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**Laboratory Diagnosis
of Diarrhoeal Diseases
1985-1991**



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PREFACE

The **Specialized Bibliography Series** is a part of the larger effort to facilitate the exchange of information and to establish an information network in the field of diarrhoeal disease and related subjects, -- an effort being carried out by the Diarrhoeal Diseases Information Services Centre (DISC) of the International Centre for Diarrhoeal Disease Research, Bangladesh (ICDDR,B). The present issue, the sixteenth of the series, includes citations of 547 papers on the use of diagnostic kits and tools in diarrhoeal diseases, published during 1985-1991. Of the 547 papers, 367 include abstracts. The **Annotated Bibliography on Laboratory Diagnosis of Diarrhoeal Diseases** was compiled from existing information available at the DISC of ICDDR,B, and it is possible that inadvertent omissions may have occurred.

We hope that the present bibliography will contribute towards generating greater interest and awareness on research in the laboratory diagnosis of diarrhoeal diseases and the results obtained from these studies. Most of the published papers, cited in this bibliography, are available from the DISC of ICDDR,B to interested persons/organisations for consultation and dissemination. We will consider this attempt successful if the bibliography serves the interests of the diarrhoeal disease researchers, laboratory technicians and other interested groups.

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The assistance of DISC staff members who directly or indirectly contributed in successfully completing this bibliography is acknowledged. MEDLINE database on CD-ROMs was of immense help in finding out information on relevant literature.

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INTRODUCTION

Gastrointestinal infections, particularly diarrhoeal diseases, constitute one of the major challenges of the medical profession. Despite a virtual revolution that has taken place in the diagnosis and treatment of diarrhoeal diseases in the past two decades or so, diarrhoeal diseases continue to exact a heavy toll on the lives of young children, particularly in developing countries.

Accurate diagnosis of causes of diarrhoeas is important to impart rational therapy to patients, to define the extent of the problem due to particular pathogens and eventually to institute control measures. In the past several years, a plethora of new diarrhoeal agents has been identified by improved techniques that has considerably narrowed the gap that has existed in our knowledge on the aetiologies of infectious diarrhoeas. In some instances, conventional culture techniques for identification of pathogens have been superseded by rapid and simple assay systems for direct identification of antigens in body fluids. The sensitivities of existing enzyme-linked immunoassay systems have been increased by introduction of newer substrates. Development of monoclonal antibodies against an array of pathogens has greatly improved the specificity of diagnosis.

Yet another achievement is the wider application of simple staphylococcal coagglutination test for the detection of a number of diarrhoeal pathogens. The development of nucleic acid probes for virulence determinants of intestinal pathogens has introduced a new dimension in large-scale screening of pathogens for epidemiological studies, although their full potential in the routine diagnosis is yet to be realised. Replacement of hazardous and limited shelf-life radioactive labels of nucleic acid probes with stable, non-radioactive labels have made these diagnostic tools even more appealing to wider use. The "jewel in the crown" of achievements so far has been the development of polymerase chain reaction technique. The exquisite sensitivity of this detection system theoretically allows the detection of even a single copy of a particular nucleic acid sequence in the specimen, opening limitless possibilities for diagnosis and treatment.

The major challenge, however, remains the diagnosis and treatment of diseases in the rural areas of developing countries where the problems are acute. Let us hope that the simplification of existing techniques and development of newer ones will continue to take place in the days ahead to tackle this challenge, when village health workers will be armed with simple diagnostic tools to do on-the-spot diagnosis.

In the following pages we have attempted to compile a list of references on the laboratory diagnosis of diarrhoeal diseases that have appeared in print since 1985. We have tried to make this list as exhaustive as possible and yet it is likely that we would have missed some important references for which we offer our apologies. It is hoped that this volume will serve as a ready-reference to laboratory personnel involved in research and diagnosis of diarrhoeal diseases, not only in developing countries, but in the developed countries as well.

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An index to co-authors is appended at the end of the main text for easy search by co-authors' names. The numbers cited in the co-author and the subject indexes refer to the sequential numbers of the citations.

ANNOTATED BIBLIOGRAPHY ON LABORATORY DIAGNOSIS OF DIARRHOEAL DISEASES, 1985 - 1991

001 Abe A, Komase K, Bangtrakulnonth A, Ratchtrachent O-A, Kawahara K, Danbara H. Trivalent heat-labile- and heat-stable-enterotoxin probe conjugated with horseradish peroxidase for detection of enterotoxigenic *Escherichia coli* by hybridization. *J Clin Microbiol* 1990 Dec;28(12):2616-20

"A 1,268-bp polynucleotide probe for heat-labile and heat-stable enterotoxins (LTh, ST1a, ST1b) was conjugated with horseradish peroxidase (HRP). The HRP-conjugated trivalent probe was applied to the detection of enterotoxigenic *Escherichia coli* (ETEC) by colony and stool hybridizations. The binding of the probe to its targets was assayed by the addition of HRP substrates hydrogen peroxide and luminol in the presence of an enhancer, and the chemiluminescence was recorded by exposure to X-ray film. Slot blot hybridization demonstrated that the HRP-conjugated trivalent probe specifically hybridized with the DNA isolated from ETEC strains. The trivalent probe also specifically identified bacterial colonies of ETEC that produced LTh, ST1a, ST1b, LTh-ST1a, or LTh-ST1b. Treatment of targets with sodium dodecyl sulfate and proteinase K remarkably reduced nonspecific hybridization to DNAs of non-ETEC strains. Furthermore, this probe was able to detect stool specimens seeded with 10^4 original ETEC cells per 5 mg of feces. These results suggest that the HRP-conjugated trivalent probe is a candidate for use in the clinical laboratory to detect ETEC." (Authors' abstract)

002 Aceti A, Pennica A, Caferro M, Celestino D, Paparo BS, Sebastiani A. Application of time-resolved fluorimmunoassay (TR-FIA) for the diagnosis of invasive amoebiasis. *Trans R Soc Trop Med Hyg* 1987;81(5):764-7

"A development of solid-phase immunoassay, time-resolved fluorimmunoassay (TR-FIA), was used for anti-amoebic antibodies. The test used a chelate of the lanthanide europium as label. The long fluorescent life-time and large Stoke's shift of europium chelates permit sensitive detection in a time-resolved fluorimeter. The TR-FIA was compared with counterimmunoelectrophoresis (CIEP), indirect haemagglutination (IHA) and enzyme-linked immunosorbent assay (ELISA) using 61 sera from patients with invasive amoebiasis, 344 sera submitted routinely for amoebic serology without clinical data, 125 sera from patients with diseases other than amoebiasis, and 86 sera from "healthy" individuals. Overall agreement between TR-FIA and ELISA was 97.1%, between TR-FIA and IHA 93.2%, and between TR-FIA and CIEP 94.0%. Sensitivity, specificity and positive and negative predictive values were calculated to compare the 4 diagnostic methods in invasive amoebiasis. TR-FIA showed a higher validity than other methods. The application of such highly detectable labels in immunometric assays promises to open up entirely new areas of research." (Authors' abstract)

003 Acheson DWK, Keusch GT, Lightowlers M, Donohue-Rolfe A. Enzyme-linked immunosorbent assay for Shiga toxin and Shiga-like toxin II using P_1 glycoprotein from hydatid cysts. *J Infect Dis* 1990 Jan;161(1):134-7

"Shiga toxin from *Shigella dysenteriae* type 1 strains and Shiga-like toxins (SLT) I and II from *Escherichia coli* bind to terminal α -D-Galp-(1 $\rightarrow$ 4)-D-Galp containing glycolipids. Hydatid cyst fluid isolated from sheep infected with *Echinococcus granulosus* contains a glycoprotein (P_1 gp) with a terminal α -D-Galp-(1 $\rightarrow$ 4)-D-Galp disaccharide. Preparations of P_1 gp were shown to interact directly with Shiga toxin and to inhibit the binding and cytotoxicity of Shiga toxin to HeLa cells. A sandwich ELISA

was developed using preparations of P₁gp as the toxin capture molecule, which, with an appropriate polyclonal antibody, was capable of detecting as little as 80 pg/well Shiga toxin and 132 pg/well SLT-II. Thus, the P₁gp-toxin interaction forms the basis for a simple antigen-capture ELISA that may be useful clinically for the rapid detection and quantitation of Shiga and Shiga-like toxins." (Authors' abstract)

004 Addiss DG, Mathews HM, Stewart JM, Wahliquist SP, Williams RM, Finton RJ, Spencer HC, Juranek DD. Evaluation of a commercially available enzyme-linked immunosorbent assay for *Giardia lamblia* antigen in stool. J Clin Microbiol 1991 Jun;29(6):1137-42

"The lack of a quick, simple, and inexpensive diagnostic test has limited the ability of public health officials to rapidly assess and control outbreaks of *Giardia lamblia* in child day-care centers. We evaluated the performance of a commercially available enzyme-linked immunosorbent assay (ELISA) for the detection of a *G. lamblia*-associated antigen in stool. Stool specimens were collected from the diapers of 426 children attending 20 day-care centers, fixed in 10% formalin and polyvinyl alcohol, and examined by microscopy by formalin concentration and trichrome staining techniques. Specimens were also tested visually and spectrophotometrically by ELISA. Of 99 tests positive by microscopy, 93 were visually positive by ELISA (sensitivity, 93.6%). Of 534 tests negative for *G. lamblia* by microscopy, 32 (6.0%) were ELISA positive. However, on the basis of examination of multiple specimens from the same child, none of these could be considered false-positive ELISAs; the specificity of the ELISA was therefore 100%. The sensitivity of both microscopy and ELISA improved as the number of specimens per child increased. An optical density value of >0.040 was 98.0% sensitive and 100% specific for *G. lamblia*. This ELISA, which appeared to be more sensitive for *G. lamblia* than did microscopic examination of stool, should be useful as an epidemiologic tool, particularly in day-care settings, and may also have a role in confirming clinical diagnoses of giardiasis." (Authors' abstract)

005 Agbalika F, Wullenweber M, Prevot J. Preliminary evaluation of the ELISA as a tool for the detection of rotaviruses in activated sewage sludge. Zentralbl Bakteriol Mikrobiol Hyg [B] 1985 May;180(5-6):534-9

"Samples of activated sludge were taken in three different treatment plants near Nancy (France) and in West Berlin (F.R.G.), and were investigated for their content of rotaviruses. After elution with beef extract (0.25%, pH 7.3) and ultrasonication, the samples were concentrated by protamine flocculation and, in a few cases, in parallel by ultracentrifugation. A commercially available ELISA test kit (Rotazyme, Abbott Diagnostics) was routinely used for rotavirus detection. Although both the Rotazyme and a laboratory-designed ELISA gave positive results for the sludge concentrates, confirmation with a blocking test and electron microscope (EM) was negative in all cases so far investigated. Therefore, the exclusive use of ELISA cannot be recommended for similar investigations as it may lead to false positive results. In this light, an additional verification step with neutralizing antibodies should be included by the ELISA kits' manufacturers to enable a quick confirmation of the user's results." (MEDLINE)

006 Ahmed MU, Taniguchi K, Kobayashi N, Urasawa T, Wakasugi F, Islam M, Shaikh H, Urasawa S. Characterization by enzyme-linked immunosorbent assay using subgroup- and serotype-specific monoclonal antibodies of human rotavirus obtained from diarrheic patients in Bangladesh. J Clin Microbiol 1989 Jul;27(7):1678-81

"By enzyme-linked immunosorbent assay with group A-, subgroup-, and serotype-specific monoclonal antibodies (MAbs), we tested 414 stool specimens collected from pediatric and adult patients hospitalized with acute gastroenteritis between January and June 1988. Of 414 specimens tested, 124 (30%) were positive for group A rotavirus. The subgroup was determined in 110 specimens (88.7%); 16.1% were subgroup I, and 72.6% were subgroup II. Two specimens reacted with both subgroup I- and subgroup

II-specific MABs. Serotype determinations showed that serotype 1 (38.4%) was predominant over serotypes 2 (28.2%), 3 (2.5%), and 4 (23%). Three specimens reacted with more than one serotype-specific MAB. While the frequency of serotype 1 was highest in the two hospitals in Mymensingh, serotype 2 was most prevalent in one hospital in Dhaka. All human rotavirus strains with subgroup I and serotype 2 specificities showed a short electropherotype, and all but one strain with subgroup II and serotype 1, 3, or 4 specificities exhibited a long electropherotype." (Authors' abstract)

007 Ahren CM, Gothefors L, Stoll BJ, Salek MA, Svennerholm A-M. Comparison of methods for detection of colonization factor antigens on enterotoxigenic *Escherichia coli*. J Clin Microbiol 1986 Mar;23(3):586-91

Faecal *Escherichia coli* isolates from 196 patients with watery diarrhoea and 68 healthy individuals (controls) were analysed in Bangladesh immediately after isolation for the presence of colonisation factor antigen (CFA) I or II (CFA/I or CFA/II respectively) by a mannose-resistant haemagglutination (MRHA) test with six species of erythrocytes and by a slide agglutination test with absorbed CFA/I or CFA/II antisera. The presence of CFAs was confirmed by immunodiffusion analyses done in Sweden. By these methods, it was found that 49 of 69 enterotoxin-producing *E. coli* strains isolated from patients carried CFA/I or CFA/II, whereas none of the non-enterotoxigenic *E. coli* isolates or the three toxin-positive strains isolated from healthy individuals carried these adhesins. All *E. coli* strains retained their MRHA ability after transportation to Sweden followed by one subculture and after storage at -70°C (but not at room temperature) for 1 to 2 years without further subculturing. After 5 to 10 subcultures of the fresh isolates, however, 70% of the initially CFA/I and 80% of the initially CFA/II-carrying strains analysed did not haemagglutinate. The efficacy of different methods for detecting CFAs on the fresh isolates was compared with that of immunodiffusion. The sensitivity of MRHA with human blood group A erythrocytes for the detection of CFA/I was high (97%), but the specificity was only 69%. The sensitivity of MRHA with bovine erythrocytes for the detection of CFA/II in Bangladesh was very low but increased considerably when chicken erythrocytes were also used. Whereas both false-positive and false-negative reactions were obtained when absorbed CFA antisera were used for agglutination, antisera against purified CFAs were equally effective as immunodiffusion in identifying CFA/I- and CFA/II-carrying strains. (Modified author's abstract)

008 Akhtar SQ. Application of Biken test (modified Elek test) for sampling of heat-stable enterotoxin of *Escherichia coli* isolated in Bangladesh. Biken J 1986;29(3-4):73-5

||
"The usefulness of Biken agar 2 as a source of heat-stable (ST) toxin for the suckling mouse assay was examined. Strains of *Escherichia coli*, isolated in Bangladesh from patients with gastroenteritis, were found to produce ST toxin (n=152), both ST and heat-labile (LT) toxin (n=60) and not to produce either of the toxins (n=25) by using the standard suckling mice assay using broth culture. Sampling from Biken agar 2 gave results comparable to those obtained using standard broth cultures. This is the first field survey of evaluation of the Biken test for sampling ST toxin of *E. coli*. The result further clarifies the applicability of the Biken test for sampling ST toxins, and the usefulness of the Biken test for detection of LT and St toxin produced by *E. coli*." (Modified author's abstract)

009 Alabi SA, Odugbemi T. Biochemical characteristics and a simple scheme for the identification of *Aeromonas* species and *Plesiomonas shigelloides*. J Trop Med Hyg 1990 Jun;93(3):166-9

"The biochemical characteristics of 55 strains of *Aeromonas* spp. and 16 *P. shigelloides* of human faecal origin were examined. Results of tests for the production of oxidase, acid from xylose, dulcitol, adonitol, mannitol, or inositol; greenish pigmentation on nutrient agar, and growth in 6.5% NaCl broth were found to be specific and reproducible. However, other tests such as haemolysis on blood agar (BA), production of DNase,

amylase, ornithine or lysine decarboxylase; citrate utilization and the methyl red, Voges-Proskauer (MRVP) varied with different strains. On the basis of our results we propose a simple scheme for the preliminary identification of *Aeromonas* and *Plesiomonas* isolates in laboratories with limited resources. We hope that other laboratories will evaluate it to determine its validity." (Authors' abstract)

010 Albert MJ. Enteric adenoviruses: brief review. Arch Virol 1986;88(1-2):1-17

In 1978, the World Health Organization initiated a programme for global prevention and control of childhood diarrhoeas. As a result, the relative importance of various pathogens in the aetiology of diarrhoea in many parts of the world has been recognised. Rotavirus ranks as the most prevalent viral pathogen in childhood diarrhoea. After a decade of intense research since its discovery, many vaccines for its control are in sight. Several studies have now established enteric adenoviruses as definite pathogens causing diarrhoea. They probably rank as the second most important viral pathogen. Structural study of enteric adenovirus strains has shown an icosahedron capsid with a diameter of 70-80 nm and fibers 28-33 nm long, which are typical of established adenoviruses. The growth characteristics, taxonomy, genome profiles, and cloning and mapping of genomes are discussed. No systematic study of the antigenic structure of enteric adenovirus has so far been undertaken. However, some information about their antigenic structure can be obtained from serological methods used to identify them, and by analogy with established adenoviruses. As a group, adenoviruses possess several structural proteins which can be resolved by SDS-polyacrylamide gel electrophoresis. The leading symptom of enteric adenovirus infection is diarrhoea. It may be associated with fever, vomiting, or respiratory problems. The disease is usually mild, but fatal cases have also been reported. Enteric adenoviruses are excreted in large quantities of up to 10¹⁰ particles per gram of stool at the acute stage of the disease, suggesting that they multiply actively in the gastrointestinal tract, most likely in the small intestinal mucosa. The epidemiology of the disease, including seroepidemiology, seasonal variation, outbreaks, and mode of spread, are described, and detection methods are outlined. Type-specific enzyme-linked immunosorbent assay has been recommended as the most suitable method for the diagnosis of enteric adenovirus-associated gastroenteritis.

011 Allard A, Girones R, Juto P, Wadell G. Polymerase chain reaction for detection of adenoviruses in stool samples. J Clin Microbiol 1990 Dec;28(12):2659-67

"The usefulness of the polymerase chain reaction (PCR) method for diagnosing adenovirus infections was investigated. Several primers, including primers specific for the hexon-coding region and for enteric adenovirus types 40 and 41, were evaluated. The PCR method was validated against cell culturing in routine diagnostic work and against restriction enzyme analysis of viral DNA. Sixty diagnostic specimens were selected for evaluation by the PCR method. Twenty of the 60 specimens were found positive on the basis of cytopathic effects and latex agglutination (Adenolex [Orion Diagnostica, Helsinki, Finland]), and 16 were identified and typed as adenoviruses by polyacrylamide gel electrophoresis. PCR was performed on all 60 specimens in parallel directly on diluted stool samples and on viral DNA extracted from cells inoculated with the same stool samples. When the general hexon primers were used 51 of the 60 specimens from infected cell cultures were found positive by PCR, whereas only 13 specimens were found positive when PCR was performed directly on stool samples. With the use of selective primers for enteric adenoviruses 16 of the 60 cell cultures were found to exhibit amplification products by PCR, whereas 4 were detected in stool samples. None of the 60 specimens were found positive by PCR when an adenovirus type 40-specific primer pair was used. PCR was found to be a fast, sensitive, and reliable method for the detection of adenoviruses in diarrheal disease, provided the amplifications were performed directly on diluted stool samples." (Authors' abstract)

012 Almeida RJ, Hickman-Brenner FW, Sowers EG, Puhf ND, Farmer JJ, 3d, Wachsmuth IK. Comparison of a latex agglutination assay and an enzyme-linked

Immunsorbent assay for detecting cholera toxin. J Clin Microbiol 1990 Jan;28(1):128-30

"To determine the pandemic potential of *Vibrio cholerae*, one must demonstrate both the presence of O1 antigen and the production of enterotoxin (CT). Tissue culture or enzyme-linked immunosorbent assays (ELISAs) for CT have been limited to research and reference laboratories. A kit for detecting CT by reversed passive latex agglutination is now commercially available and was used to test 168 strains of *V. cholerae* O1 and non-O1. When compared with the routine ELISA, the latex test was 98% accurate (86 of 88) for serogroup O1 strains and 100% accurate (80 of 80) for non-O1 strains. For both O1 and non-O1 study strains, the sensitivity of the latex agglutination test was 0.97 and the specificity was 1.00 when results were compared with ELISA results. The latex test is commercially available and has the advantages of being less complicated and less time-consuming than the ELISA." (Authors' abstract)

013 Altwegg M, Steigerwalt AG, Altwegg-Bissig R, Luthy-Hottenstein J, Brenner DJ. Biochemical identification of *Aeromonas* genospecies isolated from humans. J Clin Microbiol 1990 Feb;28(2):258-64

"One hundred phenotypic characteristics were determined for 138 clinical and environmental *Aeromonas* strains. Cluster analysis revealed three major phenons equivalent to the *A. hydrophila*, *A. caviae*, and *A. sobria* groups, each of which contained more than one genospecies and more than one named species. An excellent correlation was found between phenotypic identification and classification based on DNA relatedness. DNA hybridization groups within each of the phenotypic groups were also separable by using a few biochemical characteristics. Key tests were production of acid from or growth on D-sorbitol (which separated DNA hybridization group 3 from groups 1 and 2 within the *A. hydrophila* phenogroup), growth on citrate (which essentially separated by DNA hybridization group 4 from groups 5A and 5B within the *A. caviae* phenogroup), and growth on DL-lactate (which separated DNA hybridization group 1 from groups 2 and 3 within the *A. hydrophila* phenogroup as well as group 5A from groups 4 and 5B within the *A. caviae* phenogroup). All except one strain in the *A. sobria* phenogroup belonged to DNA hybridization group 8. DNA hybridization groups were not equally distributed among clinical and environmental isolates, suggesting that strains of certain DNA hybridization groups might be less virulent than others." (Authors' abstract)

014 Alvarado-Aleman FJ, Ruiz-Castaneda M, Kumate J. [The flocculation-agglutination test in capillaries in the serologic diagnosis of typhoid fever in children]. Bol Med Hosp Infant Mex 1987 Feb;44(2):74-80

015 Anand P, Malaviya B, Das P, Mateen MA, Habibullah CM, Das SR. Multilayer-enzyme linked immunosorbent assay (ML-ELISA) for detection of *Entamoeba histolytica* trophozoite coproantigen. Immunol Invest 1985 Oct;14(5):443-53

"Multilayer ELISA (ML-ELISA) method for *Entamoeba histolytica* trophozoite coproantigen was developed for diagnosis of amoebiasis and its usefulness vis-a-vis microscopic examination analysed for paired specimens from 87 patients from Lucknow and Hyderabad, India. All the stool specimens positive for trophozoites were also found to be positive in ML-ELISA. Out of a total 32 samples positive for cysts in microscopy 21 (65.62%) gave positive reactions for coproantigen. Results on 26 samples drawn from otherwise healthy subjects with negative microscopical picture, 4 were positive for coproantigen. The results of the present study indicate that ML-ELISA test is suitable for early diagnosis of acute amoebic patients for timely treatment." (MEDLINE)

016 Anderson JM, Hartman PA. Direct immunoassay for detection of salmonellae in foods and feeds. Appl Environ Microbiol 1985 May;49(5):1124-7

"A direct enzyme immunoassay (EIA) with polyclonal antibodies was developed for

detecting salmonellae in foods and feeds. *Salmonella* cells were attached firmly to the wells of polystyrene microtitration plates with a capture-antibody technique. Spicer-Edwards anti-H immunoglobulin G was bound to protein A-beta-D-galactosidase to serve as the signal; 4-methylumbelliferyl-beta-D-galactoside was used as the substrate. The sensitivity threshold was 10(7) cells per ml. Direct EIA, indirect EIA, and pure-culture techniques were compared by using 48 samples of naturally contaminated foods and feeds. The direct EIA was more sensitive than the indirect EIA or pure-culture technique. Food samples were analyzed within 3 working days, and 32 samples were tested simultaneously in a single 96-well microtitration plate. False-positive or false-negative results did not pose a problem. This direct EIA is sensitive, rapid, and amenable to automation." (MEDLINE)

017 Ansorg R, von Recklinghausen G, Pomarius R, Schmid EN. Evaluation of techniques for isolation, subcultivation, and preservation of *Helicobacter pylori*. J Clin Microbiol 1991 Jan;29(1):51-3

"With sheep blood agar (SBA), Belo Horizonte medium, and Brussels campylobacter charcoal agar, 104 strains of *Helicobacter pylori* were detected in 309 gastric biopsies. Each medium revealed only 69 to 71% of the strains. Ten strains grew solely on SBA, and four strains each grew on Belo Horizonte medium and Brussels campylobacter charcoal agar. Subculturing of 50 fresh *H. pylori* isolates on SBA revealed a progressive reduction in growth with increasing passage. Thirty strains stopped growing between passages two and seven. Four strains survived more than 20 passages. The preservation of fresh *H. pylori* isolates at -193 and -70°C and by lyophilization was compared by use of 10% porcine mucin solution, fetal calf serum, and a commercial cryopreservative fluid. Of 30 strains, 77 to 90% could be recultivated on SBA after preservation at -70°C in all three storage media. The data indicate that for the primary isolation of *H. pylori*, not only one selective medium but several selective media with different antibiotic supplements plus at least one nonselective medium should be used to yield the highest culture rates. Frequent subculturing of *H. pylori* on SBA selects strains which may not be representative of clinical isolates. Storage of fresh *H. pylori* isolates at -70°C in 10% mucin solution is a simple and effective preservation procedure." (Authors' abstract)

018 Anzivino D, Nani E, Lavitola A, Amato G, Gulletta E. Comparative evaluation of the new titertek enterobac rapid automated system (TTE-RAS) for identification of members of the family *Enterobacteriaceae*. J Clin Microbiol 1986 Nov;24(5):741-3

019 Appassakij H, Bunchun N, Sarasombath S, Rungpitarangsi B, Manatsathit S, Komolpit P, Sukosol T. Enzyme-linked immunosorbent assay for detection of *Salmonella typhi* protein antigen. J Clin Microbiol 1987 Feb;25(2):273-7

"A double-antibody sandwich enzyme-linked immunosorbent assay (ELISA) was designed for the detection of *Salmonella typhi* protein antigen. The optimal concentration of antibody for coating the plate was found to be 50 µg/ml. The optimum conditions for antibody coating and antigen and conjugate incubation were 37°C for 3 h, 37°C for 2 h, and 4°C overnight, respectively. The enzyme-substrate reaction was allowed to take place at 30°C for 1 h. The established ELISA was found to be reproducible, with an inter-run coefficient of variation of less than 12% for the detection of an *S. typhi* protein antigen concentration of 0.5 to 50 µg/ml. The minimal detectable level of the antigen was 0.5 µg/ml. Cross-reactions were observed with the high level (50 µg/ml) of protein antigens obtained from *Salmonella typhimurium*, *Escherichia coli*, *Salmonella paratyphi A*, and *Salmonella enteritidis*. The ELISA established was used for the detection of *S. typhi* protein antigen in serum from 62 patients with typhoid, 30 patients with clinically diagnosed typhoid fever, 21 patients with paratyphoid, 17 patients with pyrexia caused by other bacteria, and 160 normal, healthy individuals. It was found that the sensitivity, specificity, accuracy, positive predictive value, and negative predictive value of this assay were 83.87, 89.04, 87.93, 67.53, and 95.31%, respectively. (Authors' abstract)

020 Araj GF, Chugh TD. Detection of *Salmonella* spp. in clinical specimens by capture enzyme-linked immunosorbent assay. J Clin Microbiol 1987 Nov;25(11):2150-3

"A capture enzyme-linked immunosorbent assay (ELISA; Kirkegaard and Perry Laboratories, Gaithersburg, Md.) was used to detect *Salmonella* spp. in clinical and artificially inoculated specimens. In patients with bacteremia caused by *Salmonella* spp., 48% (12 of 25) and 82% (13 of 16) of serum and urine specimens, respectively, were positive for *Salmonella* spp., as determined by ELISA. All serum and urine specimens collected from healthy individuals (25 specimens) or patients whose blood cultures grew gram-negative bacteria other than *Salmonella* spp. (18 specimens) were negative for *Salmonella* spp., as determined by ELISA. For blood culture bottles in which *Salmonella* spp. (16 specimens) was grown the ELISA was positive (100%), while it was negative for all the 65 blood culture bottles in which gram-negative bacteria other than *Salmonella* spp. (42 specimens) or gram-positive bacteria (23 specimens) were grown. All samples of urine (16 specimens), stool (8 specimens), serum (16 specimens), culture media (12 specimens), and blood culture bottles (reported sterile after 2 weeks of incubation; 16 specimens) that were artificially inoculated with 10^5 to 10^7 CFU of four species of *Salmonella* per ml were positive by ELISA. Similar specimens inoculated with or containing various species other than *Salmonella* were negative by this test. Thus, ELISA offers a promising opportunity for the rapid detection of *Salmonella* spp. in clinical microbiology laboratories." (Authors' abstract)

021 Arakaki T, Hasegawa H, Asato R, Ikeshiro T, Kinjo F, Saito A, Iwanaga M. A new method to detect *Strongyloides stercoralis* from human stool. Jpn J Trop Med Hyg 1988 Mar;16(1):11-7

022 Aronsson B, Granstrom M, Mollby R, Nord CE. Enzyme immunoassay for detection of *Clostridium difficile* toxins A and B in patients with antibiotic-associated diarrhoea and colitis. Eur J Clin Microbiol 1985 Apr;4(2):102-7

A sandwich enzyme immunoassay (ELISA) was developed to detect *Clostridium difficile* toxins A and B in stools from patients with antibiotic associated diarrhoea and colitis. Immune serum to crude *Clostridium difficile* toxin and non-immune serum were coated onto polystyrene microtiter plates to act as capture antibodies; toxins A and B in human stools were detected by antibodies from rabbits immunized with purified toxins A and B. The ELISA for toxin B showed cross-reactions with *Clostridium bifermentans* and *Clostridium sordellii* and lacked diagnostic sensitivity in clinical samples. The ELISA for toxin A showed no cross-reactions with other clostridia investigated and was positive in 33% (62/189) of patients with antibiotic associated diarrhoea. This compared with 38% (71/189) positive in the tissue culture assay for *Clostridium difficile* cytotoxin. With a predictive value of 96% in clinical specimens, the ELISA for toxin A constitutes a sensitive and specific tool for diagnosis of *Clostridium difficile* associated diarrhoea and colitis." (Authors' abstract)

023 Arya SC, Parande CM. Effect of stimulated poor specimen handling on ELISA for rotavirus detection. J Commun Dis 1986 Mar;18(1):57-8

024 Asagi M, Ogawa T, Minetoma T, Sato K, Inaba Y. Detection of transmissible gastroenteritis virus in feces from pigs by reversed passive hemagglutination. Am J Vet Res 1986 Oct;47(10):2161-4

"A reversed passive haemagglutination (RPHA) method was developed for the detection of transmissible gastroenteritis (TGE) virus in faecal specimens obtained from pigs. Ovine erythrocytes fixed with glutaraldehyde and treated with tannic acid were coated with anti-TGE virus swine antibodies, which were purified by affinity chromatographic technique linked with purified TGE virus. The RPHA test was done by the microtitre method. Erythrocytes coated with purified specific antibodies were agglutinated by TGE virus, but not by porcine rotavirus or porcine enterovirus. The reaction was specifically

Inhibited by antiserum against TGE virus, confirming the specificity of the reaction. A litter of seven 3-day-old pigs was orally inoculated with TGE virus, and faecal specimens were obtained once a day and serum obtained every 4th day. With the RPHA test, TGE virus was detected in the diarrhoeal faeces; all of the inoculated pigs developed virus-neutralization antibody for the TGE virus. The RPHA test detected TGE virus in faeces collected from pigs with naturally occurring diarrhoea. The RPHA test detected TGE virus in 5 of 6 faecal specimens (80%), whereas the positivity rate was only 50% (3/6) for the immunofluorescent staining of primary cultures of porcine kidney cells inoculated with the specimens. The advantages of the RPHA method are its relative simplicity and high sensitivity. (Modified authors' abstract)

025 Ashkenazi S, Cleary TG. A method for detecting Shiga toxin and Shiga-like toxin-I in pure and mixed culture. *J Med Microbiol* 1990 Aug;32(4):255-61

"Shiga toxin and Shiga-like toxins (SLTs, syn. verotoxins) are currently detected by tissue culture assays that are expensive, time-consuming and require specialised facilities and experienced personnel. We have developed a rapid method to detect Shiga toxin and SLT-I (verotoxin 1) based on their binding to globotriosyl ceramide (Gb₃). Bound toxin was then detected by an enzyme-linked immunosorbent assay (ELISA) with monoclonal antibodies. The direct detection of cytotoxins from pure culture plates and from a mixed bacterial culture was studied. Using polymyxin extraction (0.1 g/L, 30 min, 37°C) and Gb₃-based ELISA we detected toxin from reference strains *Shigella dysenteriae* 1 strain 60R (Shiga toxin) and *Escherichia coli* O26:H11 strain H30 (SLT-I), and from clinical isolates of *E. coli* O157:H7 and O26:H11 (both SLT-I) from 11 patients with diarrhoea, haemorrhagic colitis or haemolytic uraemic syndrome. Toxin production by these strains was confirmed by a radiolabelled HeLa cell assay and the structural genes were detected by DNA hybridisation. The Gb₃-based ELISA could detect SLT-I in extracts of a mixed culture even when the toxin-positive strains represented only 1% of the mixture. No cross-reactivity was found with bacteria that produce other cytotoxins, such as other *E. coli* and *Shigella*, *Salmonella*, *Aeromonas* and *Campylobacter* spp." (Authors' abstract)

026 Avram G, Zavate O, Combiescu AA, Persu A, Ivan A, Constantiniu S, Pancu V, Popovici S, Boghean T, Nicola P, et al. [Detection of the rotavirus group antigen by a screening test using the ELISA-IC kit in subjects with acute gastroenteritis, at the pediatric services of Moldavia]. *Virologie* 1987 Jul-Sep;38(3):169-75

"The rotaviral antigen was detected by a screening test using the ELISA-IC kit in 17.6% out of 415 children with acute gastroenteritis. The highest frequency (28.9%) was found in children hospitalized in pediatric services with a diagnosis of diarrhoeic disease associated to acute respiratory infection. The rotavirus infection incidence was about three times higher during the cold season than during summer (30.4% versus 10.5%). The 6-11 month age group was the most severely affected." (MEDLINE)

027 Ayyagari A, Ray P, Kochhar R, Bhasin D, Siddeshi ER, Singh K, Malik AK, Mehta SK. Evaluation of different methods for detection of *Helicobacter pylori* in patients with gastric disease. *Indian J Med Res* [A] 1990 Mar;91:126-8

028 Ayyildiz A, Demir Y, Tuncel E, Babacan M, Leloglu S. [Comparison of the Gruber-Widal and ELISA techniques used to study *Salmonella typhi* and *Salmonella paratyphi* infections in patients]. *Mikrobiyol Bul* 1986 Jan;20(1):14-24

"In this study, antibody levels were determined against the agents of typhoid and paratyphoid fever in 168 patient's sera and 40 healthy control sera by Gruber-Widal and ELISA techniques. We compared the results of these two techniques, and discussed. The needed antigens for both techniques were prepared from the local strains of the agents of these infections which were isolated in our laboratory. The Widal's tube agglutination test was carried out by classical method, and the ELISA technique was

done by the method of Woller, *et al.* As a result, we found that the titers obtained by ELISA were as 4-6 times higher as than those of Widal's. Additionally nonspecific reactions were less seen in ELISA than in Widal." (MEDLINE)

029 Azmi F, Khan MA, Tariq S, Iqbal J, Rab A, Ghafoor A. Comparison of ELISA, direct electron microscopy and antibody capture technique for detection of rotavirus in human stools--an experience in a developing country. *J Pak Med Assoc* 1987 Jul;37(7):172-4

030 Banchuin N, Appassakij H, Sarasombath S, Manatsathit S, Rungpitarangsi B, Komolpit P, Sukosol T. Detection of *Salmonella typhi* protein antigen in serum and urine: a value for diagnosis of typhoid fever in an endemic area. *Asia-Pacific J Allergy Immunol* 1987 Dec;5(2):155-9

"Using haemoculture as the gold standard, a double antibody sandwich ELISA for the detection of *Salmonella typhi* Barber protein antigen (BP) was compared with the Widal test. Specimens used were serum and urine obtained from normal healthy individuals and from patients with typhoid fever, paratyphoid fever, pyrexia caused by other bacteria and pyrexia with negative haemoculture. The ELISA for antigenuria gave a significantly higher sensitivity, specificity, accuracy and positive predictive value than the Widal test ($p < 0.05$). The ELISA for antigenaemia gave a significantly higher sensitivity and positive predictive value only. All other values were not significantly different. The timing of specimen collection was critical for sensitivity in the ELISA for antigenaemia and antigenuria, and the best results could be obtained by carrying out both assays simultaneously. The clearance of BP from serum into urine occurred around 16 days after the onset of fever in one patient. In two patients, BP could be detected in sera up to 3 weeks after the onset of fever. In two patients, serum BP could still be detected although haemoculture was negative." (MEDLINE)

031 Baron EJ, Schenone C, Tanenbaum B. Comparison of three methods for detection of *Cryptosporidium* oocysts in a low-prevalence population. *J Clin Microbiol* 1989 Jan;27(1):223-4

032 Basta M, Karmali M, Lingwood C. Sensitive receptor-specified enzyme-linked immunosorbent assay for *Escherichia coli* verocytotoxin. *J Clin Microbiol* 1989 Jul;27(7):1617-22

033 Bate G. Comparison of minitek anaerobe II, API An-Ident, and RapID ANA systems for identification of *Clostridium difficile*. *Am J Clin Pathol* 1986 Jun;85(6):716-8

034 Baumann D, Gottstein B. A double-antibody sandwich ELISA for the detection of *Entamoeba histolytica* antigen in stool samples of humans. *Trop Med Parasitol* 1987 Jun;38(2):81-5

"A double-antibody sandwich ELISA was developed to detect detergent-solubilized antigens of *Entamoeba histolytica* in stool samples of humans. The test system was evaluated for its methodical and diagnostic sensitivity and specificity. In recovery experiments the lower limit of detection was 400 ng *E. histolytica* (HK9) protein/ml stool, corresponding to approximately 2000 amoebic trophozoites/ml stool. Samples of 97 patients with suspected intestinal amoebiasis were examined. Specific antigens were detected by ELISA (= positive reaction) in 14 (93%) out of 15 stool samples containing trophozoites of *E. histolytica*. In contrast, 68 (93%) of 73 samples with other protozoa, including *Entamoeba coli*, *E. hartmanni*, *Endolimax nana*, *Iodamoeba buetschlii* and *Giardia lamblia*, did not react in the test system (= negative reaction). The test was shown to detect only trophozoites of *E. histolytica* and not the cyst stage. This fact could facilitate the differentiation between cyst carriers and persons excreting trophozoites. The results of this preliminary study justify a further large scale evaluation of the test system." (MEDLINE)

035 Baveja UK, Roy T, Kaur M, Kulpoti DDS, Aggarwal SK. Dot immunobinding assay versus sandwich ELISA in diagnosis of invasive amoebiasis. Indian J Med Res 1991 May;93:161-5

"Sera from 34 patients of amoebic liver abscess (ALA) and 11 patients with amoebic dysentery (AD) were examined for the presence of specific *Entamoeba histolytica* (EH) antibodies and amoebic antigen by enzyme linked immunosorbent assay (ELISA) and dot immunobinding assay (DIB). Both techniques were found to be equally sensitive for detecting antibodies (89.5 and 91.9% respectively) and highly specific (100%) in patients of ALA. ELISA was found to be more sensitive (94.4%) in detecting circulating amoebic antigen compared to DIB (68%) in patients of ALA. Specific antibodies, in significant levels, were detected in 3 and 5 patients of AD by ELISA and DIB assay, respectively. As DIB assay is easier to perform and less expensive, is recommended for detection of antibodies in patients with invasive amoebiasis." (Authors' abstract)

036 Beards GM. Serotyping of rotavirus by NADP-enhanced enzyme-immunoassay. J Virol Methods 1987 Nov;18(2-3):77-85

"A method is described for the serotyping of rotaviruses directly from samples of faeces. Serotype-specific monoclonal antibodies were used in a solid phase enzyme-immunoassay utilising a novel substrate system based on the de-phosphorylation of NADP, followed by cyclic colour reaction. This method is shown to be approximately 5-10 times more specific and sensitive than assays utilising a conventional substrate for alkaline phosphatase, and allowed serotypes to be ascribed to samples which could not be typed by other methods. The value of this method for rotavirus epidemiology and vaccine trials is discussed." (MEDLINE)

037 Belaya YA, Nikolayeva LG, Bystrova SM, Belaya OF. Diagnostic significance of the coagglutination reaction in patients with the diarrhoea syndrome. J Hyg Epidemiol Microbiol Immunol 1989;33(2):181-7

"Specific soluble *Shigella*, *Salmonella* and *Yersinia enterocolitica* antigens were determined in biological fluids (saliva, urine, coprofiltrate) from 268 patients with the diarrhoea syndrome using the coagglutination reaction. The findings suggest that the coagglutination reaction (COA) is a simple and efficient method suitable for the fast diagnosing of acute intestinal infection (AII) in the early days from the onset of the disease. COA enables the identification of specific antigens associated with the causative agents of intestinal infections in 79-54% of patients with shigellosis and salmonellosis. COA was shown to possess a high diagnostic potential in AII of unknown etiology. *Shigella*, *Salmonella* and *Yersinia* antigens were determined in 47.7, 23.4 and 10.8% of cases respectively where no bacterial excretion could be confirmed. Two and less frequently three antigens being identified simultaneously in 10.8% cases. The identification of opportunistic microorganisms in AII using the COA does not appear to be sufficient to confirm their etiological significance as *Shigella* and *Salmonella* antigens were simultaneously determined in most patients." (Authors' abstract)

038 Bendall RP, Gray JJ. Haemorrhagic colitis and haemolytic-uraemic syndrome: false positive reaction with a rotavirus latex agglutination test. J Clin Pathol 1991 Jul;44(7):609-10

039 Bendig DW. Diagnosis of giardiasis in infants and children by endoscopic brush cytology. J Pediatr Gastroenterol Nutr 1989 Feb;8(2):204-6

040 Bernard S, Lantier I, Laude H, Aynaoud JM. Detection of transmissible gastroenteritis coronavirus antigens by a sandwich enzyme-linked immunosorbent assay technique. Am J Vet Res 1986 Nov;47(11):2441-4

"A new sandwich enzyme-linked immunosorbent assay, using monoclonal and polyclonal

antibodies, was developed to detect transmissible gastroenteritis virus antigens from cell culture and from intestinal wash or feces obtained from experimentally infected pigs. This technique was shown to be suitable for the detection of virulent field strain unadapted to cell culture. Cross reactions had not been observed with other enteric pathogens, rotavirus, porcine epizootic diarrhea virus, and *Escherichia coli*." (MEDLINE)

041 Bernard S, Shirai J, Lantier I, Bouteau E, Aynaud JM. Lactogenic immunity to transmissible gastroenteritis (TGE) of swine induced by the attenuated Nouzilly strain of TGE virus: passive protection of piglets and detection of serum and milk antibody classes by ELISA. *Vet Immunol Immunopathol* 1990 Jan;24(1):37-47

042 Berry PR, Rodhouse JC, Hughes S, Bartholomew BA, Gilbert RJ. Evaluation of ELISA, RPLA, and Vero cell assays for detecting *Clostridium perfringens* enterotoxin in faecal specimens. *J Clin Pathol* 1988 Apr;41(4):458-61

"Three hundred and ninety two faecal specimens from 70 separate outbreaks of suspected *Clostridium perfringens* food poisoning were examined by enzyme linked immunosorbent assay (ELISA), reversed passive latex agglutination (RPLA), and Vero cell assays for the presence of enterotoxin. Although the most time consuming method, ELISA was the most specific and reproducible. RPLA was slightly more sensitive than ELISA, but it showed some non-specific reactions. The Vero cell assay was the least sensitive and least reproducible method, being affected by some non-specific cytotoxic and cytotoxic reactions. Normal rabbit serum should be included in the Vero cell assay as a control for the neutralisation of cytotoxic effects." (Authors' abstract)

043 Berthon P, Bernard S, Salmon H, Binns RM. Kinetics of the *in vitro* antibody response to transmissible gastroenteritis (TGE) virus from pig mesenteric lymph node cells, using the ELISAPOT and ELISA tests. *J Immunol Methods* 1990 Aug 7;131(2):173-82

"A method is described for *in vitro* studies of viral humoral immune responses in the pig. After oral immunization with transmissible gastroenteritis (TGE) coronavirus, antibody production from primed mesenteric lymph node cells was revealed by an *in vitro* boost with viral antigen. For the latter the leukocytes were co-cultured with UV-inactivated virus using a variety of different methods of antigenic stimulation. Enumeration of specific antibody-secreting cells (ASC) and titration of secreted anti-virus antibodies were performed with ELISAPOT (using 3-amino 9-ethyl carbazole as the peroxidase chromogen) and ELISA tests respectively, according to the Ig isotype. The results showed a close relationship between ASC numbers and secreted antibody titres. The best *in vitro* antibody synthesis was observed when the sensitized cells were maintained in contact with virus during the whole culture period. Antibody responses were defined by a kinetic profile characterized by a narrow peak, with a maximum occurring after 4 and 6 days of culture and with the IgA response appearing earlier than the IgG. This methodology, which analyses specific antibody responses at the cellular level, may permit studies on the mechanisms of Ig isotype regulation. Extended to leukocytes from other organs of the immune system, it may also constitute an *in vitro* model to study antibody responses expressed in different lymphoid tissues of the pig." (MEDLINE).

044 Bettelheim KA, Maskill WJ. Investigation by enzyme-linked immunosorbent assay of *Salmonella* H antigen-antibody reactions. *J Clin Microbiol* 1985 May;21(5):772-4

045 Bazirtzoglou E, Romond C. Rapid identification and enumeration of *Clostridium perfringens* in the human faecal flora. *Microbial Ecol Health Dis* 1990 May-Jun;3(3):159-63

046 Bhadra RK, Bag PK, Pal SC, Nair GB. Comparison of selective medium supplemented with blood & a charcoal-based blood-free medium for primary isolation of campylobacters from human faeces. *Indian J Med Res* 1991 Jan;93:22-5

"A recently developed charcoal-based, blood-free selective medium - charcoal cefoperazone deoxycholate agar (CCDA) was compared with the conventional Butzler's agar (BA) for primary isolation of enteric campylobacters from human stool specimens. CCDA yielded higher percentage (93.6%) of positive isolations as compared to BA (76.6%). The rate of isolation on CCDA was significantly higher ($p < 0.0001$) than on BA. Cost-wise also, the CCDA medium was two times cheaper than BA. In addition to *Campylobacter jejuni* and *C. coli*, CCDA permitted the recovery of a recently described catalase-negative campylobacters which did not grow on BA. Based on these results the routine use of CCDA is recommended, particularly in developing countries, as blood is not required for this medium." (Authors' abstract)

047 Bhadra RK, Pal SC, Nair GB. Simplified method for the detection of DNA hydrolysis by enteric campylobacters. *Indian J Med Res* [A] 1991 Mar;93:87-9

"A simplified medium was developed for the detection of DNase produced by enteric campylobacters. Sensitivity and reproducibility of the test were similar to that of the improved toluidine blue DNA agar method. Logistically, the simplified DNA hydrolysis test was cheaper (5.5 times) than the earlier medium. Based on this study we recommend the routine use of the simplified medium to perform the DNase test for biotyping enteric campylobacters." (Authors' abstract)

048 Bhaduri S. Evaluation of different techniques for detection of virulence of *Yersinia enterocolitica* [letter]. *J Clin Microbiol* 1990 Apr;28(4):828-9

049 Bhawe GG, Wagle NM, Joshi UM. Detection of amoebic antigen by enzyme linked Immuno-sorbent assay (ELISA). *J Postgrad Med* 1985 Jul;31(3):146-9

050 Biddle WL, Harms JL, Greenberger NJ, Miner PB, Jr. Evaluation of antibiotic-associated diarrhea with a latex agglutination test and cell culture cytotoxicity assay for *Clostridium difficile*. *Am J Gastroenterol* 1989 Apr;84(4):379-82

051 Bielefeldt Ohmann H. Double-immunolabeling systems for phenotyping of immune cells harboring bovine viral diarrhea virus. *J Histochem Cytochem* 1987 Jun;35(6):627-33

"Optimal staining conditions were defined for simultaneous detection of bovine viral diarrhea virus (BVDV) and mononuclear leukocyte surface antigens in tissue sections and cytopins. Because of the extreme lability of the virus antigens and the variable stability of the epitopes on the cell differentiation antigens, cryopreservation had to be used. This method gives slightly sub-optimal preservation of morphology. However, the specificity and sensitivity of the immunolabeling ensured reliable identification of the double-labeled cells, i.e., the phenotypic identification of virus-infected cells within the immune system." (MEDLINE)

052 Blais BW, Yamazaki H. Extraction of *Salmonella* antigens in non-sedimentable forms for enzyme immunoassay. *Int J Food Microbiol* 1989 Aug;9(1):63-71

"Heating *Salmonella typhimurium* in ethylenediaminetetraacetate was found to dissociate its antigens into forms that are non-sedimentable at 10,000 x g. The treatment caused a marked increase in the rate of immunoreaction and the sensitivity in an enzyme immunoassay for the detection of *Salmonella* antigens. The method permits the extraction of *Salmonella* antigens from solid-rich samples and the preparation of solid-free samples by means of centrifugation. When the level of the antigens in the supernatant was too low for the immunoassay, the antigens were readily concentrated by passing a large volume of the supernatant through a macroporous hydrophobic cloth coated with anti-*Salmonella* antibody. Using this method, the detection of as few as 10 *Salmonella* cells per gram of chicken meat was possible within a total of 18 h, which included 16 h of enrichment in either tetrathionate, selenite cystine, or nutrient broths." (MEDLINE).

053 Bock RE, Burgess GW, Douglas IC. Development of an enzyme linked immunosorbent assay (ELISA) for the detection of bovine serum antibody to bovine viral diarrhoea virus. Aust Vet J 1986 Dec;63(12):406-8

"An enzyme linked immunosorbent assay (ELISA) was developed to detect antibody to bovine viral diarrhoea virus (BVDV) in bovine serum. The ELISA results were compared with those of the serum neutralisation test (SNT) using serums from 6 experimentally infected calves bled at intervals from 0 to 154 days postinfection and 886 field samples. The optical density (OD) produced by a single dilution of test serum was compared with a standard curve and the result expressed in ELISA units. Despite wide variation between absolute ELISA and SNT results, an agreement of 97% was obtained when reciprocal SNT titres ≥ 8 and ELISA units ≥ 10 were taken as indicative of a specific reaction. The ELISA was shown to be an efficient method of measuring antibody in bovine serum samples and would assist in any large scale screening of cattle herds for BVDV antibody." (MEDLINE)

054 Bongaerts GP, Bruggeman-Ogle KM, Mouton RP. Improvements in the microtitre GM1 ganglioside enzyme-linked immunosorbent assay for *Escherichia coli* heat-labile enterotoxin. J Appl Bacteriol 1985 Nov;59(5):443-9

"A variant of the microtitre GM1-ELISA for *Escherichia coli* heat-labile enterotoxin was studied. The test was improved by both reducing the assay time from 2½ d to 8 h and by determining the most appropriate GM1 coating concentration. Coating the plates with ≥ 3 µg of GM1/ml yielded a maximal sensitivity and ensured a linear relationship between the enterotoxin concentration and the extinction observed when using the final assay-procedure. Thus an optimal accuracy was obtained. This ELISA was 4- to 8-times more sensitive than the Vero cell monolayer assay. The sensitivity of this ELISA and of the chinese hamster ovary cell monolayer assay were identical." (MEDLINE)

055 Bopp CA, Threath VL, Moseley SL, Wells JG, Wachsmuth IK. A comparison of alkaline phosphatase and radiolabelled gene probes with bioassays for enterotoxigenic *Escherichia coli*. Mol Cell Probes 1990 Jun;4(3):193-203

"Alkaline phosphatase-conjugated oligonucleotide probes (APO), 32 P-labelled oligonucleotide (RO) and cloned polynucleotide (RP) probes were evaluated for their ability to detect enterotoxigenic *Escherichia coli* (ETEC) as defined by bioassay. These three sets of probes were applied to 301 *E. coli* strains that had previously been defined by the Y1 adrenal cell assay for heat-labile enterotoxin (LT) and the infant mouse assay for heat-stable enterotoxin (ST). The correlation of the APO probe for LT with the bioassay was 98% with five discrepancies and a positive predictive value (PPV) of 100%. For the APO/ST probe the correlation with the bioassay was 98% with seven discrepancies and a PPV of 96%. The correlation of the RO probe for LT was 99% with four discrepancies and a PPV of 100%, while the overall correlation for the two RO/ST probes was 97% with eight discrepancies and a PPV of 97%. For the RP probes, the correlation for LT was 99% with four discrepancies and a PPV of 100% and for ST was 98% with seven discrepancies and a PPV of 98%. These findings suggest that the APO probes were as sensitive as the RO and RP probes in detecting ETEC by colony hybridization and could be a practical alternative to bioassays and radiolabelled probes for ETEC since they do not require expensive equipment or extensive technical training." (MEDLINE)

056 Borczyk AA, Harnett N, Lombos M, Llor H. False-positive identification of *Escherichia coli* O157 by commercial latex agglutination tests [letter]. Lancet 1990 Oct 13;336(8720):946-7

057 Boulanger J, Petric M, Lingwood C, Law H, Roscoe M, Karmali M. Neutralization receptor-based immunoassay for detection of neutralizing antibodies to *Escherichia coli* verocytotoxin 1. J Clin Microbiol 1990 Dec;28(12):2830-3

058 Bowman RA, Riley TV. Laboratory diagnosis of *Clostridium difficile*-associated diarrhoea. Eur J Clin Microbiol Infect Dis 1988 Aug;7(4):476-8

059 Brandt CD, Arndt CW, Evans GL, Kim HW, Stallings EP, Rodriguez WJ, Parrott RH. Evaluation of a latex test for rotavirus detection. J Clin Microbiol 1987 Sep;25(9):1800-2

"A latex agglutination (LA) test (Slidex Rota-Kit; bioMerieux, Marcy-l'Etoile, France) was a rapid, easily used method for detecting rotavirus (RV) in pediatric fecal specimens. With 45 RV-positive and 50 RV-negative diarrhea specimens, the sensitivity of the LA test was 82%, and the specificity was 100%. Six other specimens produced indeterminate results. The frequency of positive LA tests appeared to be proportional to the concentration of virions in the stool." (Authors' abstract)

060 Brayton PR, Tamplin ML, Huq A, Colwell RR. Enumeration of *Vibrio cholerae* 01 in Bangladesh waters by fluorescent-antibody direct viable count. Appl Environ Microbiol 1987 Dec;53(12):2862-5

061 Brooks R, Brown L, Franklin R. Comparison of a protein-stabilized Rotazyme II test, with standard Rotazyme II, and electron microscopy for detection of rotavirus. Diagn Microbiol Infect Dis 1988 Dec;11(4):205-8

"We compared the performance of Rotazyme II with protein-stabilized diluent to standard Rotazyme II and to direct electron microscopy, for the detection of rotavirus. Sensitivity and specificity were: standard Rotazyme II, 79 and 100%, respectively; Rotazyme II with protein stabilizers, both 100%, and electron microscopy, 75 and 100%, respectively. Commercial availability of this product should improve rotavirus detection for laboratories utilizing Rotazyme II methodology." (Authors' abstract)

062 Brooks RG, Brown L, Franklin RB. Comparison of a new rapid test (testpack rotavirus) with standard enzyme immunoassay and electron microscopy for the detection of rotavirus in symptomatic hospitalized children. J Clin Microbiol 1989 Apr;27(4):775-7

063 Brown AL, Shields TR, Peetz RH, Mellencamp MW, Beckenhauer WH. An enzyme immunoassay for measuring *Escherichia coli* pilus antigens. Dev Biol Stand 1986;64(2):53-61

"Quantitative enzyme-linked antibody centrifuge (ELAC) assays have been developed for measuring *Escherichia coli* adhesion pilus antigens K99, K88, 987P and F41 in *E. coli* bacterins used for protecting newborn animals against neonatal enteric colibacillosis. The test consists of reacting an alkaline phosphatase conjugate of specific pilus antigen antibody directly with dilutions of the bacterin on test and a standard reference bacterin. Following incubation the unbound conjugate is removed by two centrifugation steps and the bacteria with bound conjugate are reacted with substrate. The optical density is measured at 405 nm. Unknown samples are compared to a standard bacterin known to develop sufficient immunity in sows to provide protective levels of passive maternal immunity to their piglets. A relative potency (RP) value is calculated and used for standardizing bacterial concentrates and for final potency testing. The advantages to be realized are: shortening of the test period, specificity, and reduction in test animal usage." (MEDLINE)

064 Brown DWG, Beards GM, Guang-Mu C, Flewett TH. Prevalence of antibody to group B (atypical) rotavirus in humans and animals. J Clin Microbiol 1987 Feb;25(2):316-9

"Enzyme-linked immunosorbent assays were developed for the detection of group B rotavirus antigen and antibody. The specificities of both assays were evaluated for antigens and serum specific for rotavirus groups A to D. Serum collected in the United

Kingdom from different animal species exhibited the following high prevalence of group B rotavirus-specific antibody: pigs, 97%; cattle, 71%; sheep, 91%; and goats, 91%. In human serum, a lower prevalence of group B-specific antibody was detected; serum from blood donors showed 10% prevalence, and serum from veterinarians showed 4% prevalence." (Authors' abstract)

065 Brown M. Laboratory identification of adenoviruses associated with gastroenteritis in Canada from 1983 to 1986. *J Clin Microbiol* 1990 Jul;28(7): 1525-9

066 Brown M, Shami Y, Zywicko M, Singh-Naz N, Middleton PJ. Time-resolved fluorimmunoassay for enteric adenoviruses using the europium chelator 4,7-bis(chlorosulphonyl)-1,10-phenanthroline-2,9-dicarboxylic acid. *J Clin Microbiol* 1990 Jun;28(6):1398-1402

067 Candeias JA, Fagundes-Neto U, Racz ML, Pedra MA, Ferrelra VC, Trabulsi L. Rotavirus identification in jejunal juice and stools of acute and chronic forms of infantile gastroenteritis. *Braz J Med Biol Res* 1989;22(7):833-9

"1. The present study investigates, by means of a combined enzyme immunoassay for rotaviruses and adenoviruses (EiARA), the occurrence of rotaviruses in stools and jejunal juices from 31 children with acute diarrhea and 18 with chronic diarrhea. 2. Stools from 8 acute cases contained rotaviruses (26%). In two of these cases rotaviruses were also detected in the jejunal juice. 3. In the chronic diarrhea group we identified rotaviruses in the stools of one patient and in the jejunal juice of another. 4. Some of the electrophoretotypes of the rotaviruses identified showed different patterns of RNA migration. 5. Abnormalities of the jejunal mucosa were characterized in 6 acute rotavirus-positive cases. No morphological or functional abnormalities of the intestinal mucosa were observed in the chronic diarrhea rotavirus-positive cases." (MEDLINE)

068 Carlin NIA, Lindberg AA. Monoclonal antibodies specific for *Shigella flexneri* lipopolysaccharides: clones binding to type IV, V, and VI antigens, group 3,4 antigen, and an epitope common to all *Shigella flexneri* and *Shigella dysenteriae* type 1 strains. *Infect Immun* 1987 Jun;55(6):1412-20

"Monoclonal antibodies reactive with *Shigella flexneri* O antigens were generated in both mouse and rat systems. Antibody-producing hybridomas were screened in an enzyme-linked immunosorbent assay using chemically defined lipopolysaccharides as antigens, and the epitope specificities were determined with a panel of lipopolysaccharides and synthetic O-antigen-specific glycoconjugates as antigens. To verify the specificity seen in the enzyme-linked immunosorbent assay, the antibodies were used in agglutination against a large number of *S. flexneri* strains. Monoclonal antibodies with the following specificities were identified: type, antigen IV (reactive with serotype 4a and 4b bacteria); type antigen V (reactive with serotype 5a and 5b bacteria); type antigen VI (reactive with serotype 6 bacteria); group antigen 3,4 (reactive with serotype 1a, 2a, 3b, 4a, 5a, and Y bacteria); and group antigen 1 (reactive with an epitope present on all *S. flexneri* and *Shigella dysenteriae* type 1 bacteria). Furthermore, a monoclonal antibody defining a new O-antigenic epitope present on some *S. flexneri* strains of serotypes 4a, X, and Y was characterized (4X). The monoclonal antibodies analyzed in this study define epitopes described by polyclonal antisera (type antigens IV, V, and VI), define a hitherto uncharacterized epitope (group antigen 1), and finally identify new epitopes in what has previously been considered as one epitope (group antigen 3,4 and type antigen IV). These immunochemically characterized monoclonal antibodies may have a powerful potential in studies of the importance of humoral immunity in shigellosis." (Authors' abstract)

069 Carlson JR, Sullivan PS, Harry DJ, Stork MA, Thorton SA, DuPont HL. Enzyme immunoassay for the detection of *Giardia lamblia*. *Eur J Clin Microbiol Infect Dis* 1988 Aug;7(4):538-40

070 Carnahan AM, O'Brien M, Joseph SW, Colwell RR. Enzymatic characterization of three *Aeromonas* species using API peptidase, API "Oxidase," and API esterase test kits. *Diagn Microbiol Infect Dis* 1988 Aug;10(4):195-203

"An enzymatic characterization of 16 strains of *Aeromonas* species including *A. hydrophila* (7), *A. sobria* (5), and *A. caviae* (4) was carried out using API Peptidase (strips numbered 1, 2, 3, 4, 5, and 6); API Esterase and API "Oxidase" test strips. A total of 89 substrates was used in the assay and included 59 arylamides (aminopeptides), 10 esters, and 20 carbohydrates. All three species were remarkably uniform in their reactivities. Nineteen (32%) of the arylamide substrates used were hydrolyzed by all three species. Very strong arylamidase activity was displayed by all three species for L-lysine, L-hydroxyproline, L-arginine, L-alanine, L-proline, and L-leucyl-L-alanine. Esterase activity was strongest against caproate (C6), caprylate (C8), nonanoate (C9), and caprate (C10) substrates. Only a limited number of carbohydrate substrates were hydrolyzed; strong N-acetyl- β -D-glucosaminidase activity was given by all strains. Both *A. hydrophila* and *A. caviae* gave strong β -D-glucosidase reactivities, while *A. sobria* appeared to be negative for this enzyme. The results of our preliminary study show that some of the enzymes examined may be useful in the identification and differentiation of these species. The API enzyme assays yielded rapid (4 hr) results. The assays were easy to perform, relatively inexpensive and reproducible. The importance of replicate testing and the inclusion of uninoculated (buffer only) controls as part of the assay is emphasized." (Authors' abstract)

071 Carnahan AM, Joseph SW, Janda JM. Species identification of *Aeromonas* strains based on carbon substrate oxidation profiles. *J Clin Microbiol* 1989 Sep; 27(9):2128-9

"Twenty clinical strains each of *Aeromonas hydrophila*, *Aeromonas caviae*, and *Aeromonas sobria* were evaluated for their abilities to oxidize one or more of 95 carbon sources on a GN Microplate (BIOLOG, Hayward, Calif.). Nine substrates yielded good, discriminatory values for the three species tested. The panel appears to be useful for the species identification of *Aeromonas* isolates originating from human material." (Authors' abstract)

072 Casemore DP. Laboratory methods for diagnosing cryptosporidiosis. *J Clin Pathol* 1991 Jun;44(6):445-51

073 Cerqueira-Campos M-L, Peterkin PI, Sharpe AN. Improved immunological membrane filter method for detection of food-borne *Salmonella* strains. *Appl Environ Microbiol* 1986 Jul;52(1):124-7

"An improved membrane filter method that involves the use of an enzyme-labeled antibody stain has been developed for the rapid detection of *Salmonella* species in foods. The procedure is carried out directly on a hydrophobic grid-membrane filter without requiring transfer by blotting to nitrocellulose. Pure cultures of 54 *Salmonella* species and 10 foods artificially contaminated with *Salmonella colindale* gave a positive reaction in which *Salmonella* colonies were visible as purple dots. Of 11 nonsalmonella organisms, only *Citrobacter freundii* reacted with Spicer-Edwards antiserum. Of 22 naturally contaminated food samples, 10 were positive for both the hydrophobic grid-membrane filter procedure of the Association of Official Analytical Chemists and the improved enzyme-labeled antibody stain method, and there was perfect agreement between the methods. Of these 10 positive samples, one was negative by the Health Protection Branch method; of the negative samples, two were positive by this latter method. The improved enzyme-labeled antibody stain method allows detection of *Salmonella* spp. in foods within 48 h, requires little equipment, and is inexpensive, easy to perform, and suitable for automated detection." (Authors' abstract)

074 Chakrabarti T, Chakrabarty AN, Dastidar SG. Crystal violet ring response as a marker for *in vitro* detection of pathogenic *Vibrio parahaemolyticus* & *Shigella* spp. *Indian J Med Res* 1987 May;85:508-15

Different species of bacteria were studied to see their reaction pattern with respect to crystal violet ring formation for the detection of pathogenicity *in vitro*. A total of 311 strains belonging to 11 different genera of Gram-positive and Gram-negative bacteria distributed over 34 different species were examined for their reactions to discs containing 10 and 15 µg/disc crystal violet (CV). Only 11 strains of *Vibrio parahaemolyticus* produced indistinct, irregular and thick rings around CV disc. The formation of CV rings improved with the increase in CV contents to 25 and 50 µg/disc. At contents of 75 and 100 µg CV/disc, *V. parahaemolyticus* produced a clear-cut "double ring" (DR) response. "Single rings" were given by *V. cholerae* 01/non-01, *V. alginolyticus*, *Providencia*, and *Staphylococcus aureus*. Dense rings which merged with the disc themselves and appeared to be similar to the double ring response of *V. parahaemolyticus* were produced by enterotoxigenic strains of *S. dysenteriae* 1 (4 strains) 2,4,9, *S. flexneri* 1a, *S. sonnei*, and enteropathogenic strains of *Escherichia coli*. The DR type of response to CV was found to have a correlation with enterotoxigenicity of *V. parahaemolyticus* strains. This bacterial species possessed R-plasmids which accounted for the DR response and the degree of enterotoxigenicity. This unique double ring response to crystal violet, therefore, appears to be a reliable *in vitro* marker for the pathogenic isolates of *V. parahaemolyticus*. (Modified authors' abstract)

075 Chapman PA, Rush BA, McLaughlin J. An enzyme immunoassay for detecting *Cryptosporidium* in faecal and environmental samples. *J Med Microbiol* 1990 Aug;32(4):233-7

"An enzyme immunoassay (EIA) with a monoclonal antibody to *Cryptosporidium* oocysts was developed and evaluated for the examination of faecal and environmental samples. The EIA was as sensitive as microscopy for detecting *Cryptosporidium* but also produced some positive results which could not be confirmed by a modified Ziehl-Neelsen technique or by immunofluorescence microscopy. These specimens reacted also with heterologous antibodies in EIA, suggesting false positive results." (Authors' abstract)

076 Chen YT. [Immunoperoxidase assay for the immunodiagnosis of amebiasis]. *Chung Hua Nei Ko Tsa Chih* 1985 Nov;24(11):648-50,700

077 Cherkassky BL, Minayev VI, Aleksandrova NZ, Aldova E, Hauzner O. Development of a nutrient medium to culture microorganisms of the genus *Campylobacter*. *J Hyg Epidemiol Microbiol Immunol* 1989;33(2):215-8

078 Chernesky M, Castriciano S, Mahony J, Spiewak M, Schaefer L. Ability of TESTPACK ROTAVIRUS enzyme immunoassay to diagnose rotavirus gastroenteritis. *J Clin Microbiol* 1988 Nov;26(11):2459-61

"TESTPACK ROTAVIRUS, a simple 10-min enzyme immunoassay, was compared with electron microscopy and Pathfinder enzyme immunoassay on feces from 172 patients of various ages with gastroenteritis. The percent sensitivities and specificities before blocking with antiserum were as follows: TESTPACK, 100% sensitivity and 99% specificity; Pathfinder, 95% sensitivity and 98% specificity. After blocking, the sensitivity and specificity, respectively, were 100% and 100% for TESTPACK and 95% and 99% for Pathfinder. TESTPACK ROTAVIRUS was more sensitive, but not significantly, than Pathfinder ($p > 0.1$) and the direct electron microscopy technique ($p > 0.1$). (Authors' abstract)

079 Chernesky M, Castriciano S, Mahony J, DeLong D. Examination of the Rotazyme II enzyme immunoassay for the diagnosis of rotavirus gastroenteritis. *J Clin Microbiol* 1985 Sep;22(3):462-4

"Rotazyme II, which is a shorter version of Rotazyme (less than 3 h), was compared with electron microscopy and Rotazyme for sensitivity and specificity on 229 human stool specimens. Compared with electron microscopy, the newer assay was found to be 99.4%

sensitive and 97.3% specific for an overall agreement of 98.7%. After resolution of discordant results by blocking tests, the Rotazyme II enzyme immunoassay was shown to be more sensitive than electron microscopy and, therefore, 100% specific." (Authors' abstract)

080 Chityothin O, Sethabutr O, Echeverria P, Taylor DN, Vongsthongsri U, Tharavanij S. Detection of heat-stable enterotoxigenic *Escherichia coli* by hybridization with an RNA transcript probe. J Clin Microbiol 1987 Aug;25(8):1572-3

081 Choudhry MA, Yadava JNS, Negi BS, Yadava MP. Development of vero cell miniculture assay for detection of heat-labile enterotoxin of *Escherichia coli* and its significance. J Trop Med Hyg 1989 Dec;92(6):379-82

"In the present study the vero cell miniculture assay was developed for the detection of heat-labile enterotoxin of *E. coli*. The test was found to be reliable and efficient and was comparable with other tissue culture assays. The new cell lines BHK₂₁, MDBK, LM and CK included in the present study did not respond positively to the *E. coli* toxins." (Authors' abstract)

082 Choudhury MA, Gupta S, Yadava JNS. Guinea-pig ileal loop assay: a better replacement of the suckling mouse assay for detection of heat-stable enterotoxins of *Escherichia coli*. J Trop Med Hyg 1991 Aug;94(4):234-40

"The suitability of guinea-pig ileal loop assay (GILA) for the assay of heat-stable (ST) enterotoxin was confirmed. Secretory response against *Escherichia coli* heat-stable enterotoxin in this model was determined in terms of dilatation index (DI). DI equal to 0.50 or more was considered as a positive secretory response. Kinetics of fluid accumulation and the titration of toxin in guinea-pig ileal loop suggest uniform secretory response throughout the small intestine and 31.5 µg of crude ST toxin as the minimum effective dose to induce a DI of 0.5. Guinea-pig intestine was found sensitive to both methanol soluble (STa) and methanol insoluble (STb) toxins of *E. coli* and so was considered superior to the existing suckling mouse assay (SMA), which is known to be sensitive only to STa toxin. In addition, GILA was also found to be more suitable and economical as at least 10 strains together with the positive and negative controls can be tested in one animal, whereas in SMA, four suckling mice were needed to test a single strain. Hence, in SMA individual susceptibility among mice cannot be ruled out. GILA was considered to be an alternative to the presently available test, SMA, in the determination of ST toxin of *E. coli*." (Authors' abstract)

083 Chow C-B, Wang P-S, Cheung M-W, Yan W-W, Leung N-K. Diagnostic value of the Widal test in childhood typhoid fever. Pediatr Infect Dis J 1987 Oct;6(10):914-7

"The usefulness of the Widal test in diagnosing childhood typhoid fever in endemic areas was investigated. The test was done on 150 children with other febrile illnesses and 98 bacteriologically proved cases of typhoid fever. Of the 150 children with nontyphoid fever, only one had an H agglutinin titer of 1:50. Using an H or O agglutinin titer of 1:50 or more as a criterion for diagnosis, a positive Widal test was found in 88% of typhoid fever cases on the first occasion on which the test was done. If the test was repeated at least 94% of the typhoid cases had a significant result. The Widal test is a useful diagnostic test in children in endemic areas, provided interpretation of the test is made against background information relating to agglutinin levels in normal children in the region." (Authors' abstract)

084 Christy C, Vosefski D, Madore HP. Comparison of three enzyme immunoassays to tissue culture for the diagnosis of rotavirus gastroenteritis in infants and young children. J Clin Microbiol 1990 Jun;28(6):1428-30

"Three enzyme immunoassays (EIAs), Rotazyme II, IDL, and Pathfinder, were evaluated for rotavirus detection in stool and rectal swab specimens from children with symptomatic gastroenteritis and compared with virus isolation in primary African green monkey kidney cells. Of 125 specimens tested, 49 were rotavirus positive by tissue culture isolation; of these, 49, 40 were positive by Rotazyme II, 43 were positive by IDL, and 46 were positive by Pathfinder EIAs. As compared with tissue culture isolation, the Rotazyme II, IDL, and Pathfinder EIAs had sensitivities of 82, 88, and 94%, specificities of 90, 99, and 95%, and overall agreements of 86, 94, and 94%, respectively." (Authors' abstract).

085 Chu HJ, Zee YC, Ardans AA, Dal K. Enzyme-linked immunosorbent assay for the detection of antibodies to bovine viral diarrhea virus in bovine sera. *Vet Microbiol* 1985 Jun;10(4):325-33

"A specific and sensitive enzyme-linked immunosorbent assay (ELISA) was established for the detection of antibodies to bovine viral diarrhea virus (BVDV) in bovine sera. Polyethylene-glycol concentrated, equilibrium density gradient purified BVDV was used as test antigen at an optimal amount of 1 microgram/well, whereas the optimal concentration of conjugate was at 1/2000 dilution. The standardized test encountered no non-specific reaction with test sera at a starting dilution of 1/10. A total of 50 bovine serum samples was assayed for the presence of antibodies against BVDV by ELISA and serum neutralization test (SNT). A positive correlation between the 2 tests was found. However, ELISA could be as much as 500-fold more sensitive than SNT in detecting low levels of BVDV antibodies." (MEDLINE)

086 Chu HJ, Sawyer MM, Anderson CA, Higgins RJ, Zee YC. Enzyme-linked immunosorbent assay for the detection of antibodies to bovine virus diarrhea virus in sera from border disease virus-infected sheep. *Can J Vet Res* 1987 Apr;51(2):281-3

"An enzyme-linked immunosorbent assay (ELISA) was established for the rapid detection of specific antibodies against the causative agent of border disease in ovine sera. Polyethylene-glycol concentrated, equilibrium density gradient purified bovine virus diarrhea virus was used as test antigen. The optimal amount of antigen was 0.5 microgram/well, and the optimal concentration of conjugate was at 1/4,000 dilution. A total of 20 ovine serum samples, which had been collected from animals with or without border disease, were compared by ELISA and serum neutralization test for the detection of border disease-specific antibodies. ELISA was shown to be equally specific but less time-consuming and easier to perform than serum neutralization test. A positive correlation ($r = 0.60$) between the two tests was found." (MEDLINE)

087 Chudzio T, Kasatlya S, Irvine N, Sankar-Mistry P. Rapid screening test for the diagnosis of rotavirus infection. *J Clin Microbiol* 1989 Oct;27(10):2394-6

"The early diagnosis of human rotavirus infection is essential for effective patient management and infection control. We report here a rapid, easy-to-perform, and inexpensive test for rotavirus detection. The viral RNA is extracted directly from the stools and electrophoresed on 1% agarose gels. Currently available immunoassays for routine diagnostic purposes are directed at the common group A-specific antigen. As reports become available on human gastroenteritis caused by the atypical or novel rotaviruses, this technique presents an added advantage in that it can detect both group A and non-group A rotaviruses." (Authors' abstract)

088 Clabots CR, Gerding SJ, Olson MM, Peterson LR, Gerding DN. Detection of asymptomatic *Clostridium difficile* carriage by an alcohol shock procedure. *J Clin Microbiol* 1989 Oct;27(10):2386-7

089 Combleescu AA, Persu A, Mazilu M, Moldovan L, Ficlu M, Dumitrescu A, Stefanescu M, Szegli G, Butur D. [ELISA-IC for the detection of the rotavirus group antigen, in the stools of children with acute gastroenteritis]. *Rev Ig [Bacteriol]* 1985 Oct-Dec;30(4):317-26

090 Conanen EC, Dayrit MM, Flores BB, Roa RR, Obenza J, Faraon AO, White ME. Typhoid fever pseudoe epidemic in Oroquieta city: lessons in the inappropriate use of the Widal test. *J Philip Med Assoc* 1989 Oct-Dec;65(2):205-9

091 Corazza GR, Menozzi MG, Strocchi A, Rasciti L, Valra D, Lecchini R, Avanzini P, Chezzi C, Gasbarrini G. The diagnosis of small bowel bacterial overgrowth: reliability of jejunal culture and inadequacy of breath hydrogen testing. *Gastroenterology* 1990 Feb;98(2):302-9

"The reliability of a single jejunal culture in the diagnosis of small bowel bacterial overgrowth has recently been questioned. Seventy-seven patients thought to have bacterial overgrowth, defined as a jejunal culture yielding at least 10^6 organisms per milliliter of aspirate, took part in the study. Bacterial overgrowth was found in 74% of the patients with predisposing conditions and in 32% of those with no clear causes of bacterial colonization. The intestinal juice of some patients was taken at two different levels of the proximal jejunum, using both the closed- and open-tube systems. Highly significant correlations ($r_s = 0.90$, $p < 0.001$) were found between the numbers of bacteria per milliliter at the 2 jejunal levels and between the numbers of bacteria per milliliter of jejunal aspirate obtained from the closed and open tubes ($r_s = 0.84$, $p < 0.001$). Compared with the jejunal culture, the gas chromatography of volatile fatty acids in jejunal aspirate and the glucose- and lactulose-hydrogen breath tests showed sensitivities of 56%, 62%, and 68% and specificities of 100%, 83%, and 44%, respectively. This work demonstrates the reliability of jejunal cultures and the inadequacy of breath hydrogen testing in the prediction of positive jejunal cultures. When results of testing for volatile fatty acids in jejunal aspirates are positive, this always indicates the presence of bacterial overgrowth; thus, this procedure would avoid the more complicated, time-consuming, and costly bacteriological analysis of jejunal samples." (Authors' abstract)

092 Coulson BS, Grimwood K, Bishop RF, Barnes GL. Evaluation of end-point titration, single dilution and capture enzyme immunoassays for measurement of antirotaviral IgA and IgM in infantile secretions and serum. *J Virol Methods* 1989 Oct;26(1):53-65

"In order to facilitate measurement of antirotaviral IgA in large collections of faeces and secretions, adaptations of enzyme immunoassay methods for estimating antirotaviral IgA and IgM in duodenal fluid, saliva, faeces and serum were studied. To quantitate specific IgA, a single dilution of each sample was assayed. Results were expressed as antirotaviral IgA units derived from a standard curve. Units were calculated by log-logit analysis on computer. There was strong correlation between antirotaviral IgA units and end-point titres in 257 faecal samples (correlation coefficient $r = 0.92$) and in 182 duodenal fluids and salivary samples (correlation coefficient $r = 0.74$). The assay was validated using acute and convalescent faeces from children with or without rotavirus infection. Immune conversions in IgA were detected in 33 (75%) of the children by units and 34 (77%) by titres. None of nine children with gastroenteritis due to other infectious agents showed immune conversions to rotavirus. A monoclonal capture IgM assay showed similar end-point titres and numbers of immune conversions when compared with a direct assay for antirotaviral IgM in serum and secretions. Use of the capture method eliminated false-positive reactions with the cell control. The assay for antirotaviral IgA units in secretions is simple, rapid, reproducible and reliable, and has proven of value in longitudinal epidemiological studies of rotavirus coproIgA profiles. Both the capture IgM technique and the single dilution IgA method permit analysis of large numbers of specimens and are appropriate for examination of immune responses to natural rotavirus infection or during vaccine trials." (MEDLINE)

093 Coulson BS, Unicom LE, Pitson GA, Bishop RF. Simple and specific enzyme immunoassay using monoclonal antibodies for serotyping human rotaviruses. *J Clin Microbiol* 1987 Mar;25(3):509-15

"An enzyme immunoassay for serotyping human rotaviruses in stools and in cell culture was developed. Hyperimmune rabbit antisera to rotaviruses were used as capture antibodies, and rotavirus-neutralizing mouse monoclonal antibodies specific for serotypes 1, 2, 3, and 4 were used as detection reagents. Partial purification of monoclonal antibodies and inclusion of skim milk powder in antibody diluents contributed to assay specificity. The sensitivity of this assay was greater than that of a direct enzyme immunoassay in which rotaviruses of the appropriate serotype were adsorbed directly to the solid phase. When fecal extracts were concentrated threefold, this serotyping enzyme immunoassay was of equal specificity and approached the sensitivity of electron microscopy for rotavirus detection. This assay is simple and rapid and is suitable for serotyping the large numbers of isolates obtained from epidemiological studies and vaccine trials." (Authors' abstract)

094 Cranage MP, Campbell AD, Venters JL, Mawson S, Coombs RR, Flewett TH. Detection and quantitation of rotavirus using monoclonal antibody coupled red blood cells: comparison with ELISA. *J Virol Methods* 1985 Aug;11(4):273-87

"A total of 125 faecal extracts from infants were tested by reverse passive haemagglutination (RPH) using red cells coated with a monoclonal antibody against the major group-specific rotavirus antigen (VP 6). Results were compared with those obtained using a rabbit anti-rotavirus capture, guinea pig anti-rotavirus detector-based ELISA. The specificity of the assay was confirmed by use of 'normal' immunoglobulin coupled red cells and by inhibition with rabbit antiserum. The antibody-coated red cells could be stabilised by treatment with glutaraldehyde and subsequent freeze-drying with no detectable loss of activity even after storage at 45 degrees C for 4 wk. Good correlation was obtained between RPH and ELISA. Purified bovine rotavirus could be detected by RPH down to approximately 10(5) particles in a 25 microliters vol. Similar results were obtained with polyclonal antibody coupled cells and an ELISA using monoclonal antibody. Experiments using subgroup-specific monoclonal antibodies indicated the feasibility of rapid subgroup determination." (MEDLINE)

095 Cromlen JL, Himmelreich CA, Glass RI, Storch GA. Evaluation of new commercial enzyme immunoassay for rotavirus detection. *J Clin Microbiol* 1987 Dec;25(12):2359-62

"We evaluated a new commercial enzyme immunoassay (EIA) for rotavirus (Rotavirus EIA; International Diagnostic Laboratories, Chesterfield, Mo.). A total of 161 consecutive stool samples (including 18 from infants less than 30 days old) submitted to the diagnostic laboratory at Children's Hospital, Washington University Medical Center, St. Louis, Mo., for rotavirus detection were tested by Rotavirus EIA and by Rotazyme II (Abbott Laboratories, North Chicago, Ill.) according to the instructions of the manufacturer. In addition, 16 samples from infants less than 30 days old without diarrhea were tested by both assays. Samples showing discrepant results after repeat testing were examined by electron microscopy. Nine samples yielding discrepant results were also tested by using a reference EIA directly on the specimen and on culture supernatants from two passages in MA 104 cells. Rotavirus EIA and Rotazyme II yielded concordant results for 85% of the samples. All of the 26 discrepant samples tested negative by Rotavirus EIA and positive (15 samples) or equivocal (11 samples) by Rotazyme II. These samples included 11 from symptomatic infants more than 30 days old, 2 from symptomatic infants less than 30 days old (neonates), and 2 from neonates without diarrhea. Rotavirus was not detected in any of the 24 that were examined by electron microscopy or in any of the 9 that were tested by the reference EIA. The sensitivity, specificity, positive predictive value, and negative predictive value were 100% for Rotazyme EIA and 100, 90, 70, and 100%, respectively, for Rotazyme II. Rotavirus EIA was comparable to Rotazyme II in ease of performance. We conclude that Rotavirus EIA is equally sensitive and more specific than Rotazyme II for detecting rotavirus. Rotavirus EIA is a practical and accurate rotavirus assay for use in clinical laboratories." (Authors' abstract)

096 Cryan B. Comparison of three assay systems for detection of enterotoxigenic *Escherichia coli* heat-stable enterotoxin. J Clin Microbiol 1990 Apr;28(4):792-4

"In this study, a commercial DNA-DNA hybridization kit for the detection of *Escherichia coli* heat-stable enterotoxin is compared with a competitive enzyme-linked immunosorbent assay (ELISA) and the suckling mouse bioassay. Taking the suckling mouse assay as the "gold standard," the gene probe was the more specific and the ELISA was the more sensitive of the assays. The ELISA and the suckling mouse test are semiquantitative. The ELISA was the most rapid method, most amenable to automation, and most suitable for the examination of large numbers of specimens. The gene probe is particularly applicable in relatively primitive laboratory conditions. The suckling mouse assay was the least suitable system for the examination of large numbers of specimens." (Authors' abstract)

097 Cryan B. Comparison of the synthetic oligonucleotide gene probe and infant mouse bioassay for detection of enterotoxigenic *Escherichia coli*. Eur J Clin Microbiol Infect Dis 1990 Mar;9(3):229-32

"A commercial DNA/DNA hybridisation kit for the detection of *Escherichia coli* heat stable enterotoxin gene sequences was compared to the suckling mouse bioassay using 183 isolates of *Escherichia coli* from clinical specimens. The gene probe assay had a specificity of 99% and a sensitivity of 90.4% compared to the infant mouse method. Using the colony blot method of preparing the bacterial DNA and a hybridisation temperature of 50°C optimal results were obtained. The gene probe method is not affected by the incubation conditions of the test organisms. It is technically straightforward and can be applied to large numbers of specimens with fewer logistic difficulties than with the bioassay." (Author's abstract)

098 Curlale MS, Klatt MJ, Robison BJ, Beck LT. Comparison of colorimetric monoclonal enzyme immunoassay screening methods for detection of *Salmonella* in foods. J Assoc Off Anal Chem 1990 Jan-Feb;73(1):43-50

"A colorimetric enzyme immunoassay (EIA) method for detection of *Salmonella* in foods has been compared to the AOAC colorimetric monoclonal EIA screening method (986.35, 15th ed.; 46.B21-46.B29, 14th ed.). The assays use the same monoclonal antibodies and have similar reactivity toward *Salmonella*. However, the new assay uses antibody-coated microtiter wells instead of coated magnetic beads to capture *Salmonella* antigens. Compared with the bead assay, the coated-well assay format requires significantly less time to complete, and was consistently able to detect lower levels of *Salmonella* in mixed culture. Compared to the standard AOAC culture method for food samples, the plate assay was as productive. No false negatives were obtained by the immunoassay; the false negative rate was 1.1% by the culture method. The rate of agreement between the 2 methods was 99.1%. The official final action bead assay method for *Salmonella* in foods, 986.35, and the same assay for use with low-moisture foods, 987.11, have been modified official first action to use antibody-coated microtiter strip-wells." (MEDLINE)

099 Dagan R, Hall CB, Powell KR, Menegus MA. Epidemiology and laboratory diagnosis of infection with viral and bacterial pathogens in infants hospitalized for suspected sepsis. J Pediatr 1989 Sep;115(3):351-6

"A prospective study was conducted to determine the frequency and distribution of bacterial and viral pathogens in infants hospitalized with suspected sepsis and to evaluate the potential of virus detection for improving patient management. A causative organism was detected in 157 (67%) of 233 previously healthy infants less than 3 months of age, who had been hospitalized for suspected sepsis: 19(8%) had bacterial infections, 135(58%) had viral infections, and 3(1%) had mixed viral-bacterial infections. Viral infections occurred in a seasonal pattern: enteroviruses were responsible for most of the hospitalizations during summer and fall (65/110; 63%) and respiratory syncytial and

influenza A viruses were responsible for most of the infections during winter (44/81;55%). In contrast, bacterial infections were not seasonally distributed. Virus was detected in 33% of the 138 infected infants within 24 hours, and in 64% within 3 days. We conclude that viral infections are prevalent among infants hospitalized for suspected sepsis, and most can be detected early enough to influence patient management." (Authors' abstract)

100 Davidson I, Aronovici A, Weisman Y, Malkinson M. Enzyme immunoassay studies on the serological response of turkeys to hemorrhagic enteritis virus. *Avian Dis* 1985 Jan-Mar;29(1):43-52

101 de la Vega SL, Osornio-Vargas AR, Ruiz-Palacios GM. [Coagglutination test for detecting human rotaviruses in feces: comparison with the ELISA test] *Bol Med Hosp Infant Mex* 1987 Jul;44(7):373-9

102 Delem A, Vesikari T. Detection of serum antibody responses to RIT 4237 rotavirus vaccine by ELISA and neutralization assays. *J Med Virol* 1987 Mar;21(3):231-8

"Immunoglobulin class-specific determination by ELISA and a virus neutralization test (NT) were compared for the detection of the serum antibody responses after vaccination of infants with the RIT 4237 rotavirus vaccine (of bovine origin). A capture method was applied for specific IgM determination by ELISA because of its greater sensitivity and reproducibility over the conventional ELISA-IgM test. NT was improved through the use of an enzyme-labelled antibody for the detection of non-neutralized virus. In 6-12-month-old children, the most sensitive single test to detect an antibody response was the ELISA-IgM capture method, but the combination of ELISA-IgM and IgG tests or ELISA-IgM and NT detected the highest seroconversion rate of 79%. In newborn infants high levels of maternal antibody made the ELISA-IgG test unsuitable. ELISA-IgM gave a response rate of 31%, but NT with homologous virus was the most sensitive indicator of a serological response to the vaccine in this age group, yielding a 45% response rate." (MEDLINE)

103 De Mol P, Hemelhof W, Retore P, Takeda T, Miwatani T, Takeda Y, Butzler JP. A competitive immunosorbent assay for the detection of heat-stable enterotoxin of *Escherichia coli*. *J Med Microbiol* 1985 Aug;20(1):69-74

"A competitive ELISA procedure for the detection of *Escherichia coli* heat stable enterotoxin (ST) with monoclonal antibody has been developed. This test is 10 times more sensitive than the suckling-mouse bioassay and it is specific, simple and cheap. A set of 882 strains of *E. coli* isolated from man were tested both by ST-ELISA and suckling-mouse bioassay, the latter serving as the reference method. Positive results in both tests were obtained with 152 strains. The remaining strains gave negative results in both tests, with the exception of two strains, known to be ST producers, that gave negative results in the suckling-mouse assay, but gave positive results by the ELISA method." (Authors' abstract)

104 Dennehy PH, Gauntlett DR, Spangenberg SE. Choice of reference assay for the detection of rotavirus in fecal specimens: electron microscopy versus enzyme immunoassay. *J Clin Microbiol* 1990 Jun;28(6):1280-3

"Two previously demonstrated sensitive and specific enzyme immunoassays (EIAs) for rotavirus, one using polyclonal and monoclonal antisera (TestPack Rotavirus [TPK]; Abbott Laboratories) and the other using only monoclonal anti-rotavirus antibodies (Rotaclear [RTC]; Cambridge BioScience Corporation), were evaluated as potential reference assays for rotavirus testing in comparison with direct negative-staining electron microscopy (EM), the current laboratory standard. Two hundred and seven stool samples collected consecutively during the winter of 1989 from children with acute diarrhea admitted to a ward for infants from 0 to 2 years of age were tested by the EIAs and by EM. TPK

specimens were read visually; RTC results were read spectrophotometrically. Specimens with discordant EIA and EM results were further evaluated by a fluorescent focus assay. Specimens positive by EM and those negative by EM but positive by fluorescent focus assay were considered to be positive for rotavirus. Of the 207 stools tested, 35 (17%) were positive for rotavirus by these criteria. EM had a sensitivity of only 80%. Specificities were 100% for RTC and EM and 89% for TPK. These findings indicate that EM, although very specific, is relatively insensitive compared with a highly sensitive monoclonal antibody-based EIA. An EIA with high sensitivity and specificity, such as RTC, is a more appropriate reference standard for rotavirus testing." (Authors' abstract)

105 Dennehy PH, Gauntlett DR, Tente WE. Comparison of nine commercial immunoassays for the detection of rotavirus in fecal specimens. *J Clin Microbiol* 1988 Sep;26(9):1630-4

"One hundred fecal specimens obtained from patients with acute gastroenteritis were tested for rotavirus with nine commercial immunoassays to evaluate the sensitivity, specificity, predictive value, and diagnostic accuracy of these assays. Kits evaluated included two monoclonal antibody-based enzyme immunoassays (EIAs) (Rotaclone and Pathfinder Rotavirus), three polyclonal antibody-based EIAs (Rotavirus Immunoassay, Rotazyme II, and Wellcozyme Rotavirus), and four latex agglutination assays (Rotastat, Virogen Rotatest, Meritec-Rotavirus, and The Wellcome Latex Test). Thirty-eight of the 100 specimens were found to contain rotavirus by a reference microplate EIA. The accuracy of the reference assay was determined by RNA electrophoresis and a blocking assay on discordant specimens. The two monoclonal antibody EIAs had superior sensitivities (100%) and identified two positive specimens which were negative by the reference method but positive by the blocking assay. Among the polyclonal EIAs, all had sensitivities of greater than 90%, but specificities were variable; Rotazyme II, with a specificity of 50%, showed considerable discrepancy from other polyclonal EIAs. The latex tests had sensitivities ranging from 70 to 90% and specificities of 80 to 100%. Latex agglutination tests were more rapid than EIAs and did not require expensive equipment. The final choice of assay system will depend on the cost, speed, and accuracy requirements of the clinical laboratory." (Authors' abstract)

106 Dennehy PH, Gauntlett DR. Evaluation of a new enzyme immunoassay (TESTPACK Rotavirus) for the detection of rotavirus in fecal specimens. *Diagn Microbiol Infect Dis* 1988 Dec;11(4):201-3

"One hundred and forty fecal specimens were tested for rotavirus using two immunoassays, TESTPACK Rotavirus and Pathfinder Rotavirus. Five discordant specimens were evaluated by a blocking assay. The sensitivity, specificity, positive predictive value, and negative predictive value were 98, 100, 100, and 99% for TESTPACK and 92, 100, 100, and 96% for Pathfinder, respectively." (Authors' abstract)

107 DePaola A, Hopkins LH, McPhearson RM. Evaluation of four methods for enumeration of *Vibrio parahaemolyticus*. *Appl Environ Microbiol* 1988 Feb;54(2):617-8

108 De Silva DGH, Willis P, George RH, Candy DCA, McNelsh AS. Effect of transport and storage on jejunal fluid microflora. *J Diarrhoeal Dis Res* 1985 Dec;3(4):205-8

"The effect of transport medium and the effect of freezing and thawing on colony counts in jejunal fluid were studied. Findings show that for studies of microflora it may not be necessary to transport specimens of jejunal fluid in transport medium (brain heart infusion broth with 10% glycerol) if the samples are processed rapidly. The use of transport medium may even cause loss of some organisms due to dilutions. Storage of jejunal fluid at -20°C either in brain heart infusion broth with 10% glycerol or in an undiluted form, resulted in marked decreases in viable counts of microflora, although storage of pure cultures of enterobacteria preserved viability." (Authors' abstract)

109 Desmonts C, Minet J, Colwell R, Cormier M. Fluorescent-antibody method useful for detecting viable but nonculturable *Salmonella* spp. in chlorinated wastewater. *Appl Environ Microbiol* 1990 May;56(5):1448-52

110 de Suarez EP, Perez-Schael I, Perozo-Ruggeri G, de Davila D, Romer H, Tapia FJ. Immunocytochemical detection of *Entamoeba histolytica*. *Trans R Soc Trop Med Hyg* 1987;81(4):624-6

"Human anti-*Entamoeba histolytica* immunoglobulin was used to detect *Entamoeba histolytica* in 74 positive samples from several different sources, using an indirect immunoperoxidase method. In 73 samples, the protozoan was easily identified. Trophozoites and cysts of all cultured *Entamoeba* strains examined were strongly stained, and as few as 3 trophozoites per microscope slide could be detected. In addition, 51 negative control samples were also tested and non-specific reactions were not observed. These preliminary results show that this method is both sensitive and specific, and can easily detect trophozoites and cysts of different *E. histolytica* strains." (Authors' abstract)

111 de Vries WP, Houben AW, Stobberingh EE. Comparison of four commercial assays for detection of rotavirus in childhood gastroenteritis. *Eur J Clin Microbiol* 1986 Oct;5(5):542-4

112 Dezfoulian M, Bartlett JG. Identification of toxigenic *Clostridium botulinum* type E by enzyme immunoassay. *Diagn Microbiol Infect Dis* 1991 Jan/Feb;14(1):11-5

"A sensitive double-sandwich enzyme immunoassay for identification of toxigenic *Clostridium botulinum* type E was developed with a type-E-specific antitoxin. The antitoxin was produced in a rabbit, following inoculation of minute quantities of type-E toxoid. The toxoid was prepared from a commercially available toxin by using a detoxification method. Cross-reactivity of the antitoxin with *C. botulinum* types A and B was avoided by absorption of the serum with alum-precipitated toxoids A and B. The immunoassay proved to be specific for *C. botulinum* type E because it did not react with culture filtrates of various *Clostridium* species, including nontoxigenic "E-like" organisms and *Clostridium sporogenes*." (Authors' abstract)

113 Dickle N, Speirs JI, Akhtar M, Johnson WM, Szabo RA. Purification of an *Escherichia coli* serogroup O157:H7 verotoxin and its detection in North American hemorrhagic colitis isolates. *J Clin Microbiol* 1989 Sep;27(9):1973-8

"Verotoxin (VT), which is immunologically unrelated to VT1 (Shiga-like toxin I), was purified from the culture filtrate of *Escherichia coli* hemorrhagic colitis serogroup O157:H7 strain 3657 by copper ion chelate affinity chromatography followed by anion-exchange chromatography. The isoelectric point by sucrose density gradient isoelectric focusing was 5.0, the molecular weight by gel filtration on superose 12 was about 60,000, and the 50% cytopathic dose for Vero cells was about 1 pg. This toxin was found by immunological methods to be the predominant VT in *E. coli* O157 isolates associated with illness in North America, with 38 of 42 strains tested producing this toxin, 20 in combination with VT1. VT from strain 3657 is immunologically identical to the described Shiga-like toxin II (VT2) of *E. coli* strains (from the United States) K-12(pEB1) and C600(933W) but only partially related to VT of strain E32511 (from the United Kingdom), the first to be named VT2." (Authors' abstract)

114 Doern GV, Herrmann JE, Henderson P, Stobbs-Walro D, Perron DM, Blacklow NR. Detection of rotavirus with a new polyclonal antibody enzyme immunoassay (Rotazyme II) and a commercial latex agglutination test (Rotalex): comparison with a monoclonal antibody enzyme immunoassay. *J Clin Microbiol* 1986 Feb;23(2):226-9

"A total of 176 human fecal specimens were examined for the presence of rotavirus by four different assays: a monoclonal antibody enzyme immunoassay; the original polyclonal

antibody enzyme immunoassay marketed by Abbott Laboratories, North Chicago, Ill. (Rotazyme I); a modification of this assay which is now commercially available (Rotazyme II); and a latex agglutination test (Rotalex) recently introduced by Medical Technology Corp., Somerset, N.J. In addition, selected specimens were examined for the presence of rotavirus by electron microscopy, immune electron microscopy, and RNA gel electrophoresis. A total of 40 specimens were positive in the monoclonal antibody enzyme immunoassay, and 136 were negative. Using the results obtained with this procedure as the reference standard, we found the sensitivities of the Rotazyme I, Rotazyme II, and Rotalex tests to be 97.4, 100, and 81.6%, respectively. The specificities of these three procedures were 88.8, 83.9, and 100%, respectively." (Authors' abstract)

115 Dolan KT, Twist EM, Horton-Slight P, Forrer C, Bell LM Jr., Plotkin SA, Clark HF. Epidemiology of rotavirus electropherotypes determined by a simplified diagnostic technique with RNA analysis. *J Clin Microbiol* 1985 May;21(5):753-8

"The incidence and RNA electropherotypes of rotavirus in stools or rectal swabs of children with diarrhea were studied for three rotavirus seasons (1981 through 1984) in Philadelphia, Pa. We used a simplified RNA analysis method involving polyacrylamide gel electrophoresis followed by silver staining. Phosphate-buffered saline suspensions of the stools and swab eluates were examined directly by polyacrylamide gel electrophoresis-silver staining analysis and enzyme-linked immunosorbent assay (Rotazyme; Abbott Laboratories); electron microscopy was performed on solid stool specimens. The RNA analysis results were compared with electron microscopy and enzyme-linked immunosorbent assay results and exhibited a sensitivity and specificity greater than or equal to that of electron microscopy or the enzyme-linked immunosorbent assay. Ten different electropherotypes were detected among the 68 rotavirus RNA-positive specimens examined over the 3-year study. The predominant electropherotype was different in each season. Our results indicate that the polyacrylamide gel electrophoresis-silver nitrate strain RNA analysis of simple unextracted stool suspensions is a uniquely useful diagnostic technique; it rapidly provides both a definitive positive result and immediate determination of the RNA electropherotype, which is of value for epidemiological study." (Authors' abstract)

116 Donohue-Rolfe A, Kelley MA, Bennis M, Keusch GT. Enzyme-linked immunosorbent assay for Shigella toxin. *J Clin Microbiol* 1986 Jul;24(1):65-8

"An enzyme-linked immunosorbent assay (ELISA) was developed for the detection of shigella toxin. For the assay, a mouse monoclonal antibody against the B subunit of the toxin and a rabbit polyclonal antibody against the holotoxin were employed. The monoclonal antibody was used to coat wells of a microtiter plate, and the polyclonal antibody preparation was used as the detecting antibody. The amount of bound polyclonal antibody was determined by using a goat anti-rabbit immunoglobulin G-alkaline phosphatase conjugate and substrate. The ELISA was able to detect as little as 12 pg (0.06 ng/ml) of shigella toxin. The assay was specific for shigella toxin, not detecting a variety of other bacterial enterotoxins and lethal toxins. The ELISA values correlated well with cytotoxin activity during toxin purification. Shigella toxin was detected by ELISA and by immunoblot analysis in human fecal specimens from persons with *S. dysenteriae* infections, demonstrating that this toxin is produced *in vivo*." (Authors' abstract)

117 Durham PJ, Hassard LE. An enzyme-linked immunosorbent assay (ELISA) for antibodies to bovine viral diarrhea virus. *Vet Microbiol* 1990 Mar;22(1):1-10

"A single dilution enzyme-linked immunosorbent assay (ELISA) was developed to detect antibodies to bovine viral diarrhea (BVD) virus in cattle sera. Viral antigen (NADL strain) was grown in a pig kidney cell line (PK15), and after removal of nuclear debris, was purified by ultracentrifugation through a potassium tartrate cushion. Antigen grown in embryonic bovine tracheal epithelial cells was also satisfactory. The test used a high salt

buffer to minimize nonspecific reactivity, polyethylene glycol to enhance the reaction, and Protein G as the labelling agent. Comparative testing with the virus neutralization test (VNT) showed the ELISA results to have a high level of correlation with the VNT titers ($r = 0.83$). In vaccinated animals the ELISA detected antibodies earlier than the VNT. All animals sampled from a BVD-free herd were negative for BVD antibody. The single dilution test showed close agreement ($r = 0.84$) with ELISA values obtained using a serial dilution technique, and also proved to have a high level of reproducibility. The test procedures were relatively easy to carry out, and were economic in their use of materials." (MEDLINE)

118 Duthie R, French GL. Comparison of methods for the diagnosis of typhoid fever. *J Clin Pathol* 1990 Oct;43(10):863-5

119 Echeverria P, Taylor DN, Seriwatana J, Chatkaemorakot A, Khungvalert V, Sakuldaipera T, Smith RD. A comparative study of enterotoxin gene probes and tests for toxin production to detect enterotoxigenic *Escherichia coli*. *J Infect Dis* 1986 Feb;153(2):255-60

A comparative study of enterotoxin gene probes and tests for toxin production to detect enterotoxigenic *Escherichia coli* (ETEC) were done. A stool specimen and a rectal swab were collected from children aged 1-3 years with watery diarrhoea during July-August 1984 in Bangkok, Thailand. ETEC was identified from 38 (12%) of 304 children by enterotoxin assays. ETEC, as identified by testing *E. coli* for production of enterotoxin, were isolated from both rectal swabs and stools of 74% of the study cases. Bacterial growth from fecal specimens hybridized with the cloned enterotoxin gene probes of samples from 84% of the children from whom ETEC were isolated. Among the 5,701 *E. coli* samples tested, 289 (99%) of 293 colonies that hybridized with the cloned heat-labile (LT) probe produced enterotoxin. About 72% of 257 *E. coli* that hybridized with the heat-stable (ST)-H probe and all 20 isolates that hybridized with the ST-P probe produced ST in the suckling mouse assay. Nineteen isolates contained plasmids that hybridized with the LT and ST-H probes, and 54 contained plasmids that hybridized with the ST-H probe. *E. coli* that initially hybridized with the cloned ST-H gene probe hybridized with the cloning vector PBR 322. The ETEC organisms were not evenly distributed in specimens collected from children with diarrhoea. This variation may explain the lack of complete concordance between detection of genes coding for enterotoxin in specimens and testing of individual colonies for enterotoxin production.

120 Echeverria P, Taylor DN, Seriwatana J, Moe C. Comparative study of synthetic oligonucleotide and cloned polynucleotide enterotoxin gene probes to identify enterotoxigenic *Escherichia coli*. *J Clin Microbiol* 1987 Jan;25(1):106-9

Escherichia coli isolated from stools of 2,126 children in Thailand and the Philippines were examined for enterotoxin production and for DNA hybridization with synthetic oligonucleotide and cloned polynucleotide enterotoxin gene probes. A total of 233 infections due to *E. coli* that were detected by one or more of these assays were identified. Of the infection, 75% (164/233) were identified by all three methods. An additional 18% (43/233) were identified by two of three methods. Isolates from 10% (19/183) of infections with *E. coli* that hybridized with both the oligonucleotide and cloned enterotoxin gene probes were non-toxicogenic, as determined by the Y1 adrenal cell and suckling mouse assays. Although synthetic oligonucleotide probes to detect enterotoxigenic *E. coli* are more uniform and easier to use than cloned enterotoxin gene probes, the heat-labile toxin oligo probe used in this study did not identify 13% (11/87) of infections with *E. coli* that produced a heat-labile toxin, as identified with the Y1 adrenal cell assay and the cloned enterotoxin gene probe. Synthetic oligonucleotide probes enable laboratories with only minimal equipment to use DNA hybridisation assays to identify enterotoxigenic *E. coli*. (Modified authors' abstract)

121 Echeverria P, Taylor DN, Leksomboon U, Blacklow NR, Pinnol S, Nataro JP,

Kaper J, Rowe B. Identification of enteric pathogens in the small and large intestine of children with diarrhea. *Diagn Microbiol Infect Dis* 1986 Apr;4(4):277-84

"Enteric pathogens were identified in children with diarrhea from duodenal specimens obtained with a string capsule and from fecal specimens. Rotavirus was identified in stools of 43 of 100 children, and was recovered from the small intestine from nine (21%) children who were excreting this virus. *Shigella* was isolated from stools from 22, *Salmonella* from 17, enterotoxigenic *Escherichia coli* from eight, and *Aeromonas hydrophila* from one of 100 children with diarrhea. In contrast to rotavirus, *Salmonella*, *Shigella*, enterotoxigenic *Escherichia coli*, and *A. hydrophila* were not isolated from the small intestine. Nonenterotoxigenic *Aeromonas* species were recovered from the small intestine, but not the stool of five children. These children were also infected with *Shigella* or with rotavirus; this suggests that *Aeromonas* was not the cause of their diarrhea. None of 51 *Escherichia coli* isolated with the string capsules, or 67 isolated from stool that agglutinated in commercial enteropathogenic *Escherichia coli* antisera were of classical enteropathogenic *Escherichia coli* serotypes. One hundred and five of these 118 *Escherichia coli* did not hybridize with a deoxyribonucleic acid probe for plasmid mediated factors conferring adherence to HeLa cells. Examination of specimens collected with a string capsule from children with diarrhea did not identify any more enteric pathogens than examining stools. Furthermore testing *Escherichia coli* for agglutination in commercial enteropathogenic *Escherichia coli* antisera did not identify *Escherichia coli* of enteropathogenic serotypes." (Authors' abstract)

122 Echeverria P, Hanchalay S, Taylor DN. Serological response to plasmid-encoded antigens in children and adults with shigellosis. *Diagn Microbiol Infect Dis* 1988 Jun;10(2):75-80

123 Echeverria P, Taylor DN, Seriwatana J, Sethabutr O, Chatkaeomrakot A. Use of nucleic acid probes in the diagnosis of diarrheal disorders. *Indian J Pediatr* 1987 Jan-Feb;54(1):15-25

"DNA probes have recently been used for the identification of enteric pathogens. The principle of this technique is described and those enteric pathogens which have been identified by DNA hybridization techniques are discussed. This technique has been used primarily to study the epidemiology of enterotoxigenic, enteroinvasive and enteropathogenic *Escherichia coli*, and identify enteropathogenic determinants in other species of bacteria. DNA hybridization techniques have only been used by well-equipped research laboratories. The recent development of non-radioactive markers for DNA probe should make it feasible for laboratories without ready access to radioisotopes to use this technique in their studies of enteric diseases." (Authors' abstract)

124 Edwards S, Chasey D, Napthine P, Banks J, Hewitt-Taylor C, Cranage MP. A comparison of three rapid diagnostic methods for the detection of rotavirus infection in calves. *Vet Microbiol* 1987 Jan;13(1):19-25

"Three techniques for the detection of rotavirus in faecal samples from calves with neonatal gastroenteritis were compared. A preliminary study indicated that reverse passive haemagglutination (RPHA) was at least as sensitive as the enzyme-linked immunosorbent assay (ELISA). These two immunoassays were compared with the detection of viral RNA by polyacrylamide gel electrophoresis (PAGE) on 209 field samples. Of the 77 samples in which at least one test gave a positive result, 69 were positive by both RPHA and PAGE, but only 49 were also positive by ELISA, indicating a lower sensitivity for the latter test. The overall agreement between RPHA and PAGE was 96%. The reasons for the discrepancies between the tests are discussed." (MEDLINE)

125 Ehara M, Ishibashi M, Watanabe S, Iwanaga M, Shimodori S, Naito T. Fimbriae of *Vibrio cholerae* 01: observation of fimbriae on the organisms adherent to the intestinal epithelium and development of a new medium to enhance fimbriae production. *Trop Med* 1986 Mar;28(1):21-3

Evidence for the presence of fimbriate *Vibrio cholerae* during colonization of the small intestinal surface, and for the reproducibility of fimbriae on the TCG agar are presented. The fimbriate phase of enteropathogenic *V. cholerae* 01 strains was observed under electron microscope during the colonization to the surface of rabbit small intestine. There was no difference in the shape of fimbriae between the two serotypes of El Tor Inaba and El Tor Ogawa. Pathogenic strains were confirmed as capable of producing numerous fimbriae on a solid medium (TCG agar) with the same structure, namely 5 to 7 nm in width and flexible fibres as seen *in vivo*. A cap-like structure observed under electron microscope represented the base plate of the fimbriae in the cytoplasm. Numerous holes identified in the surface of the cell appeared to be channels through which the fimbriae emerge, since their diameter was the same as that of the fimbrial strand, and their frequency and peritrichous distribution corresponded to the degree and pattern of cell fimbriation. This observation suggests that the vast majority of the fimbriae were already detached at cell harvest and that enteropathogenic *V. cholerae* strains produce several hundred of fimbriae per cell. TCG medium containing thiopropine, sodium bicarbonate and EGTA was successful in producing fimbriae from *V. cholerae*; however, the reason still remains to be ascertained.

126 Elden J, Sato S, Yolken R. Specificity of dot hybridization assay in the presence of rRNA for detection of rotaviruses in clinical specimens. *J Clin Microbiol* 1987 Sep;25(9):1809-11

127 Elden JJ, Firoozmand F, Sato S, Vonderfecht SL, Yin FZ, Yolken RH. Detection of group B rotavirus in fecal specimens by dot hybridization with a cloned cDNA probe. *J Clin Microbiol* 1989 Mar;27(3):422-6

128 Endtz HP, Ruijs GJHM, Zwinderman AH, van der Reijden T, Bielev M, Mouton RP. Comparison of six media, including a semisolid agar, for the isolation of various *Campylobacter* species from stool specimens. *J Clin Microbiol* 1991 May;29(5):1007-10

"A recently described semisolid blood-free selective motility medium (SSM) (J. Goossens, L. Vlaes, I. Galand, C. Van den Borre, and J.P. Butzler, *J. Clin. Microbiol.* 27:1077-1080, 1989) was compared with two charcoal-based selective media (charcoal-based selective medium [CSM] and modified charcoal cefoperazone deoxycholate agar [CCDA]), two blood-based media (Skirrow medium [SKM] and CampyBAP), and a passive, 0.65- μ m-pore-size cellulose acetate membrane filter technique for the recovery of campylobacters from stools of patients with diarrhea. A total of 1,980 specimens were tested, 161 of which were found to be positive for campylobacters. *Campylobacter jejuni* was isolated in 148 specimens (91.9%), *C. coli* was isolated in 12 (7.5%), and *C. upsaliensis* was isolated in 1 (0.6%). After 72 h of incubation with a single medium, the cumulative percentages of *Campylobacter*-positive specimens isolated on CSM, CCDA, SKM, and SSM were 87, 83, 80, and 72%, respectively. The filter method alone enabled us to recover 61% of all campylobacters. The *C. upsaliensis* strain was isolated by this method only. The highest isolation rates were observed when two media, including CSM, were combined. The combination of CSM and SSM yielded the highest rates (96%), but these were not statistically different from the rates observed with combinations of CSM and SKM (94%) or of CSM and the filter method (91%). Extending the incubation time from 48 to 72 h led to an increase in the isolation rate regardless of the medium used ($p < 0.001$). CSM and CCDA were the most selective media. SKM and CampyBAP appeared to be the most inhibitory media for the isolation of *C. coli*." (Authors' abstract)

129 Evans DJ, Jr., Evans DG, Graham DY, Klein PD. A sensitive and specific serologic test for detection of *Campylobacter pylori* infection. *Gastroenterology* 1989 Apr;96(4):1004-8

"*Campylobacter pylori* has been associated with gastritis, duodenal ulcer, gastric ulcer, and nonulcer dyspepsia. Evidence that *C. pylori* may be the causative agent or at least

a major contributory factor in peptic ulcer disease has generated intense interest in the development of reliable methods for detecting *C. pylori* infections. We have developed a specific and sensitive enzyme-linked immunosorbent assay (ELISA) that detects serum immunoglobulin G antibodies directed against high molecular weight cell-associated proteins (HM-CAP) of *C. pylori*. In a blinded fashion we tested sera from 300 individuals and found that all of 147 HM-CAP ELISA-negative individuals were also negative for *C. pylori*, as documented by a negative urea breath test; also, 151 of 153 *C. pylori*-positive (by urea breath test) individuals were HM-CAP ELISA-positive. *Campylobacter pylori* was cultured from the two ELISA-negative but infected patients and these isolates did possess HM-CAP antigens, showing that these two individuals had failed to seroconvert. Thus, the specificity and positive predictive value of the HM-CAP ELISA were each 100%; the sensitivity of the assay was 98.7%, and the negative predictive value was 98.6%. The HM-CAP ELISA and the urea breath test both proved valuable for detecting *C. pylori* infection, the urea breath test being a more direct method whereas the ELISA is less expensive and easier to perform. Furthermore, the results of a serologic test such as the HM-CAP ELISA would not be influenced by recent ingestion of bismuth compounds or antimicrobial therapy, which might suppress *C. pylori* and cause a transient false-negative result in the urea breath test." (Authors' abstract)

130 Ezaki T, Takeuchi N, Liu S, Kai A, Yamamoto H, Yabuuchi E. Small-scale DNA preparation for rapid genetic identification of *Campylobacter* species without radioisotope. *Microbiol Immunol* 1988;32(2):141-50

131 Fedorko DP, Lehman SM, Yu PKW, Germer JJ, Anhalt JP. Increased efficiency of stool culture for the detection of *Salmonella* and *Shigella*. *Diagn Microbiol Infect Dis* 1989 Nov/Dec;12(6):463-6

"The Wampole Bactigen *Salmonella*-*Shigella* Latex Agglutination Test (SSLA) (Wampole Laboratories, Cranbury, New Jersey) was evaluated as a possible substitute for blind subculture of selenite broths from stool cultures. Recovery rates of *Salmonella* and *Shigella* from eosin-methylene blue (EMB) agar were reviewed to determine if this medium could be eliminated from primary stool culture. *Salmonella* was detected in 17 of 822 stools by both SSLA and culture. There were 52 false-positive SSLA for *Salmonella* (sensitivity 100%, specificity 93%). Of three *Shigella* isolated on culture, one was SSLA positive, one was SSLA negative, and one was negative by both SSLA and subculture of selenite broth. There were eight false-positive SSLA for *Shigella* (specificity 99%). Of 50 *Salmonella* and 11 *Shigella* isolated from 6200 stools in 1.5 years, two *Shigella* were isolated on EMB only. The SSLA test is a useful screening test for *Salmonella*. By eliminating unnecessary subcultures of selenite broth, it reduces turnaround time by 24 hr for negative stool cultures. The combination of primary culture with SSLA screening of enrichment broth should be adequate for the detection of *Salmonella* and *Shigella* from stool specimens. Our data suggest that EMB or other differential medium should be retained for primary culture to enhance detection of *Shigella*." (Authors' abstract)

132 Fenwick SG, Murray A. Detection of pathogenic *Yersinia enterocolitica* by polymerase chain reaction [letter]. *Lancet* 1991 Feb 23;337(8739):496-7

133 Fernandez-Lago L, Santoyo FJ, Vizcaino N, Chordi A. Class-specific immune response to *Yersinia enterocolitica* serotype O9 antigens as determined by enzyme-linked immunosorbent assay. *J Clin Microbiol* 1991 Jun;29(6):1243-8

"An enzyme-linked immunosorbent assay (ELISA) using lipopolysaccharide (S-LPS) as the antigen was used to analyze the antibody response in rabbits orogastrically and intravenously infected with virulent (plasmid-bearing) *Yersinia enterocolitica* O9 strains (pYV+) and with the avirulent (plasmid-cured) derivatives (pYV-). A significant response of immunoglobulin G (IgG), IgA, and IgM antibodies against the S-LPS antigen

was evident in sera from the rabbits orogastrically infected with pYV+ strains. This immune response was stronger and persisted longer than those obtained with the corresponding pYV- strains. In contrast, few differences were observed in the titers and evolution of IgG, IgA, and IgM antibodies against the S-LPS antigen in rabbits intravenously infected with pYV+ and pYV- strains. These results suggest that the necessity of the virulence plasmid for the establishment of infection by *Y. enterocolitica* serotype O9 is conditioned by the infection route used. When the S-LPS ELISA was compared with the radial immunodiffusion test using the native hapten as the antigen, the results showed that the ELISA technique was more sensitive. However, only those sera obtained between 2 and 8 weeks postinfection from rabbits intravenously infected with plasmid-bearing strains were positive in the radial immunodiffusion test." (Authors' abstract)

134 Ferner WT, Miskuff RL, Yolken RH, Vonderfecht SL. Comparison of methods for detection of serum antibody to murine rotavirus. *J Clin Microbiol* 1987 Aug;25(8):1364-9

"Mice are frequently used as animal models for the study of rotaviral infections. Since natural infection is common in laboratory mice, it is important that rotaviral studies, as well as other studies utilizing suckling mice, employ animals of known immune status to murine rotavirus. A variety of homologous and heterologous enzyme immunoassay systems and an immunofluorescence technique were thus compared to determine the immunoassay that is most effective at detecting adult mice seropositive for rotaviral antibody. It was determined that a homologous enzyme immunoassay inhibition technique utilizing murine rotavirus-derived reagents was the most efficient serologic assay evaluated. A serologic response was consistently detected by this assay by 5 days after experimental rotaviral inoculation of adult mice. A homologous antibody-binding enzyme immunoassay, a heterologous inhibition enzyme immunoassay utilizing antigenically related simian rotavirus (SA-11) reagents, and an immunofluorescence technique utilizing Nebraska calf diarrhea virus antigens were found to be less sensitive for detecting serum antibody to murine rotavirus." (Authors' abstract)

135 Fleck SL, Hames SE, Warhurst DC. Detection of *Giardia* in human jejunum by the immunoperoxidase method. Specific and non-specific results. *Trans R Soc Trop Med Hyg* 1985;79(1):110-3

"Rabbit antiserum to cultured trophozoites of *Giardia intestinalis* (*G. lamblia*) was used to detect organisms in jejunal biopsies using the PAP immunoperoxidase technique. In 30 sections examined from seven cases of giardiasis associated with histological changes in malabsorption, trophozoites were seen in the lamina propria in one instance, although they were otherwise seen in the intestinal lumen and surface mucus. One anti-*Giardia* serum reacted with the neutrophil polymorphs in all infected jejunal biopsies, and in jejunal and rectal biopsies from patients not suffering from giardiasis. The reaction with *Giardia* and with polymorphs could be absorbed out using washed *Giardia* trophozoites but not with preparations of bacteria." (MEDLINE)

136 Flores J, Green KY, Garcia D, Sears J, Perez-Schael I, Avendano LF, Rodriguez WB, Taniguchi K, Urasawa S, Kapikian AZ. Dot hybridization assay for distinction of rotavirus serotypes. *J Clin Microbiol* 1989 Jan;27(1):29-34

137 Flowers RS, Eckner K, Gabls DA, Robison BJ, Mattingly JA, Silliker JH. Enzyme immunoassay for detection of *Salmonella* in foods: collaborative study. *J Assoc Off Anal Chem* 1986 Sep-Oct;69(5):786-98

"A collaborative study was performed in 25 laboratories to validate an enzyme immunoassay (EIA) procedure utilizing 2 specific monoclonal antibodies for rapid detection of *Salmonella* in foods. The EIA was compared with the standard culture procedure for detection of *Salmonella* in 6 food types: ground black pepper, soy isolate, dried whole eggs, milk chocolate, nonfat dry milk, and raw deboned turkey. Uninoculated and

Inoculated samples were included in each food group analyzed, with the exception of poultry which was naturally contaminated. There was no significant difference in the productivity of the EIA and culture procedures at the 5% level for any of the 6 foods. The enzyme immunoassay screening method has been adopted official first action. (MEDLINE)

138 Flowers RS, Klatt MJ, Robison BJ, Mattingly JA, Gabis DA, Silliker JH. Enzyme immunoassay for detection of *Salmonella* in low-moisture foods: collaborative study. J Assoc Off Anal Chem 1987 May-Jun;70(3):530-5

"A collaborative study was performed in 15 laboratories to evaluate a modification of the enzyme immunoassay (EIA) method for detection of *Salmonella* in foods (46.B21-46.B29). The modified EIA requires 18-24 h pre-enrichment, 6-8 h selective enrichment, and 14-18 h M-broth post-enrichment prior to performing the assay, which requires 1-2 h. Total assay time is 40-52 h. The modified method was compared with the standard culture method for detection of *Salmonella* in 5 low-moisture foods: nonfat dry milk, milk chocolate, meat and bone meal, dry whole egg, and ground pepper. The modified method has been adopted official first action for use with low-moisture foods." (MEDLINE)

139 Fongsiri K, Suthienkul O, Taylor DN, Saiborisuth S, Sricharnorn S, Kongvisaisook S, Echeverria P. A comparison of methods for the isolation of *Campylobacter* species from stool specimens. Southeast Asian J Trop Med Public Health 1989 Mar;20(1):139-46

"Eight methods used for isolation of *Campylobacter* species were compared. Using a combination of methods *Campylobacter* species were isolated from 30(11%) of 270 children with diarrhea seen at Bamrasnaradura Infectious Disease Hospital, Nonthaburi, Thailand. The membrane filter method using a gas generating envelope at 37°C identified 73% of the total positive specimens and was found to be the best isolation method for *Campylobacter* species from stool specimens. This method identified two strains that failed to grow on antibiotic containing media, and also gave a higher isolation rate of *C. jejuni* than could be isolated with conventional methods. The combination of the membrane filter method and a selective antibiotic method identified 90% of all isolates. At present the cost of the membrane filter method is higher than other methods. Therefore, the selective antibiotic method (Campy-BAP) with sheep blood under gas mixture at 42°C is recommended for laboratories with limited supplies. Diagnosis by direct smear with 1% basic fuchsin revealed high degree of sensitivity and specificity. This rapid, inexpensive, and simple method could be used to make a presumptive diagnosis of *Campylobacter* enteritis when isolation methods are unavailable." (Authors' abstract)

140 Fontana M, Zuin G, Paccagnini S, Ceriani R, Quaranta S, Villa M, Principi N. Simple clinical score and laboratory-based method to predict bacterial etiology of acute diarrhea in childhood. Pediatr Infect Dis J 1987 Dec;6(12):1088-91

141 Forrer CB, Rodden JM, Clark HF, Friedman HM. Discrepant rotavirus results in two laboratories using the same enzyme immunoassay. Am J Clin Pathol 1989 Jan;91(1):85-7

142 Fukushima H. New selective agar medium for isolation of virulent *Yersinia enterocolitica*. J Clin Microbiol 1987 Jun;25(6):1068-73

A selective agar medium for isolation of virulent *Yersinia enterocolitica* (VYE agar) was developed for the rapid and accurate isolation of virulent *Y. enterocolitica* from environmental samples highly contaminated with environmental *Yersinia* organisms as well as for isolation from clinical specimens. VYE agar provided a quantitative recovery of 51 different strains of virulent *Y. enterocolitica* at 32°C after incubation for 24 hours. The

cefsulodin, irgasan, josamycin, and oleandomycin content of the medium resulted in a high selectivity, while the mannitol and esculin content provided some differentiation. The greatest advantage of VYE agar is that virulent *Y. enterocolitica*, which forms red colonies, is easily differentiated from most environmental *Yersinia* organisms and other Gram-negative bacteria, which from dark colonies with a dark peripheral zone as a result of esculin hydrolysis. Use of VYE agar led to a high recovery of *Y. enterocolitica* biotype 3B serotype 0:3 strains from experimentally inoculated meat samples, compared with that obtained using the CIN agar. Biotype 2 serotypes 0:5, 27 and 0:9 and biotype 1 esculin-negative serotypes 0:4, 32, 0:8, 0:13a, 13v, 0:18, 0:20, and 0:21 (American types) were readily differentiated from other environmental organisms capable of growing on VYE agar. Epidemiological studies on *Y. enterocolitica* should be greatly facilitated by the use of this selective agar medium. (Modified authors' abstract)

143 Fule RP, Kaundinya DV. Comparison of two methods for the detection of heat labile enterotoxin of enterotoxigenic *E. coli* from infantile diarrhoea. Indian J Pathol Microbiol 1986 Apr;29(2):115-20

144 Gandhi BM. A dot immunobinding assay (DIA) on nitrocellulose membrane for the serological detection of antibodies to *Entamoeba histolytica*. Trans R Soc Trop Med Hyg 1986;80(6):883-5

A dot-immunobinding assay on nitrocellulose membranes for serological detection of antibodies to *Entamoeba histolytica* is described. The experiment was carried out by the Department of Gastroenterology and Human Nutrition of the All-India Institute of Medical Sciences, involving 32 subjects comprised of normal healthy controls, and 45 patients with amoebic liver abscess. *E. histolytica* cyst passers were included in the study. The normal control group included males and females of different socio-economic status. Only one of the 32 healthy controls gave a positive dot test. The remaining 31 included 28 samples which were completely negative and gave no colour on the dot test and three samples which gave a mild blue discolouration which were considered negative for the purposes of this study. There was no difference, when, in 15 cases, on washing with Tris-saline buffer, there was complete removal of red blood cells and no interference with the intensity of the final colour of the product. Forty-two of the 45 samples collected from those with amoebic liver abscess gave a deep blue colour (positive test) while the remaining three gave a slightly blue discolouration and these samples were taken as negative. All blood samples were then run simultaneously in the protein enzyme-linked immunosorbent assay (ELISA) technique. A test of proportions was applied. No differences in sensitivity and specificity for serological diagnosis of amoebic liver abscess was noted between the dot immunobinding technique on nitro-cellulose membranes and the ELISA done on micro-ELISA plates.

145 Gandhi BM, Irshad M, Chawla TC, Tandon BN. Enzyme linked protein-A: an ELISA for detection of amoebic antibody. Trans R Soc Trop Med Hyg 1987;81(2):183-5

"Enzyme linked protein-A was used to develop an enzyme linked immunosorbent assay (ELISA) system for the detection of circulating antibodies to amoebic antigen. The specificity of protein-A to bind IgG only through Fc receptors, makes the test more specific for the detection of IgG antibodies to amoebic antigen. The ELISA system was used to detect amoebic antibody in control subjects (56), patients with amoebic liver abscess (79) and *Entamoeba histolytica* cyst-passers (10) and the results compared with those of indirect haemagglutination assay (IHA). The ELISA was more sensitive and detected 74.7% of cases with amoebic antibody in amoebic liver abscess compared with 66.7% detected by IHA. The test was more specific, sensitive and easy to perform and is recommended as a test of choice for the serological diagnosis of amoebic liver abscess." (MEDLINE)

146 Gary GW Jr, Kaplan JE, Stine SE, Anderson LJ. Detection of Norwalk virus antibodies and antigen with a blotin-avidin immunoassay. J Clin Microbiol 1985 Aug;22(2):274-8

"Biotin-avidin Immunoassays (BAIs) were developed to detect Norwalk virus antigen and to measure Norwalk virus antibody. The BAI detected Norwalk virus infections by a fourfold titer rise in antibody in sera or by antigen in stool, with a sensitivity similar to or greater than that of the radioimmunoassay (RIA), and the BAI appeared to be more sensitive than the RIA for detecting antibody in single serum specimens. The BAI antigen test detected Norwalk antigen in all stools that were positive by the RIA, and in two stools negative by the RIA. Of 106 serum pairs tested by both the RIA and BAI, 94 demonstrated seroconversion, or lack of seroconversion, in both assays; 12 gave discordant results. Titers by the BAI generally exceeded those by the RIA by two- or fourfold. The BAI had advantages over the RIA in that it had added sensitivity in the detection of Norwalk antibody, was less time consuming, did not require frequent labeling of antibody, and did not have the problems associated with the use of isotopes." (Authors' abstract)

147 Germani Y, Begaud E, Brethes B, Guesdon JL. [Application of the vapor test for the detection and immunologic determination of *Escherichia coli* heat labile enterotoxin]. Ann Inst Pasteur Immunol 1986 Jan-Feb;137C(1):39-50

"The principle of this thin-layer immunoassay (vapour condensation technique or TVAP) is based on the ability of antibodies to absorb firmly to polystyrene surfaces and to retain their reactivity. A condensation pattern consisting of large confluent water drops is noticable when an antibody-antigen reaction takes place. We used this technique to detect and assay the *Escherichia coli* heat-labile enterotoxin (ETEC LT+) and compared the results of 53 strains (40 positives and 13 negatives) with single radial immune haemolysis, GM1-ELISA and Vero cell culture tests. With the reagents used, this reaction was specific for a toxin dilution up to 1/14. As little as 0.025 micrograms/ml of cholera toxin could be detected. The TVAP-test is simple, rapid and cost-effective. It is thus quite suitable for use in diarrhoeal endemic areas." (MEDLINE)

148 Germani Y, Begaud E, Guesdon JL. Assay for heat-labile *Escherichia coli* enterotoxin using sandwich erythroimmunoassay. Med Microbiol Immunol (Berl) 1987;176(2):89-97

"We investigated the possibility of using a sheep erythrocyte-antibody conjugate as reagent in a sandwich erythroimmunoassay (SERIA) procedure to detect and titrate *Escherichia coli* heat-labile enterotoxin (LT) with the naked eye. In this assay, which is based on the immunological similarity between *Vibrio cholerae* toxin (CT) and LT, rabbit anti-CT IgG was used as immunosorbent, and sheep erythrocytes, sensitized with the rabbit anti-CT antibodies, were used as indicator. The sensitivity of the test was demonstrated by a comparative study using an enzyme-linked immunosorbent assay (ELISA). The results obtained by SERIA with 130 samples correlated well with those of a Vero cell assay and a GM1-ELISA. The test is easy, relatively cheap and as sensitive as other standard techniques; it is particularly suited for field laboratories, especially in tropical countries, and a large number of strains may be examined daily." (MEDLINE)

149 Germani Y, Guesdon JL, Begaud E, Moreau JP. Antibody chimera technique applied to the detection of *Escherichia coli* heat-labile enterotoxin. J Immunol Methods 1987 Apr 2;98(1):83-9

"Covalently prepared chimera antibodies were tested in a ganglioside GM1 erythro-immunoassay (CERIA) for *E. coli* heat-labile enterotoxin (LT) detection. The antibody specific for LT was conjugated with a polyclonal antibody specific for sheep erythrocytes. The assay is based on the specific binding of LT to polystyrene-adsorbed GM1 and subsequent erythro-adsorption via chimera antibody by which the bound toxin is visualized. Enterotoxin titers determined with this CERIA method were similar to those obtained with the Vero cell assay and with ELISA. 5 ng of cholera toxin/ml may be detected with the assay. The CERIA, as described, may be used either qualitatively or quantitatively and is well suited for routine laboratory diagnosis of LT in a culture supernatant of *E. coli*." (MEDLINE)

150 Germani Y, Begaud E, Guesdon J-L, Moreau J-P. GM1 erythroimmunoassay for detection and titration of *Escherichia coli* heat-labile enterotoxin. J Clin Microbiol 1986 Nov;24(5):744-8

"A GM1 ganglioside erythroimmunoassay for the detection of heat-labile *Escherichia coli* enterotoxin (LT) was developed for use in poorly equipped laboratories in developing countries. This assay is based on the immunological similarity between *Vibrio cholerae* toxin and LT and uses cholera toxin antiserum and sheep anti-rabbit immunoglobulin covalently coupled to sheep erythrocytes as conjugate. This assay has the following advantages over other currently available techniques: the reagents it uses are stable, in particular, tanned and sensitized sheep erythrocytes; GM1 ganglioside is commercially available; erythro-adsorption can be read with the naked eye; the test can be completed in 1 day; and as little as 4 ng of *V. cholerae* toxin or LT per ml can be detected accurately. The GM1 ganglioside erythroimmunoassay showed good quantitative and qualitative correlation with the Vero cell assay and the conventional GM1 enzyme-linked immunosorbent assay. The GM1 ganglioside erythroimmunoassay was somewhat less sensitive than the GM1 enzyme-linked immunosorbent assay but more sensitive than the Vero cell assay. Results obtained for 12 LT-positive and 138 LT-negative *E. coli* strains correlated with results obtained with GM1 enzyme-linked immunosorbent and Vero cell assays." (Authors' abstract)

151 Germani Y, Guesdon JL, Begaud E, Moreau JP. [Specific detection of the thermolabile enterotoxin of *Escherichia coli* in human feces by competitive immunoassay tests in the presence of protease inhibitors] Ann Inst Pasteur Microbiol 1987 Nov-Dec;138(6):681-92

"Two simple two-step competitive enzyme-linked immunoassays for human *E. coli* heat-labile enterotoxin (LT) employing microtitration plates coated with rabbit anti-LT antibody (ELISA) or GM1 ganglioside (GM1-ELISA) are described. LT of the test sample competed with the same toxin coupled with horse-radish peroxidase. ELISA and GM1-ELISA were able to detect, respectively, as low as 5 ng and 6.5 ng TLH/ml and up to 9 and 11 micrograms TLH/ml. Both techniques were applied to the study of 167 infant diarrhoeas; ETEC producing LT were identified in 17 diarrhoeal stools. When the faeces were diluted in phosphate buffer, only 17.6% (3 stools) and 29% (5 stools) of LT-positive faeces were identified in ELISA and GM1-ELISA. When the stools were diluted with phenylmethylsulphonyl fluoride (PMSF), a synthetic protease inhibitor, 82% (14 stools) and 88% (15 stools) of LT-positive stool supernatants were detected. Aprotinin, another protease inhibitor, was without effect and foetal calf serum, horse serum and bovine serum albumin enabled detection of only a low percentage of LT-positive stools." (MEDLINE)

152 Gerna G, Sarasini A, Passarini N, Torsellini M, Parea M, Battaglia M. Comparative evaluation of a commercial enzyme-linked immunoassay and solid-phase immune electron microscopy for rotavirus detection in stool specimens. J Clin Microbiol 1987 Jun;25(6):1137-9

"Using solid-phase immune electron microscopy (SPIEM) as a reference test, we examined 151 stool specimens from infants and young children with acute gastroenteritis for rotavirus detection by a one-step commercial enzyme-linked immunosorbent assay (ELISA) with labeled monoclonal antibody. Of the 83 samples determined to be positive for rotavirus by SPIEM, 82 were detected as positive by the monoclonal antibody ELISA (sensitivity, 98.7%), while 67 of the 68 specimens determined to be negative by SPIEM were correctly detected as negative by the ELISA (specificity, 98.5%). The diagnostic accuracy of the ELISA kit was 98.6%. Thus, the one-step monoclonal antibody ELISA, which can be completed in less than 90 min, appears to be highly suitable for the rapid and reliable detection of rotavirus in stools." (Authors' abstract)

153 Gerna G, Sarasini A, Coulson BS, Parea M, Torsellini M, Arbustini E, Battaglia M.

Comparative sensitivities of solid-phase immune electron microscopy and enzyme-linked immunosorbent assay for serotyping of human rotavirus strains with neutralizing monoclonal antibodies. *J Clin Microbiol* 1988 Jul;26(7):1383-7

154 Gerna G, Sarasini A, Zentilin L, Di Matteo A, Miranda P, Parea M, Battaglia M, Milanesi G. Isolation in Europe of 69 M-like (serotype 8) human rotavirus strains with either subgroup I or II specificity and a long RNA electropherotype. *Arch Virol* 1990;112(1-2):27-40

"During an epidemiological study on the prevalence of human rotavirus (HRV) serotypes 1-4 in Europe, we found that some strains could not be typed. However, when a monoclonal antibody directed to serotype 8 HRV was included in the typing assay, we detected seven 69 M-like (serotype 8) strains, six from Finland and one from Italy. The previously reported serotype 8 HRV strains, 69 M, B 37, and B 38 isolated in Indonesia, were of subgroup I specificity and presented a peculiar "super short" RNA electropherotype. In contrast, all the seven European strains possessed a long RNA pattern, and one of them had subgroup II specificity. Three of these strains were adapted to growth in cell cultures and were further characterized by neutralization and by Northern blot hybridization. They appeared to be closely related to serotype 8 HRV strain 69 M by neutralization, but showed partial homology with several human and animal strains by hybridization. The epidemiological importance of these serotype 8 strains circulating in Europe should be investigated, in view of their possible inclusion in a rotavirus vaccine." (MEDLINE)

155 Glaquinto C, Vanin M, Angiani F, Errico G, Ruga E, D'Elia R. A new screening test for rotavirus infection. *Infection* 1985 Nov-Dec;13(6):260-2

"A simple and rapid staphylococcal coagglutination test, using rabbit antisera prepared against Nebraska calf diarrhea virus (NCDV), is described for the detection of rotavirus in neonatal fecal specimens. When the samples were examined directly using the coagglutination test, more than 60% of the specimens agglutinated the control reagent. These non-specific reactions were markedly reduced by preincubation of the specimens with non-immune rabbit serum and further heating at 80°C for 45 min. Such treatment did not reduce the specific activity in the coagglutination test when rotavirus-containing stools were tested. The coagglutination test was compared with ELISA in 290 stools positive or negative for rotavirus. The sensitivity of the coagglutination test was 92%, the specificity 91% and the predictive value 31%. These results indicate that coagglutination is a suitable test for rapid screening of rotavirus infection in clinical practice." (MEDLINE)

156 Gicquelais KG, Baldini MM, Martinez J, Maggi L, Martin WC, Prado V, Kaper JB, Levine MM. Practical and economical method for using blotting DNA probes with bacterial colony blots to identify diarrhea-causing *Escherichia coli*. *J Clin Microbiol* 1990 Nov;28(11):2485-90

157 Gilchrist MJR, Brett TS, Moultney K, Knowlton DR, Ward RL. Comparison of seven kits for detection of rotavirus in fecal specimens with a sensitive, specific enzyme immunoassay. *Diagn Microbiol Infect Dis* 1987 Dec;8(4):221-8

"A sensitive, specific enzyme immunoassay (SSEIA) was compared to four commercial, enzyme-linked immunosorbent assay (ELISA) kits and three latex agglutination assay (LAA) kits: (1) *Rotavirus EIA*, International Diagnostic Laboratories (IDL), (2) *Pathfinder*, Kallestadt (KAL), (3) *Rotavirus Bio-EnzaBead*, Litton (LIT), (4) *Rotazyme II*, Abbott (RTZII), (5) *Slidex Rota-Kit*, bioMerieux (SRK), (6) *Meritec-Rotavirus*, Meridian (MER), and (7) *Rotalax*, Medical Technology Corporation (RLX). The SSEIA was chosen as the reference method due to its greater sensitivity in comparison to immunoelectron microscopy and polyacrylamide gel electrophoreses of viral RNA segments. Upon evaluation of 136 specimens (of which 44 were positive by SSEIA), the ELISA kits (LIT, KAL, IDL, and RTZII) had sensitivities of 80%, 98%, 91% and 84%; specificities of 95%, 78%, 100%, and

88%; i. positive predictive values (PPV) of 88%, 68%, 100%, and 77%; and negative predictive values (NPV) of 91%, 99%, 96%, and 92%. When compared with SSEIA, the three LAA tests (SRK, MER, and RLX) had sensitivities of 73%, 75%, and 62%; specificities of 99%, 93%, and 95%; PPVs of 97%, 85%, and 84%; and NPVs of 88%, 89%, and 84%. LAA test results appeared to be reliable, if positive, but the sensitivities of these tests were less than those of the ELISA tests. The ELISA tests that employed specimen specific negative controls were superior in minimizing false positive reactions." (Authors' abstract)

158 Girdwood RWA. Protozoan' Infections in the Immunocompromised patient - the parasites and their diagnosis. J Med Microbiol 1989 Sep;30(1):3-16

159 Glassman MS, Dallal S, Berezin SH, Bostwick HE, Newman LJ, Perez-Perez GI, Blaser MJ. *Helicobacter pylori*-related gastroduodenal disease in children: diagnostic utility of enzyme-linked immunosorbent assay. Dig Dis Sci 1990 Aug;35(8):993-7

160 Goh MH, Lal KPF, Ho LC, Ng GC, Singh M. A comparison of the ELISA with other serological tests in the serodiagnosis of amoebiasis. Southeast Asian J Trop Med Public Health 1989 Sep;20(3):399-405

161 Goka AKJ, Rolston DDK, Mathan VI, Farthing MJG. Diagnosis of giardiasis by specific IgM antibody enzyme-linked immunosorbent assay. Lancet 1986 Jul 26;2(8500):184-6

The diagnostic value of measuring specific serum anti-*Giardia* IgM responses was evaluated in patients undergoing investigation of diarrhoea in the United Kingdom and in an area endemic for giardiasis in India. An enzyme-linked immunosorbent assay (ELISA) was used to detect serum IgM and IgG antibodies against *Giardia* in 52 patients with diarrhoea. In giardiasis patients, there was a specific anti-*Giardia* serum IgM response; in two of three patients studied longitudinally, IgM levels had fallen to normal 2-3 weeks after treatment. At serum dilutions of 1/100 to 1/400 there was almost complete separation in optical density readings between patients with *Giardia* and *Giardia*-free controls, a finding equally true for both Indian and UK patients. The sensitivity and specificity of the anti-*Giardia* IgM ELISA were both 96%. Serum anti-*Giardia* IgG was detected in patients and controls, but tests were unable to distinguish current from previous infections. The serum anti-*Giardia* IgM ELISA is simple to perform and useful in identifying patients with current infections.

162 Goka AKJ, Rolston DDK, Mathan VI, Farthing MJG. Diagnosis of *Strongyloides* and hookworm infections: comparison of faecal and duodenal fluid microscopy. Trans R Soc Trop Med Hyg 1990 Nov-Dec;84(6):829-31

163 Goka AKJ, Rolston DDK, Mathan VI, Farthing MJG. The relative merits of faecal and duodenal juice microscopy in the diagnosis of giardiasis. Trans R Soc Trop Med Hyg 1990 Jan-Feb;84(1):66-7

"We compared the diagnostic accuracy of microscopical examination of multiple faecal specimens with duodenal juice examination in the diagnosis of giardiasis. Of 292 patients who had stool microscopy and duodenal aspirate, *Giardia* were identified in either stools or duodenal fluid from 73 patients (25%). Giardiasis was diagnosed in 62 (73%) with the first faecal specimen, but examination of 3 specimens increased the diagnostic yield to 85%. *Giardia*, however, were found in only 32 of 73 duodenal aspirates examined (44%). This finding is contrary to the widely held belief that duodenal fluid examination is superior to stool microscopy for the diagnosis of giardiasis. The 2 approaches are complementary, however, since *Giardia* was found in duodenal fluid, only, from 15% of patients." (Authors' abstract)

164 Goldie J, van Zanten SJOV, Jalali S, Hollingsworth J, Riddell RH, Richardson H,

Hunt RH. Optimization of a medium for the rapid urease test for detection of *Campylobacter pylori* in gastric antral biopsies. *J Clin Microbiol* 1989 Sep;27(9):2080-2

165 Goldin AJ, Apt W, Agullera X, Zulantay I, Warhurst DC, Miles MA. Efficient diagnosis of giardiasis among nursery and primary school children in Santiago, Chile by capture ELISA for the detection of fecal *Giardia* antigens. *Am J Trop Med Hyg* 1990 Jun;42(6):538-45

"Fecal samples were obtained from 722 of 820 children attending 7 nursery schools and 1 primary school in the city of Santiago, Chile. Microscopy of formal-ether concentrates showed that 33% of the children were infected with *Giardia lamblia*. Prevalences among primary school students (5-10 years of age) of *G. lamblia* (38%), *Endolimax nana* (43%), and *Entamoeba coli* (25%) were overall higher than among nursery school students (3 months-5 years of age; prevalences 29%, 21%, and 16%, respectively). There was no apparent association between socio-economic intake and levels of *G. lamblia* infection: the private nursery school had the highest recorded level of infection (40%). One hundred sixty-two triplicate stool specimens were used to compare microscopy with an enzyme-linked immunosorbent assay (ELISA) for the detection of *Giardia* fecal antigens. The ELISA was highly sensitive and specific either visually (95% and 97%, respectively) or by optical density determination (99% and 96%, respectively). Incorporation of non-immune rabbit immunoglobulin-coated control wells did not enhance sensitivity and specificity. The antigen detection ELISA is an extremely effective tool for the epidemiological investigation of giardiasis." (Authors' abstract)

166 Gomez J, Estes MK, Matson DO, Bellinzoni R, Alvarez A, Grinstein S. Serotyping of human rotaviruses in Argentina by ELISA with monoclonal antibodies. *Arch Virol* 1990;112(3-4):249-59

"Human rotaviruses are the major, recognized cause of infantile diarrhea worldwide. Characterization of naturally occurring human isolates indicates that there are six human rotavirus serotypes, four of which (serotype 1 to 4) are widespread. We utilized monoclonal antibodies specific for the VP of serotypes 1, 2, 3, and 4 as capture antibody in a sandwich enzyme-linked immunosorbent to serotype rotaviruses directly in stool samples. The stool samples were collected from 1983 through 1986, from two epidemiologic studies in the area of Buenos Aires, Argentina. All four serotypes assayed were found serotype 2 and 3 viruses, which were detected most frequently in 1983 and 1984, were virtually undetected in 1985 and 1986 (chi 2 = 23, P = <0.001 for this difference). No significant difference was noted among the three collection sites for serotype prevalence. These results indicate that the changing predominance of rotavirus serotype in a given region can involve multiple serotypes at the same time. Analysis of an outbreak of diarrhea in two neighboring families which occurred during a prospective study of community diarrhea documented inter- and intra-family spread of one serotype of virus." (MEDLINE)

167 Gomez JA, Bercovich JA, Grinstein S. [Comparison of enzyme immunoassay, counterimmunoelectrophoresis and polyacrylamide gel electrophoresis for the diagnosis of rotaviruses]. *Rev Argent Microbiol* 1985;17(2):111-4

"Detection of human rotaviruses by enzyme-linked immunosorbent assay (ELISA), provided by the World Health Organization, in 100 stool specimens was compared with the sensitivity of detection by counterimmunoelectrophoresis (CIED) and polyacrylamide gel electrophoresis (PAGE). Table 1 shows that 34 samples were found to contain rotavirus antigens by ELISA and in other 4 specimens false negative results were observed in the ELISA confirmatory test because of high antigen concentration. The ELISA confirmatory test was repeated with the 4 samples diluted 1/40 and they became positives. It was found that, of the 38 ELISA positive samples, 35 were positive by PAGE (92%) and 24 by CIED (63%). Detection of human rotaviruses in stools by PAGE were almost as sensitive as ELISA, but the direct visualization of viral RNA fragments, provided

a distinct advantage since no confirmatory test was necessary. Besides, the surveillance of rotavirus electrophoretotypes constitutes a very useful tool in epidemiological studies. In order to the future possible use of a rotavirus vaccine in our country. By the other hand, it was found CIED less sensitive than ELISA and PAGE. This disadvantage might be tolerated in a diagnostic setting, only if no other test was available for rotavirus diagnosis." (MEDLINE)

168 Gorbachev EN, Verbov VN, Galko NV, Makarova NG, Vashukova SS. [Immunoenzyme analysis in the diagnosis of human rotavirus gastroenteritis (methodological aspects)]. *Vopr Virusol* 1986 Nov-Dec;31(6):743-6

169 Gorbachev EN, Verbov VN, Artiukhov AI, Noskov FS. [The use of commercial immunoenzyme and latex preparations for the demonstration of human rotavirus antigens]. *Zh Mikrobiol Epidemiol Immunobiol* 1989 Aug;(8):68-73

"The results of using enzyme immunoassay and latex preparations for the diagnosis of rotavirus gastroenteritis are presented. High effectiveness of the enzyme immunoassay system developed in the USSR with latex diagnostic agents, such as Rotalex (Orion Diagnostica, Finland), Sildex Rota Kit (BioMerieux, France), The Wellcome Rotavirus Latex Kit (Wellcome Foundation Ltd., Great Britain), 48-63% and 21-41% respectively, has been noted. The results of the comparison of the system developed in the USSR with Wellcozyme Rotavirus, an enzyme immunoassay system manufactured by Wellcome Foundation Ltd. (Great Britain), are practically comparable. The results of the block test and the confirmation test used for control indicate that the Soviet preparation is specific. Materials on the practical evaluation of the assay system at health institutions are presented. Good prospects for the use of this system in the diagnosis of rotavirus gastroenteritis, as well as in the realization of epidemiological surveillance on this infection, are substantiated." (MEDLINE)

170 Gorelov AL, Levina GA, Prozorovskii SV. [The possibilities of using variants of immunoenzyme analysis for detecting L-form *Salmonella typhi* bacteria in biological fluids]. *Zh Mikrobiol Epidemiol Immunobiol* 1989 Aug;(8):60-4

"The possibility of using, on principle, the enzyme immunoassay (EIA) in different modifications for the detection of *S. typhi* L-forms in biological fluids (blood, urine) was established. The inhibiting variant of EIA showed the highest sensitivity: 1 ng/ml. The direct sandwich variant permitted the quantitative determination of the antigen of *S. typhi* L-form in the widest range of 20-500 ng/ml. The indirect enzyme immunometric variant permitted the detection of *S. typhi* L-forms with a sensitivity of 10(5) colony-forming units per ml only in urine." (MEDLINE)

171 Gouvea V, Glass RI, Woods P, Taniguchi K, Clark HF, Forrester B, Fang Z-Y. Polymerase chain reaction amplification and typing of rotavirus nucleic acid from stool specimens. *J Clin Microbiol* 1990 Feb;28(2):276-82

172 Gouvea VS, de Castro L, Pereira HG. A combined dot nitrocellulose-enzyme immunoassay for rotavirus and adenovirus. *J Virol Methods* 1987 Oct;18(1):57-65

"A combined enzyme immunoassay using nitrocellulose membrane as solid support is described for the detection of rotavirus and adenovirus in faeces from children with gastroenteritis. Its sensitivity and specificity are comparable to those of a previously described assay performed on plastic microplates (Pereira et al. (1985) *J. Virol. Methods* 10, 21-28). The introduction of nitrocellulose membrane as support for the immune reactions greatly simplifies the multiple washing steps and precludes the need for disposable plastic plates." (MEDLINE)

173 Goyal SM, Rademacher RA, Pomeroy KA. Comparison of electron microscopy with three commercial tests for the detection of rotavirus in animal feces. *Diagn Microbiol Infect Dis* 1987 Mar;6(3):249-54

"Three commercial test kits were evaluated to detect the presence of rotavirus antigens in bovine, porcine, and turkey feces. Two of the assays, Rotalex (Medical Technology Corporation, Somerset, NJ) and Virogen-Rotatest (Wampole Labs, Cranbury, NJ) are latex agglutination tests (LA), while the third, Pathfinder (Kallestad, Austin, TX) is an enzyme immunoassay. The clinical usefulness of these assays was evaluated by comparing their results with those of direct electron microscopy (EM). A total of 135, 92, and 211 samples of animal feces were tested by Rotalex, Virogen, and Pathfinder, respectively. All samples were examined by EM as a reference procedure. The overall agreement of the three commercial assays with EM was 53%, 66%, and 83% for Rotalex, Virogen, and Pathfinder, respectively. Based on these results, we consider Pathfinder as an attractive alternative to EM for the detection of rotavirus in animals. Of the two LA tests, Virogen was found to be a little more sensitive and specific." (Authors' abstract)

174 Grahnquist L, Stintzing G, Alam AN, Svensson L. Rapid detection of rotavirus in faeces by a slide latex agglutination test as compared with an enzyme-linked immunosorbent assay. *J Diarrhoeal Dis Res* 1987 Sep;5(3):178-80

"A commercial latex agglutination slide test for rotavirus detection (Rotascreen^R) was compared with an enzyme-linked immunosorbent assay (ELISA) in a hospital setting using 225 patients, 6-24 months of age, who came with clinical symptoms suggestive of rotavirus diarrhoea. Of 225 specimens 120 (53%) were found positive with the ELISA compared with only 69 (31%) using the Rotascreen. Twelve specimens gave equivocal results with Rotascreen, 2 of which were positive and the remaining 10 were negative using the ELISA. Excluding the results of these 12 specimens by both the assays, the Rotascreen showed a specificity of 95% and sensitivity of 54%, in comparison with results in the ELISA. Rotascreen is a rapid and simple test with a satisfactory specificity but a low sensitivity." (Authors' abstract)

175 Grandien M, Pettersson CA, Svensson L, Uhnöo I. Latex agglutination test for adenovirus diagnosis in diarrheal disease. *J Med Virol* 1987 Dec;23(4):311-6

"A commercial latex agglutination test for diagnosis of adenovirus in diarrheal disease (Adenolex, Orion Diagnostica, Finland) was evaluated by comparison with the results obtained by ELISA, electron microscopy (EM), and virus isolation. Fifty specimens originated from the diagnostic routine, and 50 were selected from a previous epidemiological study on the etiology of diarrheal disease in children. Thirteen of the 100 specimens reacted with the latex control, impairing interpretation of the results. Although the ELISA detected adenovirus antigen in 10(2) higher dilutions than the latex agglutination test, a total agreement was obtained between results by the two tests for 87 specimens including 42 positives. The two additional positives found by EM and virus isolation could not be diagnosed by the latex agglutination test. Of 37 specimens containing enteric adenoviruses (types 40 and 41), the agglutination test diagnosed all but 4 specimens containing type 41 virus. These four specimens were negative also by ELISA and adenovirus had been detected by virus isolation on the 293 cell line. The latex agglutination test gave positive results with nine specimens containing adenovirus types other than the enteric types 40 and 41. The latex agglutination test was found to be a rapid and simple method for the detection of adenovirus in diarrheal disease. Compared to ELISA and EM, the sensitivity was 100% and 95% respectively, and the specificity 100%." (MEDLINE)

176 Grauballe PC, Hornsleth A, Hjelt K, Krasilnikoff PA. Detection by ELISA of immunoglobulin G subclass-specific antibody responses in rotavirus infections in children. *J Med Virol* 1986 Mar;18(3):277-81

"IgG subclass-specific antibody responses to a human subgroup 2 rotavirus were studied in 26 children by ELISA by use of monoclonal antibodies specific to the four human IgG subclasses. One hundred twenty-nine serum samples were obtained before, during, and after an episode of rotavirus-induced diarrhoea in these patients. When

these sera were investigated, an increase in IgG1 and IgG3 subclass-specific antibodies was detected in all 26 patients. IgG3 antibodies reached a peak concentration 1 week after rotavirus was detected in faecal samples and then progressively declined over the following months, whereas the peak concentration of IgG1 subclass antibodies was found 2 months later and seemed to persist thereafter. IgG2 rotavirus-specific subclass antibodies were never found and IgG4 subclass antibodies were detected only in sera from seven of the 26 patients." (MEDLINE)

177 Green E, Warhurst D, Williams J, Dickens T, Miles M. Application of a capture enzyme immunoassay in an outbreak of waterborne giardiasis in the United Kingdom. *Eur J Clin Microbiol Infect Dis* 1990 Jun;9(6):424-8

"A capture enzyme immunoassay (EIA) for the detection of *Giardia lamblia* antigen was used to examine 136 faecal samples collected during an outbreak of waterborne giardiasis in a city in the UK. Six cases of *Giardia lamblia* infection were detected that had previously not been diagnosed by microscopy. The capture EIA provides an efficient means of processing large numbers of samples for prompt and accurate assessment of an epidemic. It may also facilitate rapid tracing of epidemic sources." (MEDLINE)

178 Green EL, Miles MA, Warhurst DC. Immunodiagnostic detection of *Giardia* antigen in faeces by a rapid visual enzyme-linked immunosorbent assay. *Lancet* 1985 Sep 28;2(8457):691-3

"An enzyme-linked immunosorbent assay (ELISA) was developed for the detection of *Giardia* antigen in human faeces. 136 faecal samples were tested; 76 were positive and 27 negative for giardia cysts on microscopy, and 33 samples contained a range of other parasites. The ELISA sensitivity exceeded 98% and its specificity was 100%, by both colorimetry and direct visual interpretation. The assay could trace the decline and disappearance of antigen in faeces rendered microscopically negative by chemotherapy. This ELISA, which can be completed in less than 3 h and which does not rely on complex equipment for interpretation, should be very useful in diagnostic and epidemiological applications." (Authors' abstract)

179 Green KY, James HD, Jr., Kapikian AZ. Evaluation of three panels of monoclonal antibodies for the identification of human rotavirus VP7 serotype by ELISA. *Bull WHO* 1990;68(5):601-10

"Three panels of monoclonal antibodies used for rotavirus serotype identification by enzyme-linked immunosorbent assay (ELISA) were evaluated at the National Institutes of Health, USA, to identify antibodies suitable for distribution to laboratories involved in WHO-sponsored trials of rotavirus vaccines. Two of the panels were comparably effective in identifying the serotype of each of the human rotavirus reference strains of serotype 1, 2, or 3. In addition, one of the panels included a monoclonal antibody that was effective in identifying strains of serotype 4. However, two different lots of a third, commercially available panel were not effective in identifying the eight strains representing the four serotypes. A third shipment of this panel was therefore tested using revised instructions and, under these conditions, it was effective in serotyping seven of the eight reference strains. It appears that a battery of monoclonal antibodies for each serotype may be required to identify antigenic variants within a serotype. Additional studies are needed to assess the extent of antigenic variation in rotavirus field strains." (Authors' abstract)

180 Grohn K, Genigeorgis C. Adaption of ELISA for the detection of *Campylobacter* antibodies and its application in seroepidemiological studies in sheep and cattle herds. *Acta Vet Scand* 1985;26(1):30-48

181 Grom J, Bernard S. Virus enzyme-linked cell immunoassay (VELCIA): detection and titration of rotavirus antigen and demonstration of rotavirus neutralizing and total antibodies. *J Virol Methods* 1985 Feb;10(2):135-44

"Virus enzyme-linked cell immunoassay (VELCIA) for detection and titration of rotavirus antigen has been developed. Wild-type porcine rotavirus antigen can be detected and titrated directly from the fecal material within 24 h. Porcine OSU strain can be titrated higher than 10^6 (-8). The method has also been introduced for the demonstration of rotavirus neutralizing and total antibodies. In VELCIA the advantages of the cell culture system for virus isolation are combined with enzyme immunodetection and spectrophotometrical reading of the test." (MEDLINE)

182 Gruhn I, Gunther H, Schmidt J, Otto P, Vogt KH. [Comparative studies of the detection of rotavirus antigen with electron microscopy, countercurrent electrophoresis and enzyme immunoassay] *Z Gesamte Hyg* 1986 Apr;32(4):266-8

183 Grundy MS, Voller A, Warhurst D. An enzyme-linked immunosorbent assay for the detection of *Entamoeba histolytica* antigens in faecal material. *Trans R Soc Trop Med Hyg* 1987;81(4):627-32

"This paper describes a method for the detection of *Entamoeba histolytica* antigens in stool samples using a multi-layer ELISA. The method is sensitive and specific, showing no interference with other intestinal parasites, e.g. *E. coli*, *E. hartmanni*, *Endolimax nana*, *Iodamoeba buetschlii*, *Hymenolepis nana*, *Giardia lamblia*, *Trichomonas* and *Ascaris*. The method provides a rapid and simple screening assay for *E. histolytica* infections and should assist in diagnosis and epidemiological studies of the disease." (Authors' abstract)

184 Guerrant RL, Shields DS, Thorson SM, Schorling JB, Groschel DHM. Evaluation and diagnosis of acute infectious diarrhea. *Am J Med* 1985 Jun 28;78(suppl 6B):91-8

"The appropriate approach to the diagnosis and management of acute infectious diarrhea is determined by the frequency and setting of the illness, the recognizable causes or syndromes, the cost and yield of available diagnostic tests, and the treatability of the disease. Acute diarrhea affects everyone throughout the world from one to more than six times each year, depending on age, location, and living conditions. The range of identifiable viral, bacterial, and parasitic etiologies is great, and the cost of indiscriminate use of etiologic studies for diagnosis is prohibitive. Because of its insensitivity for many organisms and poor selection of cases for testing, routine stool culture has been one of the most costly and ineffective microbiologic tests; the cost per positive result has traditionally exceeded \$900 to \$1,000. The appropriate treatment for the vast majority of cases (independent of their cause) is simple and effective: oral glucose- and electrolyte-containing rehydration solution. On the basis of an appropriate history and understanding of pathogenesis, fecal specimens can be selectively obtained and promptly examined for leukocytes and parasites, and the common noninflammatory diarrheas can be separated from the inflammatory infections in order to focus further studies on the latter group. The bacteria for which specific antimicrobial therapy should be considered usually cause inflammatory diarrhea in the United States. Therefore, only when the history or fecal leukocyte findings indicates an inflammatory process it is appropriate to culture for the routine invasive bacterial pathogens. In sporadic inflammatory diarrhea, culture methods should include those for *Campylobacter jejuni* as well as *Salmonella* and *Shigella*. Several special circumstances may prompt a consideration of parasites (including *Giardia*, *Entamoeba*, *Strongyloides*, *Cryptosporidium*), *Vibrio*, *Yersinia*, *Clostridium difficile*, enterotoxigenic *Escherichia coli*, food-borne agents, or sexually transmitted pathogens. The practical value of specific identification of rotaviruses (by enzyme-linked immunosorbent assay, Rotazyme, or electron microscopy) is primarily epidemiologic, particularly in hospitalized infants or young children. Using such a selective approach to fecal culture will greatly increase its yield and can reduce the cost per positive result from \$1,000 to less than \$150. (Authors' abstract)

185 Guttman-Bass N, Tchorsh Y, Marva E. Comparison of methods for rotavirus detection in water and results of a survey of Jerusalem wastewater. *Appl Environ Microbiol* 1987 Apr;53(4):761-7

"Methods for the detection of viable rotaviruses and rotavirus antigen in water were developed and compared. The methods included laboratory-developed enzyme-linked immunosorbent assays (ELISAs) with chromogenic and luminescent substrates, commercial Rotazyme and Enzygnost ELISAs, and an indirect immunofluorescent assay. Of the methods tested, the immunofluorescent assay and the Enzygnost ELISA were the most sensitive for the simian rotavirus SA-11. All of the methods were positive for human rotavirus from clinical specimens. Seeded SA-11 rotavirus was concentrated from water by adsorption to and elution from Zeta Plus filters followed by organic flocculation. Interference with the assays by components of the wastewater concentrates was minimal for the ELISAs, although the undiluted organic flocs were cytotoxic for the immunofluorescent assay. A survey of Jerusalem wastewater was carried out over the course of 1 year, and samples were assayed for rotaviruses and enteroviruses. Although enteroviruses were found in almost all of the samples, all samples were negative for rotaviruses. The concentration of rotaviruses in the wastewater was thus below the detection limit of the method used." (Authors' abstract)

186 Harford JP. An evaluation of a commercially available enzyme immunoassay test for the rapid detection of salmonellae in food and environmental samples. *Epidemiol Infect* 1987 Aug;99(1):127-36

"A total of 91 food and environmental samples were examined for the presence of salmonellae using a commercially available enzyme immunoassay kit (EIA) and a conventional culture technique. A 78% agreement was obtained, but reexamination of culture-negative, EIA-positive samples gave agreement of 86%. The problem of comparing EIA and culture results is discussed. A partially selective pre-enrichment broth was tested in 37 samples and gave better EIA ratios. Artificially contaminated cooked foods gave 100% agreement." (Author's abstract)

187 Hariharan H, Booth BA, Brickman TJ, Katt WC, Boesman-Finkelstein M, Finkelstein RA. Competitive enzyme-linked immunosorbent assay for cholera-related enterotoxins in *Salmonella typhimurium*. *J Clin Microbiol* 1986 Aug;24(2):298-300

"We report a rapid competitive enzyme-linked immunosorbent assay to screen *Salmonella typhimurium* strains for cholera-related enterotoxin antigens. Polymyxin B extracts of bacterial cells from synchase-glucose broth cultures of 7 of 15 strains gave positive results. The specificity of the test was confirmed with known heat-labile-enterotoxin-positive and -negative *Escherichia coli* strains which gave significantly different values." (Authors' abstract)

188 Harne SD, Sharma VD, Rahman H. Paw oedema test for detection of *Salmonella* enterotoxin: modification and standardization. *Indian J Exp Biol* 1990 Dec;28(12):1141-4

189 Helba I, Girgis NI, Farid Z. Enzyme-linked immunosorbent assays (ELISA) for the diagnosis of enteric fever. *Trop Geogr Med* 1989 Jul;41(3):213-7

"Serum samples from 10 patients with culture positive *Salmonella typhi* infection, 30 febrile non-typhoidal patients, and 22 healthy subjects were tested for both *Salmonella* common structural antigen (CSA) and antibody to pathogenic species of *Salmonella*. Serum, urine and stool specimens from these patients were used to detect *Salmonella* antigen(s). IgM anti-*S. typhi* lipopolysaccharide antibody gave the best discrimination between typhoid patients, febrile non-typhoidal cases, and healthy control subjects. The highest IgM titer was recorded in the acute phase of the illness. Antigen detection in serum was highly sensitive (87.5%) and specific (100%), and always occurred early in the acute phase of the illness. Detection of antigen in stool was highly sensitive (87.5%) and specific (96.6%), but it usually was not detected until 3 weeks after the onset of acute illness. Antigen detection in urine was less sensitive and specific compared with assays of stool and blood. The study provides good evidence that ELISA for detection of *Salmonella* CSA and antibody can aid in the diagnosis and confirmation of infections caused by *Salmonella*. (Authors' abstract)

190 Herbrink P, van den Munckhof HAM, Bumkens M, Lindeman J, van Dijk WC. Human serum antibody response in *Campylobacter jejuni* enteritis as measured by enzyme-linked immunosorbent assay. Eur J Clin Microbiol Infect Dis 1988 Jun;7(3):388-93

"An ELISA for detection of IgG, IgA, and IgM antibody using an acid-glycine extract from *Campylobacter jejuni* as antigen was developed. To determine the value of this assay for the diagnosis of acute *Campylobacter jejuni* infections, the IgG, IgA, and IgM immune response against *Campylobacter jejuni* was investigated at various timepoints after infection in patients with culture-proven infection. A total of 112 sera from 46 patients and 78 sera from a control group were tested. All but one of the 46 patients with culture-proven *Campylobacter jejuni* enteritis developed IgG antibodies against *Campylobacter jejuni*. IgA and IgM ELISA both showed 97% specificity, and sensitivity of 63% and 30% respectively. IgG antibody titers generally remained at a constant level for more than 50 days, whereas IgA and IgM antibody titers declined more rapidly to normal values within 30 to 50 days after onset of clinical symptoms. Detection of *Campylobacter jejuni* specific IgA antibodies in a single serum sample provided the most useful assay for serological diagnosis of *Campylobacter jejuni* enteritis. The presence of *Campylobacter jejuni* specific IgM antibodies was the sole diagnostic criterion in three cases. Serological diagnosis of *Campylobacter jejuni* enteritis should therefore include both IgA and IgM antibody determination." (Authors' abstract)

191 Hernandez F, Caballero M, Rivera P, Hird D. Mannose-resistant hemagglutination, enzyme-linked immunosorbent assay, and immune electron microscopy for detection of K99 fimbrial antigen in *Escherichia coli* from calves. J Clin Microbiol 1989 Sep;27(9):2123-4

192 Hermann JE, Perron-Henry DM, Blacklow NR. Antigen detection with monoclonal antibodies for the diagnosis of adenovirus gastroenteritis. J Infect Dis 1987 Jun;155(6):1167-71

"A monoclonal antibody-based enzyme immunoassay (EIA) was developed for direct detection of enteric adenoviruses in stool specimens from individuals with gastroenteritis. Tests specific for each of the enteric adenoviruses, adenovirus type 40 (Ad 40) and type 41 (Ad 41), were designed. The sensitivity of the assay was determined by comparing the results of the EIA with isolation of virus in Graham 293 cells from stools that contained particles having adenovirus morphology. The standard for specificity was analysis of adenovirus genome profiles after digestion with *Sma*I endonuclease. The sensitivity was 95.8% (23 of 24) for Ad 40 and 97.1% (34 of 35) for Ad 41. The specificity was 95.7% (45 of 47) and 97.2% (35 of 36), respectively. The two type-specific monoclonal antibodies could be mixed in an EIA for identification of enteric adenoviruses in stools without loss of reactivity in either type. The EIA permits rapid diagnosis and type-specific identification of enteric adenoviruses in gastroenteritis. (Authors' abstract)

193 Hermann JE, Nowak NA, Blacklow NR. Detection of Norwalk virus in stools by enzyme immunoassay. J Med Virol 1985 Oct;17(2):127-33

"The development of a solid-phase microtiter enzyme immunoassay (EIA) for detection of Norwalk virus antigen in stool samples is described. The EIA was compared with a previously developed radioimmunoassay (RIA) for detection of Norwalk virus antigen in stools obtained from 30 volunteers who received Norwalk virus. The EIA detected viral antigen in stools from 17 of the volunteers and the RIA detected viral antigen in 15. Seroconversion was a more sensitive indicator of infection in some patients. However, two samples from volunteers who were clinically ill but did not show seroconversion to Norwalk virus were positive for Norwalk virus antigen by both immunoassays. This indicates that antigen detection may be important for use in epidemiological studies. Neither of the immunoassays gave positive reactions for stools known to contain enteric

adenovirus, rotavirus, or Hawaii virus, or in stools from patients with acute diarrhea of unknown cause. The stability of the EIA reagents and ease of use should provide a means for more extensive testing for Norwalk virus in outbreaks of gastroenteritis." (Authors' abstract)

194 Herrmann JE, Nowak NA, Perron-Henry DM, Hudson RW, Cubitt WD, Blacklow NR. Diagnosis of astrovirus gastroenteritis by antigen detection with monoclonal antibodies. *J Infect Dis* 1990 Feb;161(2):226-9

"An enzyme-linked immunosorbent assay (ELISA), based on monoclonal antibodies to the astrovirus group antigen, was designed for the detection of astroviruses in stools of patients with gastroenteritis. Compared to immune electron microscopy used as the standard test, the sensitivity of the astrovirus ELISA was 91% (31/34) and the specificity was 96% (54/56). All five of the known astrovirus serotypes could be detected in 16 samples on which serotyping was done. In tests on 155 stools containing other enteric viruses, including adenoviruses, rotaviruses, caliciviruses, Hawaii virus, Snow Mountain virus, and Norwalk virus (30, 20, 70, 24, 4, and 7 samples, respectively), only 3 were positive in the astrovirus ELISA. The combined specificity for all astrovirus immune electron microscopy-negative samples was 98% (206/211). The results demonstrate that the new ELISA provides a sensitive and specific means for the diagnosis of astrovirus gastroenteritis." (Authors' abstract)

195 Herrmann JE, Blacklow NR, Perron DM, Cukor G, Krause PJ, Hyams JS, Barrett HJ, Ogra PL. Enzyme immunoassay with monoclonal antibodies for the detection of rotavirus in stool specimens. *J Infect Dis* 1985 Oct;152(4):830-2

196 Heyworth MF, Kung JE, Caplin AB. Enzyme-linked immunosorbent assay for *Giardia*-specific IgA in mouse intestinal secretions. *Parasite Immunol* 1988 Nov;10(6):713-7

"We describe an ELISA for trophozoite-specific IgA in the intestinal secretions of mice infected with *Giardia muris*. Using this method, trophozoite-specific IgA was demonstrated in intestinal secretions of *Giardia*-infected immunocompetent BALB/c mice. Such antibody was undetectable in the intestinal secretions of *Giardia*-infected athymic (nude) mice. Immunocompetent BALB/c mice are able to clear *G. muris* infection whereas nude mice are not. The study provides evidence that the chronicity of *G. muris* infection in nude mice results from lack of intestinal trophozoite-specific IgA in these animals. By means of the ELISA, trophozoite-specific IgA was demonstrated in intestinal secretions from immunocompetent mice in the absence of protease inhibitors." (Authors' abstract)

197 Hirschl AM, Rathbone BJ, Wyatt JI, Berger J, Rotter ML. Comparison of ELISA antigen preparations alone or in combination for serodiagnosing *Helicobacter pylori* infections. *J Clin Pathol* 1990 Jun;43(6):511-3

"The immunoglobulin G antibody response to *Helicobacter pylori* was assessed in 78 patients with non-ulcer dyspepsia using five different antigen preparations. All patients were endoscoped and biopsied. The *H. pylori* state was determined histologically on at least two endoscopic biopsy specimens using a modified Giemsa stain. The ultracentrifuged cell sonicate, acid glycine extract, and 120 kilodalton protein antigens were specific in diagnosing infection (95-98%), but had only moderate sensitivity (70-84%). By mixing either of the two complex antigens with the 120 kilodalton protein, the sensitivity of the test was increased to 97% without affecting the high specificity. The combination of ultracentrifuged sonicate or acid glycine extract with the 120 kilodalton protein therefore seems to be superior to the individual antigen preparations and is particularly suitable for the serodiagnosis of *H. pylori* infection." (Authors' abstract)

198 Hock GM, Foon KLP, Chuen HL, Choo NG, Singh M. A comparison of the

ELISA with other serological tests in the serodiagnosis of amoebiasis. Southeast Asian J Trop Med Public Health 1989 Sep;20(3):399-405

"The sera of 78 patients with invasive amoebiasis were tested for anti-amoebic antibodies by the techniques of enzyme-linked immunosorbent assay (ELISA), indirect haemagglutination (IHA), indirect fluorescent antibody (IFA) and counterimmunoelectrophoresis (CIEP). Results showed that the ELISA compared favourably with IHA and IFA tests in terms of sensitivity and specificity. ELISA, IHA and IFA detected 97.4%, 96.2% and 98.7% of the patients respectively. CIEP was the least sensitive of the 4 serological methods with a sensitivity of 88.5%. The advantages and disadvantages of the 4 serodiagnostic procedures are discussed." (Authors' abstract)

199 Hock GM, Foon KLP, Chuen HL, Choo NG, Singh M. Determination of anti-amoebic IgG, IgM, IgA and IgG subclasses in sera from patients with amoebiasis by enzyme-linked immunosorbent assay. Southeast Asian J Trop Med Public Health 1989 Sep;20(3):407-14

"The HK-9 strain of *Entamoeba histolytica* was cultured axenically. A crude extract of the trophozoites was used as antigen in the enzyme-linked immunosorbent assay (ELISA) for the determination of anti-amoebic IgG, IgM, IgA and IgG subclasses in sera of patients with *E. histolytica* infection. Sera from amoebiasis patients had significantly higher mean ELISA values for all classes and subclasses of immunoglobulins examined compared with healthy controls. The ELISA for IgG, IgM and IgA was positive for 76 (97.4%), 34 (43.6%) and 62 (79.5%) respectively of 78 amoebiasis sera. Analysis of IgG subclasses by ELISA showed that 76.7%, 25%, 33.3% and 66.6% of the patients were positive for anti-amoebic IgG1, IgG2, IgG3 and IgG4 respectively. IgG1 and IgG4 were the dominant IgG subclasses involved in amoebiasis. The role of anti-amoebic antibodies in amoebiasis is discussed." (Authors' abstract)

200 Hoffman SL, Planigan TP, Klauke D, Leksana B, Rockhill RC, Punjabi NH, Pulungsih SP, Sutomo A, Moechar MA. The Widal slide agglutination test, a valuable rapid diagnostic test in typhoid fever patients at the Infectious Diseases Hospital of Jakarta. Am J Epidemiol 1986 May;123(5):869-75

"The Widal slide agglutination test was evaluated as a rapid diagnostic test in typhoid fever patients at the Infectious Diseases Hospital, Jakarta, Indonesia from 1980-1982. The results of the test can be available within 45 minutes of patient admission. The study showed that, among 229 patients with *Salmonella typhi*-positive typhoid fever and 179 control fever patients, when the Widal O antibody titer was $\geq 1:20$ the sensitivity was 53%, the specificity 98%, the positive predictive value 96%, and the negative predictive value 68%. A negative Widal test (O antibody titer $< 1:20$) does not provide useful information, but when the O antibody titer is $\geq 1:20$ the clinician at the Infectious Diseases Hospital of Jakarta can be 96% certain that the patient has typhoid fever." (Authors' abstract)

201 Hofmann M, Wyler R. Enzyme-linked immunosorbent assay for the detection of porcine epidemic diarrhea coronavirus antibodies in swine sera. Vet Microbiol 1990 Jan;21(3):263-73

"An enzyme-linked immunosorbent assay (ELISA) for detecting serum antibodies to the porcine epidemic diarrhea coronavirus (PEDV) was established by using cell culture-grown PEDV as antigen for coating. Ultracentrifugation through 20 and 45% (w/w) sucrose cushions proved to be the best antigen purification method. Examination of 1024 swine sera showed a high specificity and a greater sensitivity of the ELISA, when compared with indirect immunofluorescence. Reference sera with high antibody titers to PEDV originated from two pigs experimentally infected with PEDV. Three different antigen purification methods and the advantages of the ELISA compared with an immunofluorescence test are discussed." (MEDLINE)

202 Hohdatsu T, Elguchi Y, Ide S, Baba K, Yamagishi H, Kume T, Matumoto M. Evaluation of an enzyme-linked immunosorbent assay for the detection of transmissible gastroenteritis virus antibodies. *Vet Microbiol* 1987 Jan;13(1):93-7

"An enzyme-linked immunosorbent assay (ELISA) using a detergent-solubilized antigen of purified virus was developed for detection of antibody against porcine transmissible gastroenteritis (TGE) virus in swine serum. The ELISA demonstrated antibody responses in pigs immunized intramuscularly with the attenuated TO-163 strain of TGE virus and in pigs orally infected with the virulent Shizuoka strain of the virus. The results of the ELISA were well correlated with those of the neutralization test. These results indicate the usefulness of the ELISA as a serological tool for TGE virus antibody." (MEDLINE)

203 Honda T, Sato M, Nishimura T, Higashitsutsumi M, Fukai K, Miwatani T. Demonstration of cholera toxin-related factor in cultures of *Aeromonas* species by enzyme-linked immunosorbent assay. *Infect Immun* 1985 Oct;50(1):322-3

"The production of toxins by *Aeromonas* species was examined by the suckling mouse test, the hemolysin test, and the enzyme-linked immunosorbent assay with anticholera enterotoxin. A factor that was immunologically related to cholera enterotoxin was produced by 5 of 14 strains of *Aeromonas hydrophila* and 4 of 15 strains of *Aeromonas sobria*. Analysis by these assays and by a test for heat stability suggested that the factor differed from hemolysin and from toxin that was active in the suckling mouse test." (Authors' abstract)

204 Honda T, Yoh M, Kongmuang U, Miwatani T. Enzyme-linked immunosorbent assays for detection of thermostable direct hemolysin of *Vibrio parahaemolyticus*. *J Clin Microbiol* 1985 Sep;22(3):383-6

"Several systems for enzyme-linked immunosorbent assay (ELISA) of thermostable direct hemolysin (TDH) of *Vibrio parahaemolyticus* were tested, and single-antibody sandwich ELISA systems gave satisfactory results. ELISA was able to detect as little as several nanograms of purified TDH per milliliter. The method of De Jong (*J. Clin. Microbiol.* 17:928-930, 1983) and the glutaraldehyde method were successful for preparing conjugates of alkaline phosphatase and anti-TDH antibody. TDH in fluids in intestinal loops of experimental animals challenged with living *V. parahaemolyticus* was accurately detectable by ELISA." (Authors' abstract)

205 Honda T, Wetprasit N, Arita M, Miwatani T. Production and characterization of monoclonal antibodies to a pilus colonization factor (colonization factor antigen III) of human enterotoxigenic *Escherichia coli*. *Infect Immun* 1989 Nov;57(11):3452-7

"Three monoclonal antibodies (MAbs) to a pilus colonization factor (colonization factor antigen III [CFA/III]) of human enterotoxigenic *Escherichia coli* (ETEC) were developed and characterized. All of the MAbs isolated belonged to the immunoglobulin G2a subclass. The specificity of these MAbs for CFA/III pili was demonstrated by the immunogold-labeling technique. The presence of more than one epitope in CFA/III pili was suggested. One of the three MAbs appears to recognize a polymeric conformational epitope(s) of CFA/III. CFA/III antigenicity distinct from that of other pilus colonization factors of ETEC was demonstrated by both a bacterial agglutination test and a sandwich enzyme-linked immunosorbent assay using the MAbs. Of the 100 strains of ETEC isolated from persons with traveler's diarrhea, 8% were found to carry CFA/III pili. Two enzyme-linked immunosorbent assay systems which could detect as little as several or 50 ng of CFA/III per ml were developed." (Authors' abstract)

206 Honma H, Ushijima H, Takagi M, Kitamiura T. Evaluation of a new enzyme immunoassay (TESTPACK ROTAVIRUS) for diagnosis of viral gastroenteritis. *Kansenshogaku Zasshi* 1990 Feb;64(2):174-8

"TESTPACK ROTAVIRUS, a simple 10 min enzyme immunoassay, was compared with reversed passive hemagglutination assay (RPHA) and polyacrylamide gel electrophoresis of virus RNA (RNA-PAGE) for the detection of rotaviral antigens in feces from 104 diarrheal children. The results of TESTPACK ROTAVIRUS were identical with those of RNA-PAGE. Discordant results of RPHA were further examined by latex agglutination and electron microscopy and were finally decided to be negative. Pararotaviruses, adenoviruses and small round viruses were negative in the TESTPACK ROTAVIRUS." (MEDLINE)

207 Hopley JF, Doane FW. Development of a sensitive protein A-gold immunoelectron microscopy method for detecting viral antigens in fluid specimens. *J Virol Methods* 1985 Oct;12(1-2):135-47

"Protein A-colloidal gold immunoelectron microscopy (PAG IEM) has been employed to specifically detect rotavirus and enterovirus antigen in negatively stained fluid specimens. Unlike other IEM methods, PAG IEM can detect not only viral antigen associated with morphologically recognizable particles but also viral antigens of unrecognizable ultrastructure. This rapid and sensitive immunoassay was found to be applicable to virus-infected stool specimens as well as partially purified virus preparations. The sensitivity of viral antigen detection by PAG IEM was 2- to 40-fold greater than direct IEM and 200- to 1,000-fold greater than direct electron microscopy. In addition, PAG IEM appears to offer a more reliable and sensitive alternative to standard IEM for detection and quantitation of viral antibody." (MEDLINE)

208 Hornsleth A, Aaen K, Gundestrup M. Detection of respiratory syncytial virus and rotavirus by enhanced chemiluminescence enzyme-linked immunosorbent assay. *J Clin Microbiol* 1988 Apr;26(4):630-5

209 Horstedt P, Danielsson A, Nyhlin H, Stenling R, Suhr O. Adhesion of bacteria to the human small-intestinal mucosa: a scanning electron microscopic study. *Scand J Gastroenterol* 1989 Sep;24(7):877-85

"During the period 1 January 1980 to 30 June 1986, a total of 543 small-intestinal biopsy specimens from adults with gastrointestinal symptoms were available for routine analysis with correlated scanning electron microscopy and light microscopy. Adhesion of microorganisms was found in 77 biopsy specimens. Microorganisms in 64 specimens were classified as bacteria, in 10 as microfungi and in 4 as protozoa, including 1 specimen with both bacteria and microfungi. The structural types of bacteria found were morphologically cocciform, 8; short rod-shaped, 14; and long rod-shaped, 43. One specimen demonstrated adhesion of two structural types of bacteria. Bacteria were found in specimens from all age groups in roughly equal frequency. There was no difference in villus structure when comparing specimens from the groups with and without adhering bacteria, whereas ultrastructural alteration - that is, thinning of glycocalyx layer - was significantly more frequent in the group with bacteria. Moreover, within the group of specimens with bacteria the presence of long rod-shaped bacteria was associated with both damage of villus structure and deviation of cell surface ultrastructure. An increased amount of neutrophil granulocytes as an indicator of acute inflammation was found in 6 of 51 specimens with bacterial adhesion but in none of a matched reference material. In contrast, the amount of plasma cells and lymphocytes in the lamina propria and the amount of intraepithelial lymphocytes did not differ." (Authors' abstract)

210 Hostacka A, Majtan V. Our experiences with isolation of heat-labile enterotoxin from *Escherichia coli*. *J Hyg Epidemiol Microbiol Immunol* 1989;33(1):63-70

211 Howard CJ, Clarke MC, Brownlie J. An enzyme-linked immunosorbent assay (ELISA) for the detection of antibodies to bovine viral diarrhoea virus (BVDV) in cattle sera. *Vet Microbiol* 1985 Jun;10(4):359-69

"A microtitre ELISA has been established for the quantitation of antibodies to bovine viral

diarrhoea virus (BVDV). Single dilutions of sera were assayed and units of antibody were calculated from a standard curve. In order to detect the maximum number of responding animals both IgG1 and IgG2 antibody should be assayed, although detection of IgG1 alone was nearly as effective. The ELISA was as sensitive as the virus neutralization test for detection of antibody; comparison of an ELISA that detected IgG1 plus IgG2 antibody to BVDV with the virus neutralization test gave a correlation coefficient (r) of 0.89 ($p < 0.001$ for 95 compared sera). Although similar amounts of IgG1 and IgG2 antibodies were present in sera from both experimentally- and naturally-infected cattle, antibody to BVDV in colostrum and in the sera from young calves was predominantly IgG1. The number of adult cows with antibody was 40 out of 41 while 36 of 44 calves reared in a beef unit were found to have produced antibody by the time they were 31.5 weeks old, an indication of the high prevalence of BVDV in the cattle population." (MEDLINE)

212 Hudson RW, Herrmann JE, Blacklow NR. Plaque quantitation and virus neutralization assays for human astroviruses. Arch Virol 1989;108(1-2):33-8

"Astroviruses, 28 nm-diameter, RNA-containing viruses which have been implicated in gastroenteritis can be cultivated in cell cultures containing trypsin, but do not show distinguishable cytopathic effects. However, with the 5 known astrovirus serotypes which we have been able to cultivate, 3 (types 1, 2, and 5) formed well-defined plaques in LLCMK2 cell cultures under an agar overlay containing trypsin. A virus neutralization assay based on plaque reduction was applied to these 3 serotypes. It was found that rabbit antisera prepared against individual serotypes neutralized virus type-specifically, and no cross-neutralization titers were obtained with any of the antisera to the 5 astrovirus serotypes. The type-specific neutralization observed agreed with the specificities seen by immunofluorescence (IF), whereas ELISA tests with the same antisera show cross-reactivity among all 5 serotypes. There was no virus neutralization detected with astrovirus monoclonal antibodies which were reactive with 5 serotypes by ELISA and IF. The results we have obtained permit quantitative techniques to be applied to epidemiological and biological studies of the human astroviruses." (MEDLINE)

213 Hunt ALC, Goldsmid JM. An investigation of culture media for the isolation of shigellae. Med Lab Sci 1990 Jul;47(3):151-7

"In this study we have investigated the efficiency of presently available culture media for the isolation of shigellae. XLD was found to be the medium of choice, combined with both less and more selective culture media (e.g. MacConkey, and Hektoen or DCA). By using these media in combination it was found that not only were shigellae isolated more often and efficiently, but fewer problems were encountered in isolating other *Enterobacteriaceae*." (Authors' abstract)

214 Husson MO, Mielcarek C, Izard D, Leclerc H. Alkaline phosphatase capture test for the rapid identification of *Escherichia coli* and *Shigella* species based on a specific monoclonal antibody. J Clin Microbiol 1989 Jul;27(7):1518-21

215 Hyera JM, Liess B, Frey HR. A direct neutralizing peroxidase-linked antibody assay for detection and titration of antibodies to bovine viral diarrhoea virus. Zentralbl Veterinarmed [B] 1987 Apr;34(3):227-39

216 Ianculescu M, McCapes RH, Bankowski RA, Kelly BJ, Ghazikhanian GY. Hemorrhagic enteritis of turkeys in California: serologic study of hemorrhagic enteritis virus antibody with an enzyme-linked immunosorbent assay. Avian Dis 1985 Apr-Jun;29(2):358-63

217 Iankina NF, Semenova GB. [Isolation of monospecific *Salmonella* anti-O-4 antibodies by affinity chromatography and their use in immunoenzyme reactions] Zh Mikrobiol Epidemiol Immunobiol 1987 Aug;34(8):79-82

"An immunosorbent with *Salmonella typhimurium* 1021 O-antigen has been obtained on the basis of polymer-modified porous glass. Binding has been effected due to the presence of NH₂-groups on the carrier and aldehyde groupings on O-antigen previously oxidized with sodium periodate. Specific anti-O-4 antibodies have been isolated with the use of this immunosorbent, and on the basis of these antibodies peroxidase conjugates have been obtained. This preparation has made it possible to establish the parameters of EIA for the detection of *Salmonella* O-4 antigens in the assay involving the use of the sandwich technique. The method has proved to be sufficiently simple, sensitive and specific. It is recommended for the detection of *Salmonella* O-4 antigen in biological fluids." (MEDLINE)

218 Ibrahim GF, Lyons MJ, Walker RA, Fleet GH. Immobilization of microorganisms for detection by solid-phase immunoassays. *J Clin Microbiol* 1985 Sep; 22(3):361-5

"Several cultures of gram-negative and gram-positive bacteria were successfully immobilized with titanous hydroxide. The immobilization efficiency for the microorganisms investigated in saline and broth media ranged from 80.2 to 99.9%. The immobilization of salmonellae was effective over a wide pH range. The presence of buffers, particularly phosphate buffer, drastically reduced the immobilization rate. However, buffers may be added to immunoassay systems after immobilization of microorganisms. The immobilization process involved only one step, i.e., shaking 100 ml of culture with 50 ml of titanous hydroxide suspension in polystyrene tubes for only 10 min. The immobilized cells were so tenaciously bound that vigorous agitation for 24 h did not result in cell dissociation. The nonspecific binding of 125I-labeled antibody from rabbits and 125I-labeled protein A by titanous hydroxide was inhibited in the presence of 2% gelatin and amounted to only 5.6 and 3.9%, respectively. We conclude that this immobilization procedure is a potentially powerful tool which could be utilized in solid-phase immunoassays concerned with the diagnosis of microorganisms." (Authors' abstract)

219 Ibrahim GF, Fleet GH, Lyons MJ, Walker RA. Immunological relationships between *Salmonella flagella* and their potential application for salmonellae detection by immunoassay. *Med Microbiol Immunol (Berl)* 1985;174(2):87-99

"Native flagella of ten *Salmonella* serotypes were shown to possess serotype-specific antigenic determinants as well as a multiple of common antigenic determinants, which varied in concentration. Each common antigenic determinant was shared by certain, but not all, serotypes. This has been demonstrated from agglutination and radioimmunometric assay (RIMA) results, using ten antisera raised in rabbits against purified polymeric flagellins from ten *Salmonella* serotypes as immunogens. The minimum detectable populations of salmonellae, as determined by RIMA varied considerably, due to variation in concentrations (and possibly types) of common antigenic determinants. However, the results demonstrated the feasibility of utilizing RIMA for the qualitative detection of salmonellae, using a mixture of only the ten antisera and 125I-labelled protein A as a general tracer. In this way, 77 different salmonellae were detected in less than 8 h after culturing in selective broth. The RIMA developed was specific for salmonellae and showed no cross-reactions with high populations of other members within the family *Enterobacteriaceae*." (MEDLINE)

220 Ibrahim GF, Lyons MJ, Walker RA, Fleet GH. Rapid detection of salmonellae by immunoassays with titanous hydroxide as the solid phase. *Appl Environ Microbiol* 1985 Sep;50(3):670-5

"Radioimmunometric and enzyme-immunometric assays were developed for the detection of salmonellae in pure and mixed cultures as well as in 59 food samples. The performances of titanous hydroxide suspension and microtiter plates as the solid phase for the immobilization of microorganisms were compared in these immunoassays. Detection of populations of *Salmonella* cells in pure culture, diluted with saline, was 4- to 10-fold more sensitive with the microtiter plates. However, with mixed culture of

Salmonella and other enterobacterial species, the detection sensitivity with titanous hydroxide was 100- to 160-fold more sensitive than with microtiter plates. Good correlation existed between results of a standard cultural method for the detection of salmonellae in foods and those obtained from radioimmunometric and enzyme-immunometric assays utilizing titanous hydroxide. However, a high incidence of false-positive and false-negative results with food samples occurred with the enzyme-immunometric assay utilizing microtiter plates. The results provided strong evidence for the merits of substituting titanous hydroxide for microtiter plates as the solid phase for the immobilization of salmonellae for their detection by immunoassays. The immunoassays were rapid and enabled the analysis of a large number of selective enrichment cultures of food samples for salmonellae within 8 h. (Authors' abstract)

221 Ibrahim OS, Sunderland D, Hart CA. Comparison of four methods for detection of rotavirus in faeces. *Trop Doct* 1990 Jan;20(1):30-2

"Faecal samples from 325 children with gastroenteritis and 23 children without gastroenteritis were examined for the presence of human rotavirus (HRV) using four different methods. Using the WHO-ELISA, HRV was found in the stools of 98 (30%) symptomatic and 2 (9%) asymptomatic children. A latex particle agglutination test had the highest sensitivity (92%) but the lowest specificity (96%). Both electron microscopy and polyacrylamide gel electrophoresis of HRV RNA (RNA-PAGE) were highly specific (100%) but of lower sensitivity (73% and 84% respectively). Of the four methods tested latex particle agglutination is the simplest and since it requires little extra equipment is ideally suited for bedside tests in tropical countries. It is, however, not cheap. An alternative is to use RNA-PAGE which will require some equipment and a power supply but which is relatively cheap and will also provide epidemiological data." (Authors' abstract)

222 Isaac-Renton JL, Fung CPJ, Lochan A. Evaluation of a tangential-flow multiple-filter technique for detection of *Giardia lamblia* cysts in water. *Appl Environ Microbiol* 1986 Aug;52(2):400-2

223 Isaac-Renton JL, Black WA, Mathias RG, Proctor EM, Sherlock CH. *Giardiasis* in a group of travellers - attempted use of a serological test. *Can J Public Health* 1986 Mar-Apr;77(2):86-8

224 Iwanaga M, Yamamoto K. New medium for the production of cholera toxin by *Vibrio cholerae* 01 biotype El Tor. *J Clin Microbiol* 1985 Sep;22(3):405-8

A new medium for stimulating *in vitro* cholera toxin (CT) production by *Vibrio cholerae* 01 is described. ElTor biotype and CT productions in this new medium and in syncase medium are compared. One hundred and thirteen strains of ElTor vibrios, isolated after 1980 in Kenya, Philippines and Taiwan, were used. The medium, in which bacto-peptone was added, had an electrolyte composition similar to that of the cholera stool. Strain A66 produced 125 ng of CT per ml. The concentration of bacto-peptone had a minimal effect on the CT production which increased when yeast extract was added up to 0.4 to 0.6%. Sodium bicarbonate in the range 0.2-0.4% was essential for enhancement of CT production. The pH of the medium was set at 7.6 with a minimum variation. The new medium was designated AKI medium which is composed of bacto-peptone 1.5% yeast extract 0.4%, sodium bicarbonate 0.3%, and sodium chloride 0.5% with pH 7.5±0.1 allowed the production of more CT than syncase medium using several strains of ElTor vibrios at the optimum temperature of 37°C. One such organism may produce as much as 1,000 times more CT in AKI medium than what it can in the syncase medium. This modified culture method can help in purifying enough CT produced by ElTor vibrios to permit the analysis of the toxins.

225 Iwen PC, Booth SJ, Woods GL. Comparison of media for screening of diarrheal stools for the recovery of *Clostridium difficile*. *J Clin Microbiol* 1989 Sep;27(9):2105-6

226 Jackson MP. Detection of Shiga toxin-producing *Shigella dysenteriae* type 1 and *Escherichia coli* by using polymerase chain reaction with incorporation of digoxigenin-11-dUTP. *J Clin Microbiol* 1991 Sep;29(9):1910-4

"A technique has been developed for the detection of Shiga toxin- and Shiga-like type I (Sht/SLT-I)-producing *Shigella dysenteriae* type 1 and *Escherichia coli* by using the polymerase chain reaction with the incorporation of digoxigenin-11-dUTP. Target DNA liberated from whole cells was amplified, using primer pairs homologous to the A-subunit genes of Sht/SLT-I. The TTP analog digoxigenin-11-dUTP was incorporated into the reaction mixture, permitting nonradioactive labeling of the amplified DNA. The labeled polymerase chain reaction products were hybridized to specific gene sequences immobilized on a nitrocellulose membrane and detected by using an alkaline phosphatase-conjugated antibody to digoxigenin and the enzyme substrates. Toxin-producing strains of *E. coli* and *S. dysenteriae* type 1 were identified as colored spots on the membrane. Because this technique does not require DNA purification, gel electrophoresis, or radioactive DNA probes, it is suitable for the clinical detection of Sht/SLT-I-producing strains of *S. dysenteriae* type 1 and *E. coli*." (Authors' abstract)

227 Jackson SG, Yip-Chuck DA, Brodsky MH. A double antibody sandwich enzyme-immunoassay for *Clostridium perfringens* type A enterotoxin detection in stool specimens. *J Immunol Methods* 1985 Oct 24;83(1):141-50

"A double antibody sandwich enzyme-immunoassay has been developed for detection of *Clostridium perfringens* enterotoxin. Anti-enterotoxin immunoglobulin G-alkaline phosphatase conjugates were prepared using a rapid minicolumn procedure. The assay can achieve a sensitivity of ≥ 1 ng/ml with purified enterotoxin. Sensitivity for detection of cases of *C. perfringens* enteritis in a *C. perfringens* outbreak (86 individuals tested) was between 85.7 and 98.0 per cent depending upon stringency of criteria for defining positive cases. Specificity of the assay was demonstrated by the lack of positive results in 53 individuals involved in a gastroenteritis outbreak of unknown etiology." (Authors' abstract)

228 Jackson SG, Yip-Chuck DA, Brodsky MH. Evaluation of the diagnostic application of an enzyme immunoassay for *Clostridium perfringens* type A enterotoxin. *Appl Environ Microbiol* 1986 Oct;52(4):969-70

"The diagnostic application of an enzyme immunoassay for *Clostridium perfringens* type A enterotoxin was evaluated. Test results from 100 individuals associated with *C. perfringens* gastroenteritis outbreaks and 111 control individuals were included. The assay sensitivity was 93.7%, and the assay specificity was 98.7%." (Authors' abstract)

229 Jain AK, Babu R, Tantry BV, Sen PC, Gupta JP. Enzyme linked immunosorbent assay (ELISA) in the diagnosis of amoebiasis. *J Assoc Physicians India* 1986 Feb;34(2):129-31

230 Jain U, Patel MT, Desai PK, Kalraj P. Development of simple and stable ELISA for detection of copro antigen in intestinal amoebiasis. *Indian J Exp Biol* 1990 Jul;28(7):671-5

"A simple, sensitive and stable ELISA (enzyme linked immunosorbent assay) was developed using rabbit antibody to fractionated *Entamoeba histolytica* antigen for the detection of copro antigen in the faeces of individuals with intestinal amoebiasis. In this test none of the healthy individuals, all trophozoite positive, 40% cyst passers and 6% individuals living in endemic area showed the presence of copro antigen. ELISA using polyclonal rabbit antibody could detect 1-5 trophozoites/well and 20-50 ng protein per well of NIH-200 strain of *E. histolytica* and the sensitivity of the test was comparable with that using monoclonal antibody. Cross reaction was observed only with *E. invadens* when faeces having other parasites were screened. The reagents of ELISA were

stabilized and found to be stable for more than 6 months when stored at 4°C." (Authors' abstract)

231 Jensen A. Evaluation of two commercial kits for rapid detection of human rotavirus in feces: Rotalex, a latex agglutination test and Rotavirus ELISA Kit. *Acta Pathol Microbiol Immunol Scand [B]* 1985 Apr;93(2):159-60

"96 fecal specimens from children with symptoms of gastroenteritis were tested with a latex agglutination test (Rotalex, Orion Diagnostica, Finland) and an enzyme-linked immunosorbent test (Rotavirus ELISA Kit, Dakopatts, Copenhagen, Denmark). Using the ELISA method as reference, the latex agglutination (LX) came out with three non-specific and two false negative results. The LX test was found to be a rapid, practical and non-expensive choice for a laboratory examining few samples daily." (Authors' abstract)

232 Jerse AE, Martin WC, Galen JE, Kaper JB. Oligonucleotide probe for detection of the enteropathogenic *Escherichia coli* (EPEC) adherence factor of localized adherent EPEC. *J Clin Microbiol* 1990 Dec;28(12):2842-4

233 Johansson ME, Uhnoo I, Svensson L, Pettersson C-A, Wadell G. Enzyme-linked immunosorbent assay for detection of enteric adenovirus 41. *J Med Virol* 1985 Sep;17(1):19-27

"An enzyme-linked immunosorbent assay (ELISA) for direct detection of enteric adenovirus 41 (Ad41) in stool specimens was developed and compared with an Ad40-specific ELISA described previously [Johansson et al, 1980]. Rabbit antiserum to Ad41 was obtained by immunization with purified virions. To eliminate genus-specific reactivity the serum was passed through an immunosorbent column containing soluble adenovirus components of members of subgenera A to E. The anti-Ad41 serum still displayed high reactivity against Ad40 and had to be immunoabsorbed with soluble virus components of Ad40 to be rendered type-specific. The absorbed antiserum was used in an indirect ELISA and proved to be specific for Ad41. No heterotypic reactivity against Ad40 or Ad1 through Ad35 was found. The Ad41-specific ELISA proved to be of equal sensitivity to electron microscopy. The type-specific ELISAs for Ad40 and Ad41 were evaluated by testing 76 stool specimens containing enteric adenoviruses originating from England and Scandinavia. All specimens could be typed - 41 (54%) as Ad40 and 35 (46%) as Ad41. These results were confirmed by DNA restriction site analysis. The type-specific ELISA proved to be a specific, sensitive, and a rapid technique for detection of Ad41 and allowed clear-cut discrimination from Ad40 in clinical specimens." (Authors' abstract)

234 Johnson LL, McFarland LV, Dearing P, Ralsys V, Schoenknecht FD. Identification of *Clostridium difficile* in stool specimens by culture-enhanced gas-liquid chromatography. *J Clin Microbiol* 1989 Oct;27(10):2218-21

235 Johnson TM, Skingle J, Gillett AP. Detection of rotavirus by latex agglutination [letter]. *J Clin Pathol* 1985 Dec;38(12):1403-4

236 Julkunen I, Savolainen J, Hautanen A, Hovi T. Detection of rotavirus in faecal specimens by enzyme immunoassay, latex agglutination and electron microscopy. *Scand J Infect Dis* 1985;17(3):245-9

"Faecal specimens from 570 patients with gastroenteritis were studied for rotaviruses by enzyme immunoassay (EIA), electron microscopy (EM) and latex agglutination test (LX). Specimens from 127 patients were positive and 379 were negative with all 3 methods. 84 (11%) specimens gave contradictory results in the tests. EIA gave significantly more positive results than EM (168 vs 145). 30 EM-negative and EIA-positive specimens were positive in a confirmatory test. LX was positive in 161 specimens and no significant differences to EM or EIA were observed. There were 16 (2.8%) LX-positive samples that

were negative both in EM and EIA. The positivity of these samples could not be confirmed and 15 of them were only slightly positive. Our results can be concluded: (1) Enzyme immunoassay may be more sensitive than electron microscopy but requires a confirmatory test and is tedious when only a few specimens per day are to be examined; (2) LX seems to be suitable for primary screening of faecal specimens for rotavirus; for definitely positive results the test is reliable." (MEDLINE)

237 Juntti N, Larsson B, Fossum C. The use of monoclonal antibodies in enzyme linked immunosorbent assays for detection of antibodies to bovine viral diarrhoea virus. *Zentralbl Veterinarmed [B]* 1987 Jul;34(5):356-63

238 Jure MN, Morse SS, Stark DM. Serum albumin treatment to eliminate false-positive results with laboratory rodent specimens tested by Rotazyme enzyme-linked immunosorbent assay. *J Clin Microbiol* 1987 Nov;25(11):2239-40

"A high proportion of positive results was found in fecal specimens from mice and rats tested by Rotazyme II enzyme-linked immunosorbent assay, but the presence of virus could not be confirmed. Positive specimens and nonautoclaved rodent diet contained a substance that apparently attached nonspecifically to the antibody-coated beads used in the test and reacted directly with the substrate. Pretreatment of the beads with 0.1% bovine serum albumin eliminated this nonspecific activity." (Authors' abstract)

239 Justewicz DM, Magar R, Marsolais G, Lecomte J. Bovine viral diarrhoea virus-infected MDBK monolayer as antigen in enzyme-linked immunosorbent assay (ELISA) for the measurement of antibodies in bovine sera. *Vet Immunol Immunopathol* 1987 Apr;14(4):377-84

"A specific ELISA for the detection of IgG antibodies against bovine viral diarrhoea virus (BVDV) has been developed and was compared with the seroneutralization (SN) titers for a total of 60 bovine serum samples. A BVDV-infected Madin-Darby Bovine Kidney (MDBK) cell monolayer served as test antigen. The following results were obtained: a coefficient of correlation (r) of 0.63 ($p < 0.01$) between BVDV-ELISA and SN titer in BVDV-seroconverted animals and 0.71 for all sera, including that from BVDV vaccinated animals; a sensitivity of 92%, a specificity of 91%, a concordance of 92%; and a demonstration of a parallel increase in BVDV-ELISA and SN titers in animals with seroconversion. The BVDV-ELISA permits a more complete evaluation of the humoral immune response to BVDV." (MEDLINE)

240 Jyotsna, Misra PK, Saxena PN, Malik GK, Das SR. Enzyme-linked immunosorbent assay for serodiagnosis of amoebiasis in children. *Indian J Med Res* 1985 Feb;81(2):129-32

"Enzyme-linked immunosorbent assay (ELISA) has been evaluated for the serodiagnosis of amoebiasis in children using axenic *Entamoeba histolytica*. A titer of 1:320 was taken as the cut off limit. The seropositivity among patients of intestinal amoebiasis was 71.4 per cent, while it was 5.1, 3.4 and 2.5 per cent in patient with non-amebic parasitic infections and non-parasitic gastrointestinal disorders and controls respectively." (Authors' abstract)

241 Kaneko S, Maruyama T. Evaluation of enzyme immunoassay for the detection of pathogenic *Yersinia enterocolitica* and *Yersinia pseudotuberculosis* strains. *J Clin Microbiol* 1989 Apr;27(4):748-51

242 Kang SY, Nagaraja KV, Newman JA. Rapid coagglutination test for detection of rotaviruses in turkeys. *Avian Dis* 1985 Jul-Sep;29(3):640-8

"Avian rotaviruses present in fecal samples were readily detected using a staphylococcal protein-A coagglutination test on a white porcelain plate. Staphylococci, which

produced large amounts of protein-A, were coated with rabbit anti-avian rotavirus serum. The antibody-coated staphylococci were agglutinated specifically by rotavirus present in the fecal sample. The macroscopic agglutination reaction occurred within a few minutes. A total of 40 fecal samples were tested by the coagglutination test. The sensitivity and specificity of the coagglutination test were compared with those of electron microscopy, enzyme-linked immunosorbent assay, and tissue-culture virus-isolation methods. Of the 31 fecal samples positive for rotavirus on electron microscopy, 27 (87%) were positive on coagglutination test. Of the nine electron-microscopy-negative samples, seven (78%) were also negative on coagglutination test. It was concluded that the staphylococcal protein-A coagglutination test can be used as a simple, rapid screening test for avian rotavirus." (MEDLINE)

243 Kanjanahareutai S, Sarasombath S, Sirisinha S. Gel-filtration chromatography of *Salmonella* protein antigens and their implication in immunodiagnosis. Asian Pac J Allergy Immunol 1987 Dec;5(2):109-17

"Crude Barber protein (Bp) antigens were prepared from *Salmonella typhi*, *S. krefeld* and *S. derby* by an original method that has been described previously. These antigens were subjected to gel-filtration chromatography using Sephadex G-200. A sharp peak that eluted together with the void volume was thus separated from a broad second peak that eluted from the column at positions equivalent to 118,000 to 12,000 daltons. The proteins eluted in the latter peak were arbitrarily divided into 5 fractions and, together with the first peak, subjected to polyacrylamide gel electrophoresis and immunoprecipitation with both homologous and heterologous rabbit antisera. The extent of immunological cross reactivities was determined by enzyme-linked immunosorbent assay. The preliminary results obtained by this technique showed species-specific protein antigens to have molecular weights ranging between 36,000 and 68,000 daltons." (MEDLINE)

244 Kapperud G, Lassen J, Dommarsnes K, Kristiansen B-E, Caugant DA, Ask E, Jahkola M. Comparison of epidemiological marker methods for identification of *Salmonella typhimurium* isolates from an outbreak caused by contaminated chocolate. J Clin Microbiol 1989 Sep;27(9):2019-24

"Plasmid profile analysis, restriction endonuclease analysis, and multilocus enzyme electrophoresis were used in conjunction with serotyping, bacteriophage typing, and biochemical fingerprinting to trace epidemiologically related isolates of *Salmonella typhimurium* from an outbreak caused by contaminated chocolate products in Norway and Finland. To evaluate the efficiency of the epidemiological marker methods, isolates from the outbreak were compared with five groups of control isolates not known to be associated with the outbreak. Both plasmid profile analysis and phage typing provided further discrimination over that produced by serotyping and biochemical fingerprinting. Plasmid profile analysis and phage typing were equally reliable in differentiating the outbreak isolates from the epidemiologically unrelated controls and were significantly more effective than multilocus enzyme electrophoresis and restriction enzyme analysis of total DNA. The greatest differentiation was achieved when plasmid profile analysis and phage typing were combined to complement serotyping and biochemical fingerprinting. However, none of the methods employed, including restriction enzyme analysis of plasmid DNA, were able to distinguish the outbreak isolates from five isolates recovered in Norway and Finland over a period of years from dead passerine birds and a calf." (Authors' abstract)

245 Karunasagar I, Venugopal MN, Karunasagar I, Segar K. Evaluation of methods for enumeration of *Vibrio parahaemolyticus* from seafood. Appl Environ Microbiol 1986 Sep;52(3):583-5

The efficacies of different enrichment broths in recovering *Vibrio parahaemolyticus* inoculated into fish homogenates were examined. Enumeration of *V. parahaemolyticus* was done by direct plating on thiosulfate citrate bile salt sucrose (TCBS) agar and by

the five-tube most probable number (MPN) technique with SPB without or with 0.25 or 2.5 µg of polymyxin-B per ml. Recovery by the MPN technique was very poor in all the broths, while direct plating on TCBS agar yielded better results. A decrease in the enrichment time to 8 from 18 h did not accelerate their recovery. At concentrations exceeding 2.6 µg/ml, polymyxin was inhibitory to *V. parahaemolyticus*. The results point to the need to standardise the concentration of polymyxin to be used in broth when it is added as a filter-sterilised solution after the medium is autoclaved. Direct plating on TCBS is less labourious and the results are obtained a day earlier as compared with the MPN technique.

246 Kato N, Ou C-y, Kato H, Bartley SL, Brown VK, Dowell VR, Jr., Ueno K. Identification of toxigenic *Clostridium difficile* by the polymerase chain reaction. *J Clin Microbiol* 1991 Jan;29(1):33-7

247 Katz JB, Hanson SK. Competitive and blocking enzyme-linked immunoassay for detection of fetal bovine serum antibodies to bovine viral diarrhoea virus. *J Virol Methods* 1987 Feb;15(3):167-75

"A competitive blocking enzyme-linked immunoassay (CELIA) was developed to detect bovine viral diarrhoea virus (BVDV) antibodies in undiluted fetal bovine serum (FBS). The CELIA was based on competition of serum BVDV antibodies with biotin-labelled anti-BVDV immunoglobulins (Ig) for a limited quantity of solid-phase BVDV antigen. Antigen preparation was simple, FBS could be tested undiluted, and detergent-containing washes were unnecessary. A series of dilutions of postnatal bovine BVDV antiserum prepared in FBS and a set of 147 undiluted abattoir FBS samples were tested by both CELIA and serum neutralization tests (SNT). CELIA results on both sets of specimens correlated positively with SNT titers ($r = 0.99$ and $r = 0.85$). Relative to the SNT, CELIA sensitivity was 100%; specificity was 76%. CELIA detected a level of BVDV antibody below the 1:2-titer threshold detectable with the SNT. Advantages, limitations, and theoretical differences between the CELIA and SNT are discussed. A similar comparison of CELIA with non-competitive enzyme-linked immunoassay approaches to BVDV serodiagnosis is made. It is concluded that the CELIA is valuable in selecting only BVDV-seronegative FBS for use in virologic cell culture media." (MEDLINE)

248 Katz JB, Ludemann L, Pemberton J, Schmerr MJ. Detection of bovine virus diarrhoea virus in cell culture using an immunoperoxidase technique. *Vet Microbiol* 1987 Feb;13(2):153-7

"An indirect immunoperoxidase staining technique was developed for identifying cell cultures infected with bovine virus diarrhoea virus. Infected cell monolayers stained intensely while uninfected monolayers remained colorless. Immunoperoxidase staining was as sensitive as direct immunofluorescence in detecting endpoint dilutions of virus suspensions. Using the immunoperoxidase technique, infected monolayers were detectable by macroscopic, as well as microscopic, observation." (MEDLINE)

249 Kazemi M, Finkelstein RA. Checkerboard immunoblotting (CBIB): an efficient, rapid, and sensitive method of assaying multiple antigen/antibody cross-reactivities. *J Immunol Methods* 1990 Mar 27;128(1):143-6

"A simple technique, checkerboard immunoblotting (CBIB), is described, which facilitates the examination of multiple antigen/antibody interactions, conveniently and reproducibly, using minimal amounts of reactants. Antigens, immobilized on a solid-phase membrane in parallel lanes, are allowed to react with primary antibodies, applied in lanes perpendicular to the antigens, and the reactions are developed with appropriately labeled secondary antibody and substrate. Positive reactions, at the intersections of antigen/antibody lanes, are small squares, giving a checkerboard appearance to the blot. The results are easily read visually and presented in the form of a permanent record. CBIB has wide range of applications, including the screening of hybridomas for

monoclonal antibody production. With the cholera toxin (CT)-related antigens used, homologous reactions were markedly stronger than heterologous reactions." (MEDLINE)

250 Khan AH, Das SR. Rapid micro-IHA test with FACL-SRBCs in serodiagnosis of amoebiasis. *Indian J Med Res* 1986 Apr;83:377-9

"A rapid micro-Indirect hemagglutination (IHA) technique using glutaraldehyde fixed-antigen coated-lyophilized sheep red blood cells (FACL-SRBCs) has been described. The test is suitable for serodiagnosis of invasive amoebiasis using a drop of blood from patients. FACL-SRBCs when used after storage for 90 days at -20°C gave results similar to those with freshly prepared SRBCs. The test is rapid, stable, economical and can be used for field survey of amoebiasis." (Authors' abstract)

251 Khan AH, Ghosh PK, Malaviya B, Das SR. Simple & reproducible strip-ELISA technique for detection of anti-amoebic antibodies. *Indian J Med Res* 1987 Nov;86:582-7

"Conventional plate ELISA was converted into a simple strip-ELISA. In terms of operational ease and economy so that it may be performed by even paramedical technical personnel for diagnosis of amoebiasis. Polyvinyl chloride strips available locally were used as solid immunosorbent for antigen binding and subsequently processed for antibody quantitation of serum samples. The transformed assay was evaluated by screening different categories of clinically diagnosed amoebiasis patients in comparison with the conventional plate ELISA. A significant correlation was found between the two assays ($P < 0.05$)." (Authors' abstract)

252 Khan NZ, Shah SK, Akbar MS. Comparative efficacy of blood, urine, stool and bone-marrow cultures for isolation of *Salmonella typhi* in typhoid fever. *J Bangladesh Coll Phys Surg* 1986 Feb;3(2):10-3

The relative efficacies of blood, urine, stool and bone-marrow cultures for the isolation of *Salmonella typhi* were evaluated in 8 patients aged 1-15 years, hospitalised with typhoid fever in Dhaka, Bangladesh. All the patients had received some form of antibiotic therapy before presentation. Bone-marrow cultures were positive in 7 (87.5%) patients, stool cultures in 4 (50%), blood cultures in 2 (25%) and urine culture in 1 (12.5%). *S. typhi* was isolated from bone-marrow alone in 2 patients and from stool alone in 1 patient. All isolates were sensitive to ampicillin, chloramphenicol, and cotrimoxazole. The results suggest that bone-marrow culture is the most sensitive test for the isolation of *S. typhi*. Prior antibiotic therapy suppresses *S. typhi* bacteraemia, but does not eradicate the organism sequestered in the bone-marrow.

253 Kinney JS, Viscidi RP, Vonderfecht SL, Eiden JJ, Yolken RH. Monoclonal antibody assay for detection of double-stranded RNA and application for detection of group A and non-group A rotaviruses. *J Clin Microbiol* 1989 Jan;27(1):6-12

"Fastidious viruses are generally detected in human body fluids by means of immunoassay or nucleic acid hybridization systems. These approaches can be difficult to apply to the detection of viruses which display variations in antigenic or genetic composition. Rotaviruses are examples of viruses which can display such variations. Recently identified antigenic variants, designated as non-group A rotaviruses, cannot be detected by immunoassays or nucleic acid hybridization assays which utilize reagents directed at group A rotavirus strains. The incomplete understanding of the extent of antigenic variation has inhibited the development of assay systems for all of the non-group A rotaviruses and has limited the study of their role in human disease. While rotaviruses display genetic variation, they all contain a genome which consists of double-stranded RNA. We utilized a monoclonal antibody to devise a sensitive assay for the measurement of double-stranded RNA and applied it to the detection of a wide range of rotaviruses. We found that the assay could detect double-stranded RNA from

as few as 10 PFU of standard strains of group A rotaviruses. The assay system was also capable of detecting double-stranded RNA from several strains of group B rotaviruses isolated from calves, rats, and pigs at levels below those at which viral RNA could be visualized by means of polyacrylamide gel electrophoresis. When applied to the detection of double-stranded RNA in serial stools shed by rotavirus-infected children, the assay system was capable of detecting double-stranded RNA in samples in which antigen could not be detected by immunoassay. The specific nature of the double-stranded RNA detected by this assay system could be determined by the elution of the nucleic acids from the monoclonal antibody and the reaction of the RNA with specific nucleotide probes. The measurement of double-stranded RNA offers a potential method for the sensitive detection of a wide range of rotaviruses and other members of the family *Reoviridae*." (Authors' abstract)

254 Kitamura K, Kuwata K, Sasaki Y, Ishii T, Kamachi M, Watanabe S, Kobayashi T, Takahashi T. [Two cases of amoebiasis diagnosed by immunoserological methods]. *Nippon Shokakibyo Gakkai Zasshi* 1985 Jul;82(7):1771-4

255 Kjaeldgaard P, Nissen B, Lange N, Laursen H. Evaluation of minibact, a new system for rapid identification of *Enterobacteriaceae*. *Acta Pathol Microbiol Immunol Scand* 1986 Apr;94B(2):57-61

256 Klipstein FA, Engert RF, Short HB. Enzyme-linked immunosorbent assays for virulence properties of *Campylobacter jejuni* clinical isolates. *J Clin Microbiol* 1986 Jun;23(6):1039-43

"To evaluate the capacity of enzyme-linked immunosorbent assays (ELISAs) to identify pathogenic strains among clinical fecal isolates of *Campylobacter jejuni*, 40 consecutively obtained strains from 39 sick patients and 1 asymptomatic person were tested by respective ELISAs for enterotoxin production in culture filtrates and for the invasive virulence antigen of bacterial cells. Of the 40 strains, 14 produced the enterotoxin; 15 strains, two of which were also enterotoxigenic, were invasive; and 11 strains had no detectable virulence property. The presence or absence of these virulence properties was confirmed by the demonstration that viable cells of all 12 randomly selected enterotoxigenic or invasive strains tested, but none of 9 nonpathogenic strains tested, caused fluid secretion in rat ligated ileal loops. All 12 patients examined who were infected with an invasive strain had grossly or microscopically evident blood cells in their stools or both, whereas none of those infected with an enterotoxigenic strain had overtly bloody diarrhea, and only 1 of 8 patients examined had microscopically evident blood cells in the stool. Twelve of the invasive, five of the enterotoxigenic, and three of the nonpathogenic strains also produced small amounts of cytotoxin, but there was no correlation between cytotoxin production and an abnormal response in rat ligated ileal loops. These observations show that enterotoxin production or invasiveness or both can be detected by ELISAs in three-fourths of *C. jejuni* fecal isolates and that there is usually a relationship between the specific pathogenic property of the infecting strain and the clinical manifestations." (Authors' abstract)

257 Knight IT, Shults S, Kaspar CW, Colwell RR. Direct detection of *Salmonella* spp. in estuaries by using a DNA probe. *Appl Environ Microbiol* 1990 Apr; 56(4):1059-66

258 Knisley CV, Bednarz-Prasad AJ, Pickering LK. Detection of rotavirus in stool specimens with monoclonal and polyclonal antibody-based assay systems. *J Clin Microbiol* 1986 May;23(5):897-900

"Accurate diagnosis of rotavirus is important in both clinical and research situations. A total of 100 stool specimens from children with diarrhea were tested for rotavirus by electron microscopy. These specimens were then coded and tested for rotavirus by four procedures: a monoclonal antibody-based enzyme immunoassay (EIA) (Pathfinder; Kallestad Laboratories, Inc., Austin, Tex.), two polyclonal antibody-based EIAs (Rotazyme

II; Abbott Laboratories, North Chicago, Ill.; and an EIA performed with reagents from the National Institutes of Health, Bethesda, Md. [NIH reagent EIA]), and a latex agglutination (LA) assay (Rotalex; Medical Technology Corp., Somerset, N.J.). The sensitivity of the monoclonal antibody EIA (95%) was superior to those of the polyclonal antibody EIAs (73% for Rotazyme II and 57% for the NIH reagent EIA) and the LA assay (61%). The specificity of the LA assay (98%) was slightly better than those of the other systems (88 to 96%). The positive and negative predictive values of the monoclonal antibody EIA (93 and 96%, respectively) were better than those of Rotazyme II (82 and 80%, respectively), the LA assay (96 and 76%, respectively), and the NIH reagent EIA (93 and 74%, respectively). The visual readings of the monoclonal antibody EIA correlated better with the spectrophotometric optical density readings than did the visual readings of the polyclonal antibody EIAs; however, the agreement of both with electron microscopy results was poor when 1+ or plus-minus readings were observed. The monoclonal antibody EIA is more sensitive and predictive than other rotavirus detection systems and second only to the LA assay in specificity in detecting rotavirus in stool specimens." (Authors' abstract)

259 Knutton S, Baldwin T, Williams PH, McNeish AS. New diagnostic test for enteropathogenic *Escherichia coli* (letter). *Lancet* 1988 Jun 11;1(8598): 1337

260 Kohli E, Pothier P, Denis F, Freymuth F, Goudeau A. Multicentre evaluation of a new commercial latex agglutination test using a monoclonal antibody for rotavirus detection. *Eur J Clin Microbiol Infect Dis* 1989 Mar;8(3):251-3

"A new commercial latex-agglutination test using a monoclonal antibody for detection of rotavirus (Slidex Rota-Kit 2) was compared with three other tests (Slidex Rota-Kit, Rotalex, Rotazyme II) using immunoelectron microscopy and a monoclonal enzyme immunoassay as reference tests. Slidex Rota-Kit 2 was more sensitive and specific than the other tests, and would thus appear to be a practical and accurate rotavirus assay for use in routine laboratory work." (Authors' abstract)

261 Kokka RP, Janda JM. Isolation and identification of autoagglutinating serogroup O:11 *Aeromonas* strains in the clinical laboratory. *J Clin Microbiol* 1990 Jun;28(6):1297-9

262 Kolbl S, Vogel I, Modli M, Gerstl F. [Comparison of the diagnostic methods for studying parvovirus and rotavirus infections of dogs]. *Berl Munch Tierarztl Wochenschr* 1990 Jul 1;103(7):232-6

"79 feces samples of dogs between 7 weeks till 13.5 years of age, showing clinical signs of a hemorrhagic gastroenteritis, were tested by a commercial ELISA (DiaSystems Canine Parvo, Tech-America) for canine parvovirus (CPV) and by two latex-agglutination tests for rotavirus (30 probes were tested by Rota Screen, 49 samples by Slidex Rota-Kit 2, both tests from BioMerieux). All samples were also examined by electron microscopy. The results of the simultaneous investigations showed 28 times positive and 28 times negative for CPV (70.9%). In 93.7% the investigations for rotavirus-infection showed identical results by the latex-agglutination and electron microscopy: 73 samples were negative, one case showed a positive reaction. In 4 feces samples rotavirus could only be detected by the latex-test. In one sample a double-infection (CPV/rotavirus) could be observed by all methods, in two cases the double-infection was only found by using the latex-agglutination. No other viruses could be found by the electron microscope than those described above. The results and the performance of the methods are discussed and compared with the data of other authors." (MEDLINE)

263 Kolkman JJ, Vogten AJ. *Campylobacter jejuni*: clinical and diagnostic value of serum antibody titres. *Neth J Med* 1990 Feb;36(1-2):46-52

"Eighty patients with either bacteriologically confirmed *Campylobacter jejuni* infection and/or an antibody titre value of at least 1:80, determined by ELISA, were studied. A

significant correlation was found between titre value and severity of symptoms ($P = 0.015$). Although a correlation was noted between symptoms score and endoscopic abnormalities, this was not quite statistically significant ($P = 0.053$). Comparison of patients with a titre of at least 1:1280 and those with lower titre values revealed a significantly higher symptom score ($P = 0.019$) and endoscopic score ($P = 0.015$) in patients with a higher antibody level. By using the previously recommended titre of 1:640 as a cut-off point for active infection, all significant differences were lost. Of 12 patients with positive stool culture, 7 had a titre value of at least 1:1280, suggesting a sensitivity of 58%. However, of 20 patients with negative culture, 4 showed this titre value, three of whom were studied several weeks after the onset of their illness. In those patients with clinically proven *Campylobacter* infection, the antibody response was characterized by a rapid initial rise and a slow four-fold drop in antibody titre after 3 to 5 months. Colonic involvement of the infection was seen in 63% of our patients with positive cultures. Our results support the conclusion that ELISA is a valuable method of diagnosing *C. jejuni* infections when stool cultures are likely to become negative, as is the case in prolonged complaints or complications after gastro-enteritis or in proctocolitis or after the use of antibiotics. Serial serum samples have no advantage over a single sample for antibody detection." (MEDLINE)

264 Kornaid JA, de Castagnaro NR. [Enzyme immunoassay for rotavirus using nylon as the solid phase]. *Rev Argent Microbiol* 1987 Apr-Jun;19(2):77-9

"An enzyme-linked immunoassay (EIA) to detect rotavirus in stools is described. Antibodies prepared in rabbits were immobilized on small nylon cubes as capture phase and enzyme conjugated antibodies were used to reveal the reaction. The conjugate was prepared with horseradish peroxidase by the Nakane periodate oxidation method. The solid phase consisted of 3 mm nylon cubes (66 CNL Du-cilo) previously submitted to partial acid hydrolysis to liberate amino-reactive groups. Glutaraldehyde was employed to couple the capturing antibody to the solid phase resulting in a covalent linkage between the gamma-globulin and the nylon. Phenylenediamine in citrate buffer pH 5.0 with 0.5% hydrogen peroxide was used as revealing substrate. EIA was performed as follows: stools watery extracts were incubated 1 h at 37°C with antibody-treated nylon cubes, and then with enzyme conjugate, rinsed with distilled water and substrate-added. Samples developing colour, with optical density of at least 0.350 at 492 nm, were considered positive. The method showed good correlation with a commercial kit." (MEDLINE)

265 Kongmuang U, Honda T, Miwatani T. Enzyme-linked immunosorbent assay to detect Shiga toxin of *Shigella dysenteriae* and related toxins. *J Clin Microbiol* 1987 Jan;25(1):115-8

"A sandwich enzyme-linked immunosorbent assay (ELISA) was developed for detection of Shiga toxin. Four species of *Shigella*, *Escherichia coli*, and *Vibrio parahaemolyticus* were tested for production of Shiga or Shiga-like toxin by ELISA and Vero cell bioassay. In the ELISA, most strains of *S. dysenteriae* and some strains of *E. coli* isolated from traveler's diarrhea were positive. These ELISA-positive strains were positive by Vero cell bioassay without exception. Some *E. coli* strains and most *V. parahaemolyticus* strains were toxic to Vero cells, although they were negative in the ELISA. Much of the cytotoxic activity was not neutralized by anti-Shiga toxin antiserum. The newly developed sandwich ELISA is specific and can be a substitute for the cumbersome Vero cell bioassay." (Authors' abstract)

266 Korman SH. The duodenal string test. A simple multipurpose diagnostic tool in clinical pediatrics. *Am J Dis Child* 1990 Jul;144(7):803-5

"The duodenal string test capsule is a cheap and simple device used for sampling the contents of the upper gastrointestinal tract. Its major applications in pediatrics are in diagnosis of enteric parasitic infestations, confirmation of contaminated small-bowel syndrome, diagnosis of *Salmonella* infection, and assessment of neonatal cholestasis. Pediatricians should be aware of this invaluable diagnostic aid." (Authors' abstract)

267 Korman SH, Hais E, Spira DT. Routine *in vitro* cultivation of *Giardia lamblia* by using the string test. J Clin Microbiol 1990 Feb;28(2):368-9

268 Kovacs A, Chan L, Hotrakitya C, Overturf G, Portnoy B. Rotavirus gastroenteritis: clinical and laboratory features and use of the Rotazyme test. Am J Dis Child 1987 Feb;141(2):161-6

"Clinical and laboratory features of 86 infants admitted with diarrhea and dehydration were evaluated prospectively. Human rotavirus (HRV) infection was documented in 35 infants (41%) by the Rotazyme test. Those with HRV gastroenteritis (HRV+ group) had a shorter duration of diarrhea prior to admission, more severe dehydration on presentation, and a longer hospital course than the HRV-negative (HRV-) group. Vomiting, fever, upper respiratory tract symptoms, otitis media, and cough were present in equal numbers of infants in both groups. The HRV+ infants had lower serum bicarbonate and higher serum albumin, alanine aminotransferase, aspartate aminotransferase, and uric acid concentrations than did the HRV- infants. Serum uric acid levels greater than 10 mg/dL (590 μ mol/l) were present in 69% of HRV+ vs 29% of HRV- infants. The Rotazyme test was found to be a valuable tool in diagnosis; testing on two days increased the yield from 74% to 97% of all infants finally diagnosed as HRV+. The optimal time for testing was within the first five days of illness." (Authors' abstract)

269 Krishnan C. Detection of *Clostridium difficile* toxins by enzyme immunoassay. J Hyg (Camb) 1986 Feb;96(1):5-12

"An enzyme-linked immunosorbent assay (ELISA) for the rapid diagnosis of antibiotic-associated colitis (AAC) is presented. Commercially available antisera to *Clostridium difficile* toxins contain antibodies to other antigens found in non-toxicogenic *C. difficile* and other bacteria. Removal of these unwanted antibodies by absorption increased the specificity of ELISA for detection of *C. difficile* toxins. Specimens tested included 40 faecal extracts positive for cytotoxicity from cases of AAC, 30 diarrhoeic and 30 well-formed stools negative for cytotoxicity and 50 culture filtrates of toxigenic and non-toxicogenic *C. difficile* and other clostridial species. Use of absorbed sera reduced false-positive reactions observed with faecal specimens from 23 to 8%. About 90% of specimens that were positive by the tissue culture cytotoxicity test were positive by ELISA using the absorbed sera. The relative merits of ELISA and other methods for the rapid diagnosis of AAC are discussed." (Authors' abstract)

270 Kruger G. [Current trends in the development of *Salmonella* detection methods]. Z Gesamte Hyg 1989 Nov;35(11):683-5

"The cultural methods require 4 to 7 days for presumptive evidence of *Salmonella* in foodstuffs. Attempts in time shortening have resulted in combination of pre-enrichment or selective enrichment with time saving genetical or immunological tests. Proceedings of enzyme immunoassays for applications in *Salmonella* screening are important. Involving monoclonal antibodies, fluorescent or chemiluminescent substrates, there are some commercial *Salmonella* test kits. Especially rapid EIA methods, here advantages and disadvantages are discussed." (MEDLINE)

271 Kuhn HM, Basu S, Mayer H. Comparison of enterobacterial common antigen from different species by serological techniques. Eur J Biochem 1987 Jan 2;162(1):69-74

"Enterobacterial common antigen (ECA) was isolated from a number of selected species (including *Salmonella montevideo*, *Shigella sonnei* and *Plesiomonas shigelloides*) using the extraction method described by Manneil and Mayer [Eur. J. Biochem. 86, 361-370 (1978)]. ECA of all these species behaved identically in enzyme-linked immunosorption assay (ELISA) and in its inhibition using monoclonal anti-ECA antibodies. Immunoblotting showed a ladder-like pattern of at least 20 bands for all preparations tested. ECA

modified at its lipid moiety (e.g. by phospholipases A2 and D or by mild acid hydrolysis) lost its coating capacity leaving, however, the serological reactivity as detected by inhibition assays intact. In contrast, reduction of the carboxylic groups of 2-acetamido-2-deoxy-D-mannopyranosyluronic acid destroyed the serological reactivity. Deacylated ECA was also not detectable in immunoblotting. Chemical reacylation restored the reactivity of deacylated ECA in ELISA and in immunoblot and thus proved the essential function of fatty acids for the physicochemical properties of the molecule. 2-Acetamido-2-deoxy-D-glucopyranose was identified as the reducing end of the ECA sugar chain after splitting off the lipid moiety by phospholipase D." (MEDLINE)

272 Kumar S, Band AH, Samantaray JC, Dang N, Talwar GP. A dot enzyme-linked immunosorbent assay for detection of antibodies against *Entamoeba histolytica*. J Immunol Methods 1985 Oct 24;83(1):125-33

"A visual micro-dot enzyme-linked immunosorbent assay (ELISA) based on detection of antibodies against soluble antigens of axenically grown cultures of *Entamoeba histolytica* is described. The antigen was spotted on a nitrocellulose sheet, the unsaturated sites blocked with bovine serum albumin (BSA) and incubated with 3-fold dilutions of patient sera followed by incubation with protein A conjugated to peroxidase. Enzymic activity was evidenced using the substrate 4-chloro-1-naphthol. A positive reaction produced a blue spot. The sensitivity of the assay was better and comparable to the indirect haemagglutination assay (IHA) and plate ELISA. The entire assay could be completed within 3 h. Antigen-loaded and pre-blocked nitrocellulose strips could be stored up to 3 months at room temperature and 37°C." (Authors' abstract)

273 Kurstak E, Tijssen P, Kurstak C, Morisset R. Enzyme immunoassays and related procedures in diagnostic medical virology. Bull WHO 1986;64(3): 465-79

274 Lahesmaa-Rantala R, Granfors K, Toivanen A. Detection of circulating *Yersinia*-immunoglobulin complexes by enzyme immunoassay (EIA). J Immunol Methods 1986 May 22;89(2):191-9

"An enzyme immunoassay (EIA) was developed for the detection of *Yersinia*-immunoglobulin complexes of known Ig class. Immune complexes (ICs) were attached to polystyrene microtiter plates by rabbit anti-human immunoglobulins, and the existence of *Yersinia enterocolitica* O:3 antigens was demonstrated using Fab fragments of alkaline phosphatase (AP)-conjugated antibody against the same serotype. Simultaneous binding of *Yersinia* antigens and immunoglobulins was a prerequisite for the detection of ICs. The method will be valuable for research into the immunopathogenetic mechanisms leading to reactive arthritis after *Yersinia* infection." (MEDLINE)

275 Lampel KA, Jagow JA, Trucksess M, Hill WE. Polymerase chain reaction for detection of invasive *Shigella flexneri* in food. Appl Environ Microbiol 1990 Jun;56(6):1536-40

"The polymerase chain reaction (PCR) was used to amplify a 760-base-pair (bp) fragment with the 220-kbp invasive plasmids of enteroinvasive *Escherichia coli*, *Shigella flexneri*, *Shigella dysenteriae*, *Shigella boydii*, and *Shigella sonnei* as templates. This PCR product was easily detected by agarose gel electrophoresis. A 210-bp *AccI*-*PstI* fragment lying within the amplified region was used as a probe in southern hybridization blots and showed that the PCR-generated product was derived from the invasive plasmid. The application of PCR as a rapid method to detect enteroinvasive bacteria in foods was tested by inoculating lettuce with 10^4 *S. flexneri* cells per g in *Shigella* broth base. Plasmid DNA was isolated from cultures of inoculated and uninoculated lettuce in broth after 0, 4, and 24 h of incubation. With the PCR, the 760-bp fragment was generated only from lettuce inoculated with *S. flexneri*, as shown by gel electrophoresis and confirmed both by southern blotting and by nucleotide sequencing of the amplified region. Because the isolation of plasmid DNA, the performance of PCR, and gel

electrophoresis all can be completed in 6 to 7 h, invasive enteric bacteria can be detected in less than 1 day." (Authors' abstract)

276. Lanser JA, Anargyros PA. Detection of *Escherichia coli* adhesins with DNA probes. *J Clin Microbiol* 1985 Sep;22(3):425-7

"DNA hybridization assays for genes encoding the *Escherichia coli* adhesins K88 and K99 were developed. These assays were used to screen a variety of animal *E. coli* strains, and the results were compared with results obtained by serological methods. All methods compared well for determining the presence of adhesin K99. However, the DNA hybridization detected more K88-positive strains than did either enzyme immunoassay or slide agglutination. Agreement was improved when strains were grown on blood or nutrient agar instead of Minca medium." (Authors' abstract)

277. Lecomte C, Pin JJ, De Moerlooze L, Vandenberg D, Lambert AF, Pastoret PP, Chappuis G. ELISA detection of bovine viral diarrhoea virus specific antibodies using recombinant antigen and monoclonal antibodies. *Vet Microbiol* 1990 Jun;23(1-4):193-201

"A panel of monoclonal antibodies was prepared by immunization of BALB/c mice with Moredun (BD) virus strains. These antibodies were characterized by immunofluorescence and seroneutralization against BD, BVD and hog cholera (HC) virus strains, and radioimmunoprecipitation of BVD-infected cells extracts. The MAbs reacting with the majority of the Pestivirus strains recognize the 80 kDa antigen of the BVD cytopathic strains. The 80 kDa antigen of the BVD/Osloss virus strain has been cloned and expressed in *E. coli* as a fusion protein with beta-galactosidase. The fusion protein has been purified from inclusion bodies and used successfully as an antigen for ELISA detection of BVDV specific antibodies in bovine sera. A competitive ELISA using MAbs is more specific than a direct assay. These results compare well with the ones obtained with antigen extracted from BVDV-infected cells." (MEDLINE)

278. Lee CL, Lee YS. Use of avidin-biotin-peroxidase complex in immunoassay for detecting cholera toxin. *Chung Hua Min Kuo Wei Sheng Wu Chi Mien I Hsueh Tsa Chih* 1990 Feb;23(1):98-100

279. Lee CN, Kao CL, Ho HJ, Lee CY. Comparison of Rotazyme, Slidex Rota-Kit, counterimmunoelectrophoresis with solid-phase immune electron microscopy for the detection of human rotavirus. *Taiwan I Hsueh Hui Tsa Chih* 1987 Mar;86(3):227-32

280. Lee HA, Wyatt GM, Bramham S, Morgan MRA. Enzyme-linked immunosorbent assay for *Salmonella typhimurium* in food: feasibility of 1-day *Salmonella* detection. *Appl Environ Microbiol* 1990 Jun;56(6):1541-6

"A microtitration plate, antibody-capture, enzyme-linked immunosorbent assay was developed for detection of *Salmonella typhimurium*. The assay utilizes a monoclonal detector antibody which shows no cross-reactions with non-*Salmonella* species and only a slight cross-reaction with one other *Salmonella* serotype. By using only one cultural stage (in a nonselective, chemically defined medium) prior to the enzyme-linked immunosorbent assay, low numbers of cells in food (10 cells 25 g⁻¹) were detected in 19 h. Non-*Salmonella* competing organisms did not interfere with detection of *S. typhimurium* even when present in the ratio of 10⁶:1 (non-*Salmonella*/*Salmonella* spp.). The assay shows the feasibility of rapid, 1-day testing for *Salmonella* spp. with antibody technology." (Authors' abstract)

281. Leechanachai P, Yoosook C, Saganwongse S, Matangkasombut P. Comparison of a modified enzyme-linked immunosorbent assay with immunosorbent electron microscopy to detect coronavirus in human faecal specimens. *J Diarrhoeal Dis Res* 1987 Mar;5(1):24-9

"Two commonly used methods to detect human enteric coronavirus in faecal specimens were compared. All samples were screened by a modified enzyme-linked immunosorbent assay (ELISA). Positive samples were confirmed by an ELISA with a blocking test. Randomly selected negative and positive samples by the ELISA with a blocking test were then subjected to immunosorbent electron microscopy using a protein A coated grid technique (PA-CGT). Out of 39 negative specimens using an ELISA with a blocking test, 38 (97.4%) were still negative by PA-CGT whereas coronaviruses were seen in only 26 out of 32 (81.2%) samples positive by the ELISA with a blocking test. This indicated that the ELISA with a blocking test is superior to PA-CGT for detecting human enteric coronaviruses." (Authors' abstract)

282 Leechanachai P, Yoosook C, Matangkasombut P. Epidemiological study of enteric coronavirus excretion by an enzyme-linked immunosorbent assay. *J Med Assoc Thai* 1989 Aug;72(8):452-7

"The enzyme-linked immunosorbent assay with a blocking test was used to determine the prevalence of enteric coronavirus excretion among the population of Bangkok and Khon Kaen province. The results indicated that coronaviruses were present in fecal samples from both children and adults with or without diarrhea. It is interesting that the viruses were more frequently observed in the apparently healthy individuals in Bangkok than in the patients with diarrhea whereas the excretion rates were approximately alike in both groups in Khon Kaen province. Among children aged less than 5 years (with or without diarrhea) the viruses were more frequently observed in stools from young children (1-5 years old) than from the newborn (less than 1-year-old) particularly those living in the Bang Khen slum area and in the rural Khon Kaen community. The rate of coronavirus excretion did not increase with age and some apparently healthy individuals continued to excrete the viruses for several months. The results also suggested that chronic parasitic infection associated with low socioeconomic status might influence the excretion of enteric coronaviruses in the feces." (Authors' abstract)

283 Lesmana M, Rockhill RC, Sutanti D, Sutomo A. An evaluation of alkaline peptone water for enrichment of *Vibrio cholerae* in feces. *Southeast Asian J Trop Med Public Health* 1985 Jun;16(2):265-7

The sensitivity of direct inoculation of thiosulfate citrate bile salts agar (TCBS) was compared with that of alkaline peptone water (APW) enrichment prior to direct inoculation of TCBS in the recovery of *Vibrio cholerae* from fecal specimens. *V. cholerae* biotype El Tor was isolated from 611 of 1262 fecal specimens collected from patients hospitalized in Jakarta, Indonesia. Of the 611 isolates, 535 were isolated by both TCBS - direct plate method and APW enrichment followed by plating; 15 by direct plating alone and 61 only after APW enrichment. Thus, there was a significant difference ($p < 0.001$) between the direct plating method only and the APW enrichment followed by plating in the recovery of *V. cholerae* biotype El Tor from the specimens. *V. parahaemolyticus* (21) and non-agglutinating vibrios (11) were also isolated but more often in directly inoculated TCBS than APW-TCBS cultures. Maximum isolation sensitivity of *V. cholerae* and *V. parahaemolyticus* from feces was obtained by both direct inoculation of TCBS and enrichment in APW prior to TCBS inoculation. The results support an earlier observation that both direct plating and APW enrichment should be done to recover a maximum number of *V. cholerae* from feces of cholera patients.

284 Lesmana M, Tjanidi P, Edman D, Lane E, Hendarwanto. The use of a coagglutination test to presumptively identify *Salmonella typhi* in bone marrow-oxgall medium cultures from typhoid fever patients. *Southeast Asian J Trop Med Public Health* 1990 Jun;21(2):203-6

285 Levine MM, Young CR, Black RE, Takeda Y, Finkelstein RA. Enzyme-linked immunosorbent assay to measure antibodies to purified heat-labile enterotoxins from human and porcine strains of *Escherichia coli* and to cholera toxin: application in serodiagnosis and seroepidemiology. *J Clin Microbiol* 1985 Feb;21(2):174-9

"Serum immunoglobulin G antibodies to purified heat-labile enterotoxin (LT) from human (LTh) and porcine (LTp) *Escherichia coli* strains and cholera enterotoxin (CT) were measured by an enzyme-linked immunosorbent assay. Sera from patients with LTh *E. coli* infection showed a prominent response with LTh, an intermediate response with LTp, and a meager response with CT. Of 47 persons with clinical LTh-producing *E. coli* (herein shortened to LTh *E. coli*) infections, significant rises in antitoxin were detected against LTh in 36 (77%), against LTp in 30 (64%), and against CT in only 13 (28%) patients; seroconversions also occurred in 11 of 14 (79%) patients with subclinical LTh *E. coli* infections. In North Americans with experimental LTh *E. coli* infection, anti-LTh did not remain at high levels for more than 3 months. Persons with cholera manifested antitoxin responses that were similarly potent against all three toxin antigens; in fact, net optical density values were often slightly higher against LTh than against CT. The ratio of CT/LTh ELISA net optical density in convalescent sera proved to be a sensitive means to differentiate LT *E. coli* from cholera infection. All 11 cholera patients tested had CT/LTh ratios of >0.70 , whereas in only 1 of 47 LTh *E. coli* infections did the ratio exceed that value (it was 0.71) ($p < 0.0000000001$). In single serum specimens, a net optical density of ≥ 0.30 against LTh was shown to be a useful cutoff in screening sera for recent LTh *E. coli* or past cholera infection. The CT/LTh ratio was then used to differentiate definitively. Sera from healthy 3- to 5-year-olds and 15- to 19-year-olds in Maryland, Chile, and Bangladesh were tested against LTh and CT. The serological results fit known epidemiological observations. (i) LTh infections are rare in the United States (only 2 of 60 sera had LTh net optical density values of ≥ 0.30). (ii) In contrast, evidence of recent LTh *E. coli* infections was very common in Chilean (69%) and Bangladesh (57%) 3- to 5-year-olds and not uncommon in 15- to 19-year-olds (38 and 31%, respectively) in those countries. (iii) Only Bangladeshi sera showed serological evidence of cholera infections (CT/LTh ratios of 0.70). The immunoglobulin G enzyme-linked immunosorbent assay measuring antibodies to purified LTh and CT represents a practical and effective tool for the serological study of LTh *E. coli* and cholera diarrheal infections." (Authors' abstract)

286 Lew JF, Glass RI, Petric M, Lebaron CW, Hammond GW, Miller SE, Robinson C, Boutiller J, Riepenhoff-Talty M, Payne CM, Franklin R, Oshiro LS, Jaqua M-J. Six-year retrospective surveillance of gastroenteritis viruses identified at ten electron microscopy centers in the United States and Canada. *Pediatr Infect Dis J* 1990 Oct;9(10):709-14

"To identify the prevalence, seasonality and demographic characteristics of patients with viral gastroenteritis, we reviewed 6 years of retrospective data on viral agents of gastroenteritis screened by electron microscopy at 10 centers in the United States and Canada. From 52,691 individual electron microscopic observations, a virus was detected in 16% of specimens, and the yearly positive detection rate among centers ranged from 8 to 34%. Rotavirus was the agent most commonly observed (26 to 83%), followed by adenoviruses (8 to 27%, respiratory and enteric combined), and small round viruses (SRVs) (0 to 40%) which were second most common at two of the centers. Rotavirus and astrovirus detections occurred more often in the winter but seasonal trends in detection were not apparent for the other viruses. Of all astroviruses detected 64% were found in infants (<1 year); unlike the other agents studied SRVs were detected in a large percentage of infants (48%) and older children and adults (20%). Among hospitalized patients a majority of all astroviruses, caliciviruses and SRVs were detected 7 days or more after admission in contrast to both rotaviruses and adenoviruses which were more likely to be detected earlier. The data suggest that SRVs are common agents of gastroenteritis and may be important causes of nosocomial infections. Because of the relative insensitivity of direct electron microscopy as a screening method for SRVs, astroviruses and caliciviruses, these data probably underestimate the true prevalence of disease caused by these agents." (Authors' abstract)

287 Lin TM, Schubert CM, Klefer DJ, Troy S, Cort R, Giegel JL, Lin JH, Neill L. Two rapid and simple enzyme immunoassays for human antibodies to *Entamoeba histolytica*. *J Immunoassay* 1989;10(4):301-13

"Two rapid and simple enzyme immunoassays (EIA) for antibodies to *E. histolytica* the protozoa causing amebiasis, are described. In the rapid dot EIA, a qualitative procedure, antigens were dried as a small dot (3 mm in diameter) on a thin white opaque polystyrene strip and serum samples were assayed undiluted. The assay required 3 incubation periods, 1 to 3 minutes each, and was completed in 9 minutes, with a positive reaction revealed as a blue color (precipitate) on the antigen dot and negative as colorless. The developed color is stable for permanent record. In the Microwell EIA, a quantitative procedure, antigens were dried in the Microwells. The assay also consisted 3 incubation periods of 15 minutes each, and was completed in 50 minutes. The results in absorbance values were normalized to EIA units (EU). Both tests had good reproducibility, sensitivity and specificity; and highly correlated with 3 other serologic tests. Their reagents can be stored for more than a year. Both tests could be suitable for small and physicians' office laboratories, especially in developing countries." (MEDLINE)

288 Lindberg AA, Cam PD, Chan N, Phu LK, Trach DD, Lindberg G, Karlsson K, Kamell A, Ekwall E. Shigellosis in Vietnam: seroepidemiologic studies with use of lipopolysaccharide antigens in enzyme immunoassays. Rev Infect Dis 1991 Mar-Apr;13(suppl 4):S231-7

"The performance of enzyme immunoassays (EIAs) with use of O-antigen-containing lipopolysaccharides (LPSs) extracted with phenol-water from *Shigella dysenteriae* type 1, *Shigella flexneri* serotypes 1a-5b, and *Shigella sonnei* for determination of the serum antibody response after onset of bacillary dysentery is reviewed. For the purpose of several studies, serum samples from a total of 175 Vietnamese and 47 Swedish patients, for whom *Shigella* species had been isolated from fecal specimens, were obtained at various intervals until ≤ 1 year after the onset of infection. Titers of antibodies in serum samples from infected patients were compared with those in serum samples from healthy control subjects; the combined control population of all studies comprised 426 Vietnamese and 154 Swedes. The sensitivity of the EIAs ranged from 78% to 100% for patients whose fecal culture was positive for *Shigella*. For diagnosis of *S. flexneri*, a species-specific but no serotype-specific assay based on LPS antigens is possible. Among Vietnamese patients the EIA with use of *S. flexneri* was sensitive and diagnostic only for children <3 years of age, most likely because healthy older Vietnamese children and adults have high titers of antibody to the O-antigens of *S. flexneri*. Among Swedish patients the same EIA was diagnostic for adults as well as children. Increased titers of IgA in the early phase and IgG in the convalescent phase, as determined by EIA, were the best indicators of infection due to *Shigella* species." (Authors' abstract)

289 Liprandi F, Brito B, Palencia L, Esparza J. Derivation of a monoclonal antibody against the group specific antigen of rotaviruses and its use in a diagnostic enzymatic immunoassay. Acta Cient Venez 1986;37(4):432-6

290 Lipson SM, Zellnsky-Papez KA. Comparison of four latex agglutination (LA) and three enzyme-linked immunosorbent assays (ELISA) for the detection of rotavirus in fecal specimens. Am J Clin Pathol 1989 Nov;92(5):637-43

"Eighty-two stool specimens obtained from children with gastrointestinal disease were tested for the presence of antigen to rotavirus by latex agglutination (LA) (Virogen[®] (VR), Meritec[®] (MER), Wellcome[®] (WEL), Sildex Rotatest[®] (SRT), and enzyme-linked immunosorbent assays (Rotaclone[®] (TRC), Rotazyme II[®] (RTZ), Pathfinder[®] (PTH)). Confirmatory testing was performed by isolation of rotavirus from stool specimens with the use of a shell vial centrifugation, antigen-detection tissue culture amplification method. The sensitivities and negative predictive values of VR, MER, WEL, SRT, TRC, RTZ, and PTH tests were 85, 89, 95, 91, 98, and 100%, respectively. Each test demonstrated 100% specificity and positive predictive values except the SRT, which attained a specificity of 95%. The WEL LA test may be used as a preliminary rapid screening assay following a stat request. The Kallestad PTH ELISA, however, was determined to

be the rotavirus antigen detection kit of choice for routine laboratory diagnostic testing." (Authors' abstract)

291 Lipson SM, Leonardi GP, Salo RJ, Schutzbank TE, Kaplan MH. Occurrence of nonspecific reactions among stool specimens tested by the Abbott TestPack rotavirus enzyme immunoassay. *J Clin Microbiol* 1990 Jun;28(6):1132-4

"Sixty-five stool specimens obtained from children suffering from gastroenteritis were tested for the presence of antigen to rotavirus by the Abbott TestPack Rotavirus (TestPack) enzyme immunoassay kit. The Kallestad Pathfinder enzyme immunoassay, polyacrylamide gel electrophoresis, immune electron microscopy, and virus isolation were utilized as reference assays. Fifty-four specimens were in accord by TestPack and Kallestad Pathfinder. Among 11 discordant specimens positive with TestPack but negative by Kallestad Pathfinder, rotavirus was not identified by polyacrylamide gel electrophoresis, immune electron microscopy, or isolation in primary African green monkey kidney cell cultures. TestPack displayed a performance specificity of 83%. The inordinately high number of stool specimens reported as false-positive by TestPack precludes the incorporation of this antigen detection kit into our routine regimen of diagnostic virologic testing." (Authors' abstract)

292 Litinski Iul, Gerok GI, Godovannyi BA. [Experimental study of the sensitivity of the enzyme-labeled antibody reaction and the coagglutination reaction for detection of *Shigella* and *Salmonella* antigens in urine]. *Lab Delo* 1985;35(7):434-6

293 Ljungh A, Ronnberg B, Baloda SB, Yuk YY, Wadstrom T. Coagglutination and ELISA for detection of enterotoxins and colonization factor antigens in *Escherichia coli* and *Salmonella*. *Ann Ist Super Sanita* 1986;22(3):863-8

294 Loffeld RJLF, Stobberingh E, Flendrig JA, van Spreeuwel JP, Arends JW. Diagnostic value of an immunoassay to detect anti *Campylobacter pylori* antibodies in non-ulcer dyspepsia. *Lancet* 1989 May 27;1(8648):1182-5

"An enzyme-linked immunosorbent assay (ELISA) for detection of IgG antibodies against *Campylobacter pylori* was used to examine sera from 70 patients with non-ulcer dyspepsia. 48 patients had *C. pylori* associated gastritis according to culture or histology; mean optical density (OD) of the ELISA was significantly higher than that for the 22 patients with normal antral mucosa and absence of *C. pylori*. Positive and negative predictive values for *Campylobacter*-associated gastritis were 100% above OD 2.10 and below OD 1.00, respectively. Serology might replace endoscopy in the diagnosis of *Campylobacter*-associated gastritis." (Authors' abstract)

295 Loose JH, Sedergran DJ, Cooper HS. Identification of *Cryptosporidium* in paraffin-embedded tissue sections with the use of a monoclonal antibody. *Am J Clin Pathol* 1989 Feb;91(2):206-9

296 Losonsky GA, Ferreccio C, Kotloff KL, Kaintuck S, Robbins JB, Levine MM. Development and evaluation of an enzyme-linked immunosorbent assay for serum Vi antibodies for detection of chronic *Salmonella typhi* carriers. *J Clin Microbiol* 1987 Dec;25(12):2266-9

"An enzyme-linked immunosorbent assay (ELISA) measuring, in serum, immunoglobulin G (IgG), IgM, and IgA to Vi capsular polysaccharide antigen that was tyraminated (Vi-Tyr) to increase its binding efficiency to microtiter plates was compared with the standard passive hemagglutination assay (PHA) as a screening test for chronic *Salmonella typhi* carriers. Initially, three populations were evaluated: 22 healthy U.S. adults, 17 young Chilean adults with acute typhoid fever, and 51 Chileans who had bacteriologically confirmed *S. typhi* chronic carriage. IgG-specific Vi-Tyr antibodies were preferentially present in the *S. typhi* chronic carrier state. A total of 44 of 51 (81%) chronic carriers, 0

of 22 (0%) healthy U.S. adults, and 2 of 17 (12%) Chileans with acute typhoid fever had reciprocal IgG Vi-Tyr ELISA antibody titers in serum of ≥ 200 . The IgG Vi-Tyr ELISA was then compared with the PHA as a screening test for chronic carriers in 141 Chilean female food handlers. One woman was serologically incriminated as a carrier by both the IgG ELISA and PHA; her coprocultures were positive for *S. typhi*. One other woman, identified as a carrier by PHA, was negative by culture and IgG ELISA. The IgG Vi-Tyr ELISA is as sensitive as the PHA (86 versus 76%) and as specific (95 versus 95%) in screening for chronic carriers." (Authors' abstract)

297 Lu S-q, Wang Z-y, Zhang Y-q, Wen Y. Detecting *Giardia lamblia* antigen in fecal matter with counterimmuno-electrophoresis in diagnosis of giardiasis. Chin Med J 1989 Sep;102(9):686-8

298 Lucas GP, Cambiaso CL, Vaerman JP. Receptor binding-like specificity of two anti-cholera toxin monoclonal antibodies detected by latex agglutination immunoassay. Immunol Lett 1989 Sep;22(3):227-34

"Six mouse monoclonal IgG antibodies (mAbs) reacting with the cholera toxin B-subunit (CT-B) were obtained from the spleen cells of mice immunized with CT. They were divided into two groups according to their different reactivities with CT in latex (Lx) agglutination. One group of two mAbs was called "non-repetitive" (NR) because they apparently recognized a single epitope on CT, which was unable to agglutinate Lx coated with these NRmAbs. In contrast with the other group of four mAbs that we called "repetitive" (RmAbs). The two NRmAbs, but not the four RmAbs, failed to react with CT when CT was bound first to its GM1-receptor. Thus, the NRmAbs seemed directed at an epitope close to the GM1-binding site of CT-B. These NRmAbs could represent valuable immunogenic candidates for anti-idiotypic immunization against CT and related enterotoxins." (MEDLINE)

299 Lukowicz G, Mounier M, Leboutet MJ, Denis F, Goudeau A. [Evaluation of a new latex test for detecting human rotaviruses in feces]. Pathol Biol (Paris) 1985 Jun;33(6):681-3

"We have sought the presence of rotavirus in 237 fecal samples. Three techniques were applied to each sample: ELISA, latex agglutination (LTX) with two reagents (Rotalex and SlidexRotaKit), and electron microscopy after negative staining. The specificity of ELISA and LTX is very good (more than 93.4 per cent), the sensitivity is decreasing from ELISA to SlidexRotaKit and Rotalex (respectively 83.6-74.5 and 67.3 per cent). The advantages of LTX include rapidly, simplicity, low cost price." (MEDLINE)

300 McConnell MM, Thomas LV, Day NP, Rowe B. Enzyme-linked immunosorbent assays for the detection of adhesion factor antigens of enterotoxigenic *Escherichia coli*. J Infect Dis 1985 Dec;152(6):1120-7

"Two hundred forty-four specimens of *Escherichia coli* isolated in Bangladesh and Thailand and identified as enterotoxin producers were tested for the presence of adhesion antigens by mannose-resistant hemagglutination, immunodiffusion, and enzyme-linked immunosorbent assays (ELISAs). Specific antisera to the antigens colonization factor antigen (CFA)/I, CFA/II (consisting of cell surface antigens [CS] 1, 2, and 3), and putative colonization factor antigen (PCF) 8775 (consisting of CS4, 5, and 6) were used in immunodiffusion tests and ELISAs. The results showed that the antigens could be detected in more strains by ELISA than by immunodiffusion. Twenty-nine percent of specimens of *E. coli* from Thailand and 47% from Bangladesh carried an adhesion antigen. Many of the strains had lost the ability to produce enterotoxins. Forty percent of strains from Thailand and 64% from Bangladesh that were still enterotoxigenic carried adhesion factors. These antigens were found on strains with heat-stable or heat-labile and heat-labile enterotoxin but not on strains producing only heat-labile enterotoxin. PCF8775 antigens were associated mainly with strains from Bangladesh, where 10 strains that produced only CS6 were detected." (Authors' abstract)

- 301 McNabb SJN, Hensel DM, Welch DF, Heljbel H, McKee GL, Istre GR. Comparison of sedimentation and flotation techniques for identification of *Cryptosporidium* sp. oocysts in a large outbreak of human diarrhoea. *J Clin Microbiol* 1985 Oct;22(4):587-9
- 302 McNulty CAM, Dent JC, Uff JS, Gear MWL, Wilkinson SP. Detection of *Campylobacter pylori* by the biopsy urease test: an assessment in 1445 patients. *Gut* 1989 Aug;30(8):1058-62
- 303 Madore HP, Treanor JJ, Pray KA, Dolin R. Enzyme-linked immunosorbent assays for Snow Mountain and Norwalk agents of viral gastroenteritis. *J Clin Microbiol* 1986 Sep;24(3):456-9
- "Enzyme-linked immunosorbent assays (ELISAs) for antigen detection and blocking ELISAs for serum antibody rises were developed for the Snow Mountain and Norwalk agents of viral gastroenteritis. The ELISAs were as sensitive as the existing radioimmunoassays and were specific for the Snow Mountain or Norwalk agent. The blocking ELISAs detected the same number of significant rises in antibodies to these agents as did the existing blocking radioimmunoassays." (Authors' abstract)
- 304 Mahajan RC, Sehgal R, Malla N, Ganguly NK. Dot enzyme linked immunosorbent assay for immunodiagnosis of amoebiasis. *Indian J Med Res* 1989 Mar;89:100-4
- "Dot-enzyme linked immunosorbent assay (Dot-ELISA) used for the serodiagnosis of both intestinal and extraintestinal amoebiasis, showed positivity in 94 per cent patients of amoebic liver abscess and 87.5 per cent of intestinal amoebiasis. Only one of the 18 normal controls was positive by Dot-ELISA test. This test showed good correlation with indirect haemagglutination test. As it is simple and easy to perform, the test appears to have great potential for the specific diagnosis of amoebiasis in peripheral hospitals and also at the field level where sophisticated immunodiagnostic facilities are not available." (Authors' abstract)
- 305 Makarova NG, Galko NV, Vashukova SS, Verbov VN, Gorbachev EN. [Use of Immunoenzyme analysis for the diagnosis of rotavirus gastroenteritis in children]. *Vopr Virusol* 1987 Jan-Feb;32(1):116-9
- 306 Makino Y, Maeda Y, Iwanaga T, Morita K, Igarashi A, Tazaki K, Fukunaga T. Development of class specific antibodies in sera following naturally acquired human rotavirus infections as measured by enzyme-linked immunosorbent assay (ELISA). *Trop Med* 1986 Dec;28(4):269-78
- 307 Mangali A, Chaicumpa W, Nontasut P, Chantavanij P, Tapchaisri P, Viravan C. Enzyme-linked immunosorbent assay for diagnosis of human strongyloidiasis. *Southeast Asian J Trop Med Public Health* 1991 Mar;22(1):88-92
- "Crude antigen (CA) was prepared from *Strongyloides stercoralis* filariform larvae obtained from *in vitro* culture of the human feces containing rhabditiform larvae. The lyophilized filariform larvae were ground and ultrasonicated in distilled water then the soluble antigenic preparation was delipidized. The protein content of the crude soluble antigen was 20% of the original dried larvae. The CA was passed through a gel filtration chromatography column and yielded three different protein fractions namely F₁, F₂ and F₃. CA and its fractions were used in the indirect enzyme-linked immunosorbent assay (ELISA) for detecting antibodies to *S. stercoralis* in serum samples of 5 groups of individuals. These were patients with parasitologically confirmed strongyloidiasis (group 1), patients with mixed *S. stercoralis* and other parasitic infections (group 2), non-strongyloidiasis patients with other worm infestation(s) (group 3), normal parasite-free Thais (group 4) and normal parasite-free Swedes (group 5). It was found that F₂ was the best antigen in the ELISA. The sensitivity, specificity and positive and negative

predictive values of the test using F_2 as the antigen were 95.0%, 96.4%, 95.0% and 96.4%, respectively." (Authors' abstract)

308 March SB, Ratnam S. Latex agglutination test for detection of *Escherichia coli* serotypes O157. *J Clin Microbiol* 1989 Jul;27(7):1675-7

309 Marchlewicz B, Spiwak M, Lampinen J. Evaluation of Abbott TESTPACK ROTAVIRUS with clinical specimens. *J Clin Microbiol* 1988 Nov;26(11):2456-8

310 Martin AL, Follett EA. An assessment of the sensitivity of three methods for the detection of rotavirus. *J Virol Methods* 1987 May;16(1-2):39-44

"A comparison was made of the sensitivity of a current commercial ELISA for detecting rotavirus in faecal specimens with the more complex, technically demanding systems of electron microscopy and polyacrylamide gel electrophoresis. Even after modification, the ELISA failed to detect 22% of specimens with particles identifiable by electron microscopy. Polyacrylamide gel electrophoresis failed to identify 2 out of 50 specimens with particles present but did distinguish 2 group C rotaviruses." (MEDLINE)

311 Martin AL, Kudesia G. Enzyme linked immunosorbent assay for detecting adenoviruses in stool specimens: comparison with electron microscopy and isolation. *J Clin Pathol* 1990 Jun;43(6):514-5

"A commercial enzyme linked immunosorbent assay (ELISA) for detection of adenoviruses in stool samples was compared with the use of electron microscopy and isolation in Graham 293 cells. Although specific, the ELISA was less sensitive than both electron microscopy and isolation. The ELISA had an overall sensitivity of 78% and a specificity of 100%. The sensitivity was related to the amount of virus particles present in the stool sample, increasing to 90% with about 10^7 viral particles/ml of stool. The ELISA was easy to perform, requiring no instrumentation, and is a useful first line test for detection of adenoviruses in stool samples, especially in laboratories without access to an electron microscope. Wider use of ELISAs should help in evaluating the role of adenoviruses in viral gastroenteritis." (Authors' abstract)

312 Mathai E, Jesudason MV. Coagglutination test in the diagnosis of typhoid fever. *Indian J Med Res* 1989 Sep;89:287-9

"Staphylococcal coagglutination (CoA) test for the detection of *Salmonella typhi* O (factor 9) antigen was evaluated as a diagnostic test in typhoid fever. Supernatants from 106 blood cultures with gram negative bacilli were subjected to CoA test. The sensitivity of the CoA test for the detection of *S. typhi* O antigen was 88 per cent and the specificity 97 per cent." (Authors' abstract)

313 Mathewson JJ, Winsor DK, Jr., DuPont HL, Secor SL. Evaluation of assay systems for the detection of rotavirus in stool specimens. *Diagn Microbiol Infect Dis* 1989 Mar-Apr;12(2):139-41

"We tested 41 rotavirus positive and 42 negative specimens as determined by electron microscopy. The assays systems used were an indirect NIH-ELISA, Meritec-Rotavirus, Virogen Rotatest, and Rotazyme II. Meritec and Virogen (latex agglutination assays) were the most sensitive tests, detecting 95% of the positive specimens. The NIH-ELISA detected 81% and Rotazyme detected 63%. Rotazyme was the most specific (100%), followed by the NIH-ELISA (95%) and the two latex agglutination systems (91%). To determine the level of rotavirus detection, we tested three systems against serial two-fold dilutions of ten positive stools. The NIH-ELISA detected rotavirus in an average dilution of 1:723. Rotazyme detected rotavirus in an average stool dilution of 1:366, and Meritec showed an average of 1:164. Rotavirus strains representing serotypes 1-4 were also tested. Meritec was able to detect all four serotypes. Virogen did not react with

serotype 2 strains. The NIH-ELISA and rotazyme were unable to detect serotype 3. These data suggest that some latex agglutination assays may be a useful alternative to ELISAs in the clinical laboratory." (Authors' abstract)

- 314 Mekara Y, Maneekarn N, Vithayasal V, Makonkawkeyoon S. Determination of antibody from typhoid patients against lipopolysaccharide and protein antigens of *Salmonella typhi*. *Asian Pac J Allergy Immunol* 1990 Dec;8(2):95-101

"Although the Widal test is simple, inexpensive and the most widely used for serodiagnosis of typhoid fever, the sensitivity and specificity of the test is sometimes doubtful. In this study, an enzyme-linked immunosorbent assay (ELISA) was developed for the detection of serum IgG and IgM antibodies to protein and lipopolysaccharide (LPS) antigens of *Salmonella typhi* which was compared with the Widal test in various groups of subjects. In typhoid patients with hemocultures positive for *S. typhi* (TP group), ELISA positivity was found on 100% for IgG anti-protein, 94.44% for IgG anti-LPS and 88.89% for IgM to both the protein and LPS antigens. In contrast, the Widal test was positive in only 61.11% for anti-O and 83.33% for anti-H antibodies. In healthy control subjects (HC group), only 5% of serum samples were positive for IgG anti-protein and none was positive for IgG anti-LPS or IgM to either the protein or LPS. In contrast, the Widal test was positive in 7.5% of HC group for anti-O and 17.5% for anti-H antibodies. In blood bank donors (BB group), both ELISA and Widal tests were positive in 23.40% of sera. Since the hospital records of BB group were incomplete. It might be possible that some of these subjects had recently been infected with *S. typhi*. Our data indicate that the standard Widal test was associated with false negative reactions in 16.39% of blood culture positive subjects. In non-typhoid patients with blood culture positive for bacteria other than *S. typhi* (OP group), most of the sera were negative by ELISA; 5% were positive for IgG anti-protein and 2.5% for IgM anti-protein and IgG anti-LPS, whereas 15% were positive on the Widal test for anti-O and 40% for anti-H antibodies. The final two groups of patients included in this study were those with a diagnosis of typhoid fever based on clinical signs and symptoms, in whom hemocultures were not done (WP group) or were negative (NP group). All of these subjects were positive for anti-O antibody by Widal test since this was the antigen for their diagnosis. The NP group had about 70% positive tests for IgG and IgM anti-protein and anti-LPS antigens by ELISA. The WP group showed lower percentage (28.44%) of sera positive for antibodies against protein and LPS antigens. Our data suggest that ELISA assays with *S. typhi* protein or LPS antigens have superior sensitivity and specificity to the Widal test in the diagnosis of typhoid fever and that the majority of individuals who are Widal seropositive but blood culture negative may not have typhoid fever. Further evaluation of the ELISA test is indicated." (Authors' abstract)

- 315 Melby K. Antibody response in patients infected with *Campylobacter jejuni/coli* assayed with a complement fixation test and a DIGELISA method [published erratum appears in NIPH Ann 1988 Dec;11(2):51] *NIPH Ann* 1987 Dec;10(2):29-31

"Two methods for the detection of antibodies against *Campylobacter jejuni/coli*, a complement fixation test (CFT) and a DIG-ELISA method, were tested against a panel of single sera from 36 coproculture positive patients. Twenty-five were positive in CFT compared to 31 in the other system. Twenty-three out of 25 CFT positive sera were positive in the ELISA method. Seven sera were positive only for IgA antibodies thus negative in the CFT. CFT may be useful for the detection of IgG/IgM antibodies against *C. jejuni/coli*. However, serum IgA may be a valuable marker for infections with *C. jejuni/coli*. Methods such as DIG-ELISA may identify such cases." (MEDLINE)

- 316 Merino E, Glender W, del Muro R, Ortiz-Ortiz L. Evaluation of the ELISA test for detection of *Entamoeba histolytica* in feces. *J Clin Lab Anal* 1990;4(1):39-42

"The clinical utility of an ELISA test with monoclonal antibodies to detect antigen of *Entamoeba histolytica* in feces was evaluated in 150 patients with gastrointestinal

symptoms. Each subject was examined by rectosigmoidoscopy with rectal smear and/or a triple stool search for ova-bacteria-parasite (OBP); in addition, one stool sample was collected for the ELISA test. All the tests were independent and double blind. *E. histolytica* was detected by OBP and/or rectosigmoidoscopy in 66 patients; 61 patients had other parasites; and in 23, no parasites were identified. Of all patients, 116 were positive for the ELISA test. Of these, *E. histolytica* was identified in 52. In 47, other parasites were identified and in 17, no parasites were found. The ELISA test with a monoclonal antibody against *E. histolytica* antigen showed higher sensitivity than the standard diagnostic methods: the ability to detect the presence of *E. histolytica* antigen regardless of the destruction of the parasite or of the error due to misidentification of the parasite resulting from faulty preparation of the samples." (MEDLINE)

317 Metzler J, Nachamkin I. Evaluation of a latex agglutination test for the detection of *Salmonella* and *Shigella* spp. by using broth enrichment. J Clin Microbiol 1988 Dec;26(12):2501-4

"We evaluated the Bactigen *Salmonella-Shigella* Latex Agglutination Slide Test (Wampole Laboratories, Div. Carter-Wallace, Inc., Cranbury, N.J.) for detection of *Salmonella* and *Shigella* spp. In enrichment broth cultures (gram-negative broth incubated for 24 h) as part of the routine testing of stool samples. A total of 1,128 stool samples were screened by using this test. Of 29 samples culture positive for *Salmonella* spp., 25 were positive with the *Salmonella* test (sensitivity, 86.2%; specificity, 96.2%). Of four stool samples culture positive for *Shigella* spp., two were detected with the *Shigella* latex reagent. Overall, the *Shigella* test had a specificity of 99.2%. Testing of enrichment broth cultures after 24 h of incubation was more sensitive than was testing after 6 h of incubation. When used for direct culture identification, both reagents had a specificity of greater than 98.0%. We conclude that the *Salmonella* test may be useful as an enrichment broth screening test to detect *Salmonella* spp.; however, we cannot make any conclusions about the *Shigella* test because of the low number of culture-positive specimens in this study." (Authors' abstract)

318 Midthun K, Pang L, Flores J, Kapikian AZ. Comparison of immunoglobulin A (IgA), IgG, and IgM enzyme-linked immunosorbent assays, plaque reduction neutralization assay, and complement fixation in detecting seroresponses to rotavirus vaccine candidates. J Clin Microbiol 1989 Dec;27(12):2799-804

319 Millotis MD, Galen JE, Kaper JB, Morris JG, Jr. Development and testing of a synthetic oligonucleotide probe for the detection of pathogenic *Yersinia* strains. J Clin Microbiol 1989 Jul;27(7):1667-70

320 Miotti PG, Viscidi RP, Elden J, Cerny E, Yolken RH. Centrifugation-augmented solid-phase immunoassay (CASPIA) for the rapid diagnosis of infectious diseases. J Infect Dis 1986 Aug;154(2):301-8

321 Miotti PG, Elden J, Yolken RH. Comparative efficiency of commercial immunoassays for the diagnosis of rotavirus gastroenteritis during the course of infection. J Clin Microbiol 1985 Nov;22(5):693-8

"We evaluated the performance characteristics of three commercially available immunoassays for the detection of rotavirus antigens in stool samples obtained from infants during the course of rotavirus gastroenteritis. Two of the assays, Bio-EnzaBead (Litton Bionetics, Charleston, S.C.) and Rotazyme (Abbott Laboratories, North Chicago, Ill.), are enzyme immunoassays, while the third, Rotalex (Medical Technology Corporation, Somerset, N.J.), is a latex agglutination assay. We tested a total of 122 samples obtained from 26 children with gastroenteritis; 56 samples, obtained from 21 children, were found to contain rotavirus antigen by a reference microplate enzyme immunoassay. Rotavirus antigen was found by the Bio-EnzaBead, Rotazyme, and Rotalex assays in 53, 42, and 29 samples, respectively. The true positivity of samples which were positive by the

reference microplate assay but negative by the other assays was confirmed by a specific neutralization assay or by the visualization of bands of double-stranded RNA by polyacrylamide gel electrophoresis or both. No false-positive assay results were noted with any of the commercial assays. The sensitivity of the assays was determined to a great extent by the time after the onset of illness at which the specimen was collected. Thus, the sensitivity of commercial assays with specimens collected early in the course of illness did not differ significantly from that of the microplate assay. However, significantly lower degrees of sensitivity were noted later in the course of illness. Results of our studies indicate that all three commercial assays can accurately detect rotavirus in stools from children with rotavirus gastroenteritis. However, the choice of assay systems for use in the clinical laboratory will depend on the conditions in which stool specimens are collected and tested in the laboratory." (Authors' abstract)

- 322 Miyamoto T, Miwa H, Hatano S. Improved fluorogenic assay for rapid detection of *Vibrio parahaemolyticus* in foods. *Appl Environ Microbiol* 1990 May;56(5):1480-4

"An improved fluorogenic assay for the rapid detection of *Vibrio parahaemolyticus* was developed. In the improved assay, the enrichment of *V. parahaemolyticus* was carried out in arabinose-glucuronate medium (0.5% arabinose, 0.25% glucuronate, 0.1% polypeptone, 0.1% yeast extract, 0.1% ammonium sulfate, 2% NaCl, 2µg of polymyxin B sulfate per ml, pH 8.5) at 37°C. After the cultivation, the trypsinlike activity of the bacteria was measured by fluorescence with the fluorogenic substrate benzoyl-L-arginine-7-aminomethyl-coumarin. Even in the presence of 3×10^5 cells of *Vibrio alginolyticus*, 20 cells of *V. parahaemolyticus* were clearly detected after a 6-h enrichment cultivation by the assay. Fifty contaminated samples of 14 seafoods were examined for *V. parahaemolyticus* by the fluorogenic assay after enrichment cultivation for 6 or 8 h. The results were then compared with those obtained by the conventional bromothymol blue Teepol agar assay and the most-probable-number method. There was a linear relationship between trypsinlike activity measured by the assay and the number of *V. parahaemolyticus* cells in seafood as determined by the bromothymol blue Teepol agar and most-probable-number methods. Correlation coefficients were 0.95 and 0.93 after a 6-h cultivation and an 8-h cultivation, respectively. The presence of 10 cells of *V. parahaemolyticus* per gram of seafood sample was detected after a 10-h total detection time by the fluorogenic assay." (Authors' abstract)

- 323 Moghaddam AAF, Katoull M, Parsi M. Comparison of Teknaf enteric agar and MacConkey/*Salmonella-Shigella* agar in evaluation of *Shigella* infection (letter). *Lancet* 1988 May 21;1(8595):1165-6

- 324 Mohimen A, Maitra TK, Jalan KN, Dhar A. A simple ELISA for anti-giardial antibody using the membrane fraction of the parasite as antigen. *Asian Pac J Allergy Immunol* 1985 Dec;3(2):183-6

"Most existing methods for detecting anti-giardial antibody require the use of cultured *Giardia lamblia* trophozoites as antigen in immunofluorescent and enzyme-linked immunosorbent assays. However, the maintenance of trophozoite culture systems limits the large-scale use of these antibody detection systems. An antigen extracted from *Giardia lamblia* by the detergent Triton X-100, when used in an ELISA system, produces specific, objective, quantitative and reproducible results. It is likely that such a test system could be packaged in kit form for large-scale diagnostic and epidemiological application in all parts of the world." (MEDLINE)

- 325 Monoclonal antibodies for therapy, prevention and *in vivo* diagnosis of human disease [meeting report]. *J Biol Stand* 1989 Oct;17(4):381-4

- 326 Monsur KA, Kay BA, Miah AL, Begum YA. An evaluation of the pooling method for detecting enterotoxigenic *Escherichia coli*. *J Diarrhoeal Dis Res* 1986 Dec;4(4):211-5

"The effectiveness of the common practice of using a mixture (pool) of bacterial colonies for detecting enterotoxigenic *Escherichia coli* (ETEC) was evaluated. Pools were constructed with *E. coli* whose phage lysotypes were known. After pooling and storage of isolates in the normal manner, attempts were made to recover and identify the original lysotypes in the pool. It was observed that bacteria from one or two colonies (lysotypes) almost always overgrew in the pool, thus reducing the number of recoverable individual strains. In general, enterotoxigenic *E. coli* (ETEC) did not show a survival advantage in the pools over non-ETEC strains. Therefore, the use of the pooling technique to enhance the likelihood of recovery of ETEC from faecal specimens is of doubtful value." (Authors' abstract)

327 Monsur KA, Miah AL, Huq MI, Huq F, Poddar G, Salek MA, Begum YA. Use of bacteriophage as a marker for identification of freshly-isolated individual *Escherichia coli* strains. *J Diarrhoeal Dis Res* 1985 Sep;3(3):131-7

"One or more 24 selected bacteriophages at 100 and 1000 times routine test dilution (RTD), spotted on lawns of individual *Escherichia coli* strains were found to attack over 90% of 297 *E. coli* strains tested. It was found that in most cases, it may be possible to use the phage susceptibility pattern of an individual *E. coli* strain as an identifying marker for that strain. In four cases, *E. coli* strains with the same phage pattern, isolated from different individuals, were found to belong to the same serogroup. Similarly, of 16 enterotoxigenic *Escherichia coli* (ETEC) isolates from 75 contacts of cases with watery diarrhoea, 7 were found to have the same phage pattern as ETECs from their respective index case. The toxigenic character of each pair was also found to be identical. The practicality of using phage patterns for studying the dynamics of *E. coli* population was demonstrated by its ability to detect changes in the *E. coli* population in a single patient." (Authors' abstract)

328 Moosal RB, Alcock R, Bell TM, Laidler FR, Peiris JSM, Wyn-Jones AP, Madeley CR. Detection of rotavirus by a latex agglutination test, Rotalex: comparison with electron microscopy, immunofluorescence, polyacrylamide gel electrophoresis, and enzyme linked immunosorbent assay. *J Clin Pathol* 1985 Jun;38(6):694-700

"A commercially available latex agglutination test, Rotalex (Orion Diagnostics, Finland), for detecting rotaviruses was evaluated in comparison with four other tests (electron microscopy, immunofluorescence, polyacrylamide gel electrophoresis, and enzyme linked immunosorbent assay) routinely used in our laboratories. Although Rotalex was the least complex method, it showed lack of specificity and sensitivity when carried out according to the manufacturer's instructions. Four basic modifications of Rotalex are described. These include the use of Hank's balanced salt solution, increasing the incubation time to 20 min, reading the agglutination result by an experienced observer, and the use of 50 mm square glass plates. The modified procedure gave results which were comparable with those obtained by electron microscopy, immunofluorescence, polyacrylamide gel electrophoresis, and enzyme linked immunosorbent assay. The latter techniques, when used to detect rotavirus, all gave similar results." (Authors' abstract)

329 Morotomi M, Hoshina S, Green P, Neu HC, LoGerfo P, Watanabe I, Mutai M, Weinstein IB. Oligonucleotide probe for detection and identification of *Campylobacter pylori*. *J Clin Microbiol* 1989 Dec;27(12):2652-5

330 Morris A, Ali MR, Brown P, Lane M, Patton K. *Campylobacter pylori* infection in biopsy specimens of gastric antrum: laboratory diagnosis and estimation of sampling error. *J Clin Pathol* 1989 Jul;42(7):727-32

331 Morris JA, Wells GAH, Scott AC, Sojka WJ. A comparison of methods for demonstrating colonization in the small intestine of piglets by enterotoxigenic *Escherichia coli*. *Br Vet J* 1985 Sep-Oct;141(5):484-9

- 332 Nacapunchal D, Tepmongkol M, Tharavanij S, Thammapalerd N, Subchareon A. A comparative study of four methods for detecting antibody in asymptomatic giardiasis. Southeast Asian J Trop Med Public Health 1986 Mar;17(1):96-100

"Four Immunological methods for diagnosis of giardiasis comprising complement fixation (CF) test, indirect hemagglutination (IHA) test, enzyme-linked immuno sorbent assay (ELISA) and lectin immuno test (LIT) were studied. Fifty sera from asymptomatic giardiasis patients, 40 from patients with other diseases and 50 from healthy controls were evaluated. The seropositive rates in asymptomatic giardiasis were 36% for CF, 58% for LIT, 30% for IHA and 72% for ELISA. The seropositive rates in patients with other diseases were 22.5% for CF, 52.5% for LIT, 12.5% for IHA and 67.5% for ELISA. The results suggest that the test of choice for giardiasis was CF with 88% specificity, nevertheless this test showed low sensitivity (36%). Other two tests, ELISA and LIT were more sensitive than CF with percent sensitivity of 72 and 58 respectively, but these two tests had severe disadvantages in being less specific with percentage specificity of 48 and 60 respectively." (Authors' abstract)

- 333 Nachamkin I, Barbagallo S. Culture confirmation of *Campylobacter* spp. by latex agglutination. J Clin Microbiol 1990 Apr;28(4):817-8

- 334 Nachamkin I, Lotz-Nolan L, Skalina D. Evaluation of a commercial cytotoxicity assay for detection of *Clostridium difficile* toxin. J Clin Microbiol 1986 May;23(5):954-5

- 335 Naess V, Hofstad T. Antigenicity of lipopolysaccharides from *Campylobacter jejuni* and *Campylobacter coli* in passive haemagglutination tests and enzyme-linked immunosorbent assays. Acta Pathol Microbiol Immunol Scand 1985;93C(2):97-104

"Passive haemagglutination tests and ELISA were used to study the serological activity of homologous and heterologous rabbit antisera against LPS prepared from various strains of *C. jejuni/coli*. In both test systems the homologous antisera exhibited serological activity against LPS. The heterologous antisera showed some degree of intra- and inter-species cross-reactivity. The cross-reacting was most pronounced in the ELISA. Erythrocytes sensitized with untreated LPS gave higher antibody-titres than erythrocytes sensitized with alkali-treated LPS in the haemagglutination tests." (Authors' abstract)

- 336 Nagano I, Ohtomo H, Masuda G, Tokoro M, Noda N, Goto K. [Fundamental studies on serological diagnosis of amoebiasis. 1. Application of amoebic antigen for CF test to ELISA]. Kansenshogaku Zasshi 1990 Jun;64(6):699-703

"We studied the establishment of enzyme-linked immunosorbent assay (ELISA) using amoebic antigen for complement fixation (CF) test. Optimal dilution for ELISA of sera from patients was 1:100, and that of CF-antigen was 1:400. The upper limit of the 99% critical range of the reaction of negative sera was 0.068 (cut-off level). Absorbance of sera from patients diluted 1:100 to antigen and antibody titers of ELISA were strongly correlated, so it was possible to estimate antibody titers from absorbance of serum diluted 1:100. ELISA and CF test were done to compare sensitivity of the tests using 63 sera from patients with invasive amoebic disease. The sensitivity of ELISA compared well with CF test (62 sera were positive by ELISA and 61 by CF test). Only one sample was both positive by ELISA and negative by CF test. This sample had low ELISA titers, so this discrepancy was mainly due to the sensitivity of CF test in detecting lower levels of antibody. These results suggested that the amoebic antigen for CF test can be applicable to ELISA, and this method was so sensitive and specific." (MEDLINE)

- 337 Nair GB, Saha MR, Sarkar BL, Pal SC. Comparison of the modified Elek test and Wagatsuma agar for determination of the Kanagawa phenomenon of *vibrio parahaemolyticus*. J Clin Microbiol 1985 Nov;22(5):868-9

The modified Elek test and Wagatsuma agar were compared for their ability to detect the Kanagawa activity of 142 *Vibrio parahaemolyticus* strains. The number of strains showing a positive Kanagawa reaction on Wagatsuma agar was slightly higher than in the modified Elek test. Thus, the performance of the modified Elek test was on a par with that of the Wagatsuma agar in terms of positivity. The modified Elek test, however, was far superior to Wagatsuma agar in effectively eliminating false positives and doubtful results. The results of the modified Elek test were not unduly influenced by the different types of agar used. The results indicated that Bacto-Agar and agarose were slightly better than the recommended Noble agar in terms of positive Kanagawa activity, while clarity of the precipitin bands was obtained from Noble agar and agarose. It is concluded that the modified Elek test, a simple gel diffusion technique, is a reliable substitute for the conventional Wagatsuma agar method for the detection of the Kanagawa activity of *V. parahaemolyticus*. An important advantage of this immunological method is that no fresh human blood is required.

338 Nakamura Y, Takeda A, Aoki Y, Sawada T, Yonekubo I, Tazawa S, Sato H. [Rapid detection of anti-*Campylobacter jejuni* antibodies by enzyme linked immunosorbent assay in the patients with acute enteritis (I)]. *Rinsho Byori* 1986 Apr;34(4):454-8

339 Nakamura Y, Fukuda I, Okumura K, Aoki Y, Tazawa S, Sawada T, Yonekubo I. [Rapid detection of *Campylobacter jejuni* by enzyme linked immunosorbent assay in the patients with acute enteritis (I)]. *Rinsho Byori* 1986 Mar;34(3):277-81

340 Nakata S, Estes MK, Graham DY, Wang S, Gary GW, Melnick JL. Detection of antibody to group B adult diarrhea rotaviruses in humans. *J Clin Microbiol* 1987 May;25(5):812-8

"Group B rotaviruses have been responsible for annual epidemics of severe diarrhea affecting both adults and children in China. We developed a specific and sensitive enzyme-linked immunosorbent blocking assay to detect antibody to group B rotaviruses that will be useful to assess the role of group B rotavirus infections as a cause of human gastroenteritis. We tested 219 human sera and 18 immunoglobulin pools collected from eight countries for antibodies to both group A and group B rotaviruses. Overall, a low proportion (10 of 237 or 4.2%) of sera contained antibody to group B rotaviruses. Antibody to group B rotavirus was detected in only 1 of 155 serum samples from healthy or hospitalized individuals in the United States, including patients with the chronic inflammatory bowel diseases Crohn's disease and ulcerative colitis. No antibody was detected in 15 serum samples from Australia and from an outbreak of gastroenteritis on a cruise ship or in nine immunoglobulin pools from Japan and the United Kingdom. Antibody to group B rotaviruses was detected in 8 convalescent- (but not acute-) phase serum samples from Chinese patients with group B gastroenteritis. In five immunoglobulin pools from China, in 1 of 6 serum samples from Chinese students in the United States, and in 1 each of 10 serum samples from Kenya, 20 from Thailand, and 15 from Canada. In contrast, most of these samples (226 of 237 or 95.4%) had antibody to group A rotaviruses. These results indicate that human infection with group B rotavirus has not been widespread in areas outside China. Seroconversion observed between the acute- and convalescent-phase serum samples from China also suggests that infections with this virus are primary infections. Continued surveillance for this new group of rotaviruses should determine whether the many susceptible people become infected or whether other factors influence the severe pathogenicity of human infections with these viruses in China." (Authors' abstract)

341 Nakata S, Estes MK, Chiba S. Detection of human calicivirus antigen and antibody by enzyme-linked immunosorbent assays. *J Clin Microbiol* 1988 Oct;26(10):2001-5

"Enzyme-linked immunosorbent assays (ELISAs) were developed to detect human

calicivirus (HCV) antigen and antibody to HCV. The ELISAs were specific for HCV and as sensitive as a previously developed radioimmunoassay. These ELISAs were used to search for evidence of HCV infection in the United States, where HCV gastroenteritis has rarely been reported. One hundred sixty-three stool samples collected from children hospitalized with diarrhea were examined; one sample was positive in the ELISA. Typical calicivirus particles were found in this stool sample, and these particles reacted with a hyperimmune guinea pig anti-HCV serum by immune electron microscopy. The age-related acquisition of antibody to HCV in hospitalized infants and children (from birth to 19 years old) without gastroenteritis and in healthy adults was also evaluated. The pattern of acquisition of antibody to HCV was similar to that for group A rotaviruses, namely, beginning in infancy and becoming 100% by the age of 4 years. These data suggest that HCV is associated with infantile gastroenteritis in the United States, that infections with HCV are common, and that many infections with HCV (Sapporo strain) may not require hospitalization." (Authors' abstract)

342 Narayanan J, Hartman PA, Graves DJ. Assay of heat-labile enterotoxins by their ADP-ribosyltransferase activities. *J Clin Microbiol* 1989 Nov;27(11): 2414-9

"Simple enzymatic assays to detect heat-labile enterotoxins whose modes of action are similar to that of cholera toxin were evaluated. The assays are performed by using an artificial substrate, diethylamino benzylidine-aminoguanidine, which is an ADP-ribose acceptor. The product, formed in the presence of NAD^+ , can be quantitated by spectrofluorometric, spectrophotometric, or high-performance liquid chromatographic (HPLC) methods. As little as 25 ng (spectrofluorometry) or 125 ng (spectrophotometry or HPLC) of cholera toxin can be detected in an assay volume of 250 μ l. The detection limit for heat-labile enterotoxin by either the spectrophotometric or HPLC methods was 125 ng/250 μ l. Because the results are quantitative, the enzymatic methods can be used for medium development, determination of factors that influence toxin production, and other applications that heretofore could be accomplished only with difficulty. The enzymatic methods add a new dimension to the assay of toxins that ribosylate arginine residues of proteins. Sensitivities of the assays might be improved by developing better synthetic substrates, and applications could be broadened by the development of artificial substrates containing other functional groups." (Authors' abstract)

343 Nardello S, Pizzella T, Migliaresi S, Galanti B. Performance characteristics of an enzyme-linked immunosorbent assay for the determination of immunoglobulin M anti-*Salmonella typhi* lipopolysaccharide antibodies. *Boll Ist Sieroter Milan* 1985;64(2):155-9

"Some characteristics of an enzyme-linked immunosorbent assay for the determination of immunoglobulin M anti-*Salmonella* lipopolysaccharide antibodies have been assessed. Immunoglobulin M anti-lipopolysaccharide antibodies in serum samples from most patients with typhoid fever cross-reacted with *S. typhimurium* lipopolysaccharide. However, absorbance values with *S. typhi* lipopolysaccharide were always higher than with the heterologous antigens. Serum samples from two patients with *S. paratyphi* B infection failed to show significant immunoglobulin M binding to *S. typhi* lipopolysaccharide. Serum samples from six rheumatoid arthritis patients were negative in this immunoenzymatic assay with *S. typhi* or *S. typhimurium* lipopolysaccharide as coating antigen. Finally, interference by specific immunoglobulin G in the immunoglobulin M anti-*S. typhi* lipopolysaccharide antibodies determination was excluded." (MEDLINE)

344 Nash TE, Herrington DA, Levine MM. Usefulness of an enzyme-linked immunosorbent assay for detection of *Giardia* antigen in feces. *J Clin Microbiol* 1987 Jul;25(7):1169-71

"The usefulness of a recently developed enzyme-linked immunosorbent assay which detects *Giardia lamblia* antigen in feces was determined in experimentally infected humans. *Giardia* antigen was determined in serially collected fecal specimens from humans inoculated with two *Giardia* isolates, GS/M and Isr. A total of 277 stools from 18

volunteers were tested, 74 from Isr-inoculated volunteers and 203 from GS/M-inoculated volunteers. None of the five Isr-inoculated volunteers became infected, and none of their stools contained *Giardia* antigen. In contrast, all of the 13 GS/M-inoculated volunteers became infected, and *Giardia* antigen was present one or more times in the stools of each. Of 203 stools from GS/M-inoculated volunteers, 73 contained *Giardia* cysts, and 69 of these (94.5%) had detectable antigen. In contrast, 108 of the 203 specimens were positive for *Giardia* antigen and only 73 had cysts. Most antigen-positive but cyst-negative specimens occurred during treatment, but during patency 71 stools contained antigen and 65 had cysts. The enzyme-linked immunosorbent assay is at least as sensitive as microscopic examination in diagnosing *Giardia* infections and is easier to perform." (Authors' abstract)

345 Nelmeister R, Logan AL, Egleton JH, Kleger B. Evaluation of direct wet mount parasitological examination of preserved fecal specimens. J Clin Microbiol 1990 May;28(5):1082-4

346 Newell DG. Monoclonal antibodies directed against the flagella of *Campylobacter jejuni*: cross-reacting and serotypic specificity and potential use in diagnosis. J Hyg (Camb) 1986 Jun;96(3):377-84

"Nine monoclonal antibodies directed against the flagella of *Campylobacter jejuni* strain 81116 have been investigated for serotypic and cross-reacting activity using a panel of 17 Penner serotype strains of *C. jejuni*. Four monoclonal antibodies were exclusively specific for serotype-6 strains, which was the serotype of *C. jejuni* strain 81116. Two monoclonal antibodies cross-reacted with all the flagellated strains of *C. jejuni* tested. One of these cross-reacting monoclonal antibodies, CF5, was found to react with all other *Campylobacter* species except *C. sputorum* but it did not react with other bacterial enteropathogens. An antigen-capture ELISA technique was established, using this monoclonal antibody, which could detect flagellar antigen in human faecal material. These anti-flagella monoclonal antibodies therefore may be valuable in the diagnosis and serotyping of *C. jejuni* in clinical material." (Authors' abstract)

347 Nishibuchi M, Ishibashi M, Takeda Y, Kaper JB. Detection of the thermostable direct hemolysin gene and related DNA sequences in *Vibrio parahaemolyticus* and other *Vibrio* species by the DNA colony hybridization test. Infect Immun 1985 Sep;49(3):481-6

"A specific gene probe for the *Vibrio parahaemolyticus* thermostable direct haemolysin gene was constructed and used to examine the presence or absence of the thermostable direct haemolysin gene or related DNA sequences in *V. parahaemolyticus* and other vibrios by the DNA colony hybridisation method. The gene probe consisted of a 406-base-pair, completely internal fragment covering 71% of the structural gene with PstI linkers added to the ends. Six copies of this 415-base-pair PstI fragment were cloned into plasmid pBR322, which yielded large amounts of the probe DNA. One hundred forty-one *V. parahaemolyticus* strains were tested with the gene probe, and the results were compared with those of phenotypic assays for the thermostable direct haemolysin. All Kanagawa phenomenon-positive strains were gene positive. However, 86% of the strains that exhibited weak Kanagawa phenomenon and 16% of Kanagawa phenomenon-negative strains also reacted with the gene probe. Immunological methods for the detection of the thermostable direct haemolysin [modified Elek test, enzyme-linked immunosorbent assay (ELISA)] showed better correlation with gene probe results. All gene-positive strains produced haemolysin detectable in the ELISA, although occasional strains showed weak reaction. The modified Elek test was slightly less sensitive than the ELISA. All gene-negative strains were also negative in these immunological assays. One hundred twenty-one strains of *Vibrio* spp. other than *V. parahaemolyticus* were tested with the gene probe; only strains of *V. holisae* reacted with the probe under stringent conditions. (Modified authors' abstract)

348 Nishikawa Y, Kishi T. A modification of bile salts brilliant green agar for isolation

of motile *Aeromonas* from foods and environmental specimens. *Epidemiol Infect* 1987 Jun;98(3):331-6

Aeromonas have recently been recognised as an important enteropathogen. In faecal specimens, the organism is readily identified and differentiated from coliforms by the positive oxidase reaction. However, few media have proved useful for food and environmental specimens. Bile salts brilliant-green starch agar was studied for this purpose by testing artificially contaminated foods as well as natural potential sources. The results indicate the suitability of the medium for the isolation of *Aeromonas* from foods and from environmental sources. (Modified authors' abstract)

349 Nishikawa Y, Hase A, Ishii E, Kishi T. Screening of aquatic samples for *Vibrio cholerae* serotype O1 by a dot-blot method and a latex agglutination test. *Appl Environ Microbiol* 1990 Jun;56(6):1547-50

"A dot-blot, enzyme-linked immunosorbent method and a latex agglutination test were studied for their abilities to detect *Vibrio cholerae* serotype O1 in aquatic samples by testing artificially contaminated water as well as samples from natural potential sources. Water samples were pre-enriched with alkaline peptone and then enriched with Monsur peptone water. For the dot-blot test, enriched cultures of organisms in a small portion of the Monsur peptone water were transferred to a polyvinylidene difluoride membrane with a microfiltration apparatus. The enzyme-linked immunosorbent assay was performed by using biotin-labeled antibodies and avidin-biotin-peroxidase complex; brown dots developed in the wells that contained serotype O1 vibrios. Latex agglutination tests were performed by mixing 1 drop of the culture in Monsur with 1 drop of reagent coated with monoclonal antibody specific for antigen A. The sensitivities and specificities of the methods were compared with those of the colony-blot method, which identified individual colonies of *V. cholerae* O1 in mixed bacterial cultures on isolation media. Our results indicate that the dot-blot method is as sensitive as the colony-blot method and is useful for screening for *V. cholerae* serotype O1 even in specimens that are heavily contaminated with non-O1 vibrios." (Authors' abstract)

350 Nishio O, Ooseto M, Takagi K, Yamasita Y, Ishihara Y, Isomura S. Enzyme-linked immunosorbent assay employing monoclonal antibodies for direct identification of enteric adenoviruses (Ad40,41) in feces. *Microbiol Immunol* 1990;34(10):871-7

"For detection and identification of enteric adenovirus (Ad) types 40 and 41 in stool specimens, enzyme-linked immunosorbent assay (ELISA) was developed with the use of three monoclonal antibodies: Ad group-specific, Ad40 type-specific, and Ad41 type-specific antibodies. Of 860 fecal samples from patients with acute gastroenteritis, 44 strains of Ad were isolated using Graham 293 cell cultures. Of these isolates, 20 were typed as Ad40, 18 were Ad41, and 6 were other Ads by neutralization tests with cell cultures. Results of the ELISA tests on these 860 fecal samples resulted in good agreement to those with the cell culture method. The ELISA tests using Ad type-specific monoclonal antibodies proved to be a specific and rapid technique for laboratory diagnosis of acute gastroenteritis caused by enteric Ads." (Authors' abstract)

351 Nordlander E, Phuphalsan S, Bodhidatta L, Arthur J, Echeverria P. Microscopic examination of stools and a latex slide agglutination test for the rapid identification of bacterial enteric infections in Khmer children. *Diagn Microbiol Infect Dis* 1990 May-Jun;13(3):273-6

"Eighty-five stools collected from 50 children with diarrhea at an evacuation site on the Thai-Kampuchean border were (1) examined microscopically for fecal leukocytes, (2) tested after 24 hr enrichment in brain/heart infusion broth by a latex slide agglutination test for detection of *Salmonella* and *Shigella*, and (3) examined with microbiological techniques to identify bacterial, viral, and parasitic pathogens. If the 65 specimens in which one or no pathogens are considered, 6 or more fecal leukocytes/hpf were found

on microscopic examination of stools in both children infected with *Shigella* spp., the one child infected with *Salmonella* spp., and three of eight children infected with *Campylobacter* spp. ≤ 5 leukocytes/hpf were found in 70% (7/10) of children infected with rotavirus, 100% (2/2) infected with *Cryptosporidium*, 100% (2/2) infected with *Giardia*, 89% (8/9) infected with enterotoxigenic *Escherichia coli*, and 77% (24/31) with diarrhea in whom no etiologic agent was identified. The *Salmonella* slide latex test had a sensitivity of 50%, a specificity of 92%, and a positive predictive value of 12%. The *Shigella* slide latex test had a sensitivity of 0%, a specificity of 95%, and a positive predictive value of 0%. Forty-five percent of the latex slide agglutination tests from enrichment cultures were nonspecific. Microscopic examination of diarrheal stools for fecal leukocytes, though nonspecific, appears to be the best way to differentiate *Shigella* spp. from rotavirus and parasitic infections. Examining stools for fecal leukocytes was less helpful in differentiating *Shigella* from other bacterial infections." (Authors' abstract)

352 Novara A, Oddone M, Musso A, Riva C, Cagliero T, Canese MG, Nigro N, Brunet. [Immunoenzymes and electron microscopy in the diagnosis of rotavirus infections in childhood]. *Minerva Pediatr* 1985 Oct 31;37(20):773-7

353 Nys M, Damas P, Damas F, Joassin L, Demonty J. A direct enzyme-linked immunosorbent assay (ELISA) for antibodies to enterobacterial Re core glycolipid and lipid A. Results in healthy subjects and in patients infected by Gram-negative bacteria. *Med Microbiol Immunol (Berl)* 1987;176(5):257-71

"We have developed an ELISA for IgM and IgG antibodies to the core glycolipid (CGL) of the Re mutant *Salmonella minnesota* R 595, and to lipid A. Anti-CGL antibodies have been detected in sera from 37% of healthy blood donors, whereas anti-lipid A activities were found in 13% of individuals only. The anti-CGL and anti-lipid A activities were examined in patients in a surgical intensive care unit, selected on the basis of a definite risk of infectious complications due to Gram-negative bacteria. Of the patients who developed such infections, the rate of favourable outcome was significantly higher in patients with either stable positive or increasing anti-CGL activities than in patients found to be negative. Our results provide clear evidence that anti-CGL antibodies contribute to host defence against various Gram-negative bacteria." (MEDLINE)

354 Obgolt's AA, Klishevich VP, Televnala LG. [Serological diagnosis of typhoid infection]. *Zh Mikrobiol Epidemiol Immunobiol* 1985 Jun;107(6):103-6

355 Ofek I, Courtney HS, Schifferl DM, Beachey EH. Enzyme-linked immunosorbent assay for adherence of bacteria to animal cells. *J Clin Microbiol* 1986 Oct;24(4):512-6

"Epithelial cells scraped from human oral mucosa and from pig intestines were immobilized onto the flat bottom surfaces of microtiter plates to study the adherence of various bacterial species to host cells. Bacterial adherence was quantitated either by an enzyme-linked immunosorbent assay technique with specific antibacterial serum as the first antibody followed by peroxidase-conjugated second antibody or by using biotinylated bacteria and avidin-peroxidase as the detecting agent. Unlabeled *Escherichia coli* and purified *E. coli* 987P fimbriae inhibited the adherence of biotinylated *E. coli* to immobilized enterocytes. The adherence of a mannose-sensitive strain of *E. coli* to immobilized oral epithelial cells was inhibited by mannose derivatives. The adherence of fimbriated *E. coli* 987P to immobilized enterocytes was approximately four times higher than the adherence of a nonfimbriated variant of the same strain. The adherence of *Streptococcus pyogenes* to oral cells was detected in the range of 10 to 150 bacteria per cell and was inhibited by lipoteichoic acid and albumin. The data suggest that the putative receptors which bind bacteria on the immobilized cells retain a functional form similar to that of native cells in suspension. The proposed adherence assay is easy to perform, allows the detection of specific adherence of test bacteria, and provides objective quantitation of adherence with a sensitivity of 10 bacteria per cell. Most importantly, the assay allows the testing of many variables in the same day." (Authors' abstract)

- 356 Olive DM, Sethi SK. Detection of human rotavirus by using an alkaline phosphatase-conjugated synthetic DNA probe in comparison with enzyme immunoassay and polyacrylamide gel analysis. *J Clin Microbiol* 1989 Jan;27(1):53-7
- "An alkaline phosphatase-conjugated synthetic oligodeoxynucleotide probe was compared with polyacrylamide gel electrophoresis (PAGE) detection of rotavirus RNA as well as an enzyme-linked immunosorbent assay (ELISA) for the detection of rotavirus in stools from young children with gastroenteritis. The synthetic probe did not cross-react with bacterial causative agents of diarrheal disease. Extraction of viral RNA from stool samples with a phenol-chloroform mixture was suitable for most samples. In some cases fecal pigments interfered with the reaction of the probe with viral RNA. The use of ion-exchange chromatography to further purify viral RNA removed contaminating pigments and increased the sensitivity of the probe assay. Of 260 stool specimens, 77(30%) were positive for rotavirus when tested by PAGE analysis of rotavirus RNA. The synthetic probe identified 71 rotavirus specimens when RNA obtained by phenol-chloroform extraction followed by chromatographic purification was used (sensitivity, 91.0%; specificity, 96.7%). The ELISA results also agreed well with the electrophoretic analysis (sensitivity, 98.7%; specificity 94%) and the probe assay (sensitivity, 90%; specificity, 100%). Discordant results between the ELISA and the probe assay were examined further by electron microscopy and PAGE analysis of viral RNA. The positive and negative predictive values of the probe assay in comparison with PAGE were 92.2 and 96.1%, respectively. Rotaviruses showing both long and short RNA electrophoretic patterns were detected by the probe. The probe assay coupled with chromatographic purification of rotavirus RNA is an effective method for detecting rotavirus and compares favorably with PAGE analysis and ELISA." (Authors' abstract)
- 357 Olive DM, Khalik DA, Sethi SK. Identification of enterotoxigenic *Escherichia coli* using alkaline phosphatase-labeled synthetic oligodeoxynucleotide probes. *Eur J Clin Microbiol Infect Dis* 1988 Apr;7(2):167-71
- 358 Onuigbo MAC. Diagnosis of typhoid fever in Nigeria: misuse of the Widal test. *Trans R Soc Trop Med Hyg* 1990 Jan-Feb;84(1):129-31
- 359 Oosterom J, den Uyl CH, Banffer JR, Lauwers S, Huisman J, Busschbach AE, Poelma FG, Bellemans R. Evaluation of an enzyme-linked immunosorbent assay (ELISA) for the detection of *Campylobacter jejuni* antibodies, and comparison with a complement fixation test (CFT). *Antonie Van Leeuwenhoek* 1985;51(3):321-31
- "An enzyme-linked immunosorbent assay (ELISA) was developed for the detection of total anti-*Campylobacter* immunoglobulins in human sera. In this assay disintegrated *Campylobacter* bacteria were used as the antigen. Absorption tests including other possibly enteropathogenic bacterial species showed that the ELISA system displayed a high immunological specificity for *Campylobacter*. Using this ELISA it was found that in about 80% of *Campylobacter* patients these *Campylobacter* antibodies are produced to almost maximal levels within 8 days after onset of disease, and that they may persist for at least 4 months. Indeed, *Campylobacter* antibodies were demonstrated at low levels in a large number of control sera. However, accepting an antibody titre of 1:640 as indicative of *Campylobacter* infection, the statistical sensitivity of the ELISA system was 77% and the specificity 95%. In an epidemiological survey a high association was demonstrated between the severity of *Campylobacter*-related symptoms and antibody titre values. Assessment of *Campylobacter* antibody titres by means of this ELISA and by a complement fixation test in 92 sera from index patients and contacts with and without symptoms showed a high association of results." (MEDLINE)
- 360 Otto P, Vogt KH, Prudlo J. [Rotavirus infection of the calf. 2. Use of enzyme immunoassay in the detection of virus antigen in fecal samples]. *Arch Exp Veterinarmed* 1987 Nov;41(6):939-43

- 361 Overman TL, Kessler JF, Seabolt JP. Comparison of API 20E, API Rapid E, and API Rapid NPT for identification of members of the family *Vibrionaceae*. *J Clin Microbiol* 1985 Nov;22(5):778-81
- "Sixty isolates, from nine species of the family *Vibrionaceae*, were tested by the API 20E, API Rapid E, and API Rapid NPT systems. Results were compared with those obtained with standard biochemicals. Included were 7 *Aeromonas caviae* isolates, 27 *Aeromonas hydrophila* isolates, 10 *Aeromonas sobria* isolates, 3 *Plesiomonas shigelloides* isolates, 5 *Vibrio alginolyticus* isolates, 3 *Vibrio cholerae* isolates, 1 *Vibrio fluvialis* isolate, 5 *Vibrio parahaemolyticus* isolates, and 1 *Vibrio vulnificus* isolate. The API 20E correctly identified all the isolates within 24 h. The API Rapid E correctly identified only 77%, misidentified 8%, and failed to identify 2% of the isolates in 4 h. The remaining 13% of the isolates gave a low selectivity identification, with one of the choices being correct. The API Rapid NPT correctly identified 87%, misidentified 5%, gave a low selectivity identification for 8% of the isolates, and in some instances, required up to 48 h of incubation. The API 20E is a valid system for use in the identification of the more commonly occurring members of the family *Vibrionaceae* and the most accurate and efficient of the three systems tested." (Authors' abstract)
- 362 Pacini DL, Brady MT, Budde CT, Connell MJ, Hamparian VV, Hughes JH. Polyclonal gel electrophoresis of RNA compared with plasmid- and monoclonal-antibody-based enzyme immunoassays for rotavirus. *J Clin Microbiol* 1988 Feb;26(2):194-7
- 363 Paerregaard A, Hjelt K, Krasilnikoff PA. Comparative study of four methods for detecting glandularis in children. *Pediatr Infect Dis J* 1988 Nov;7(11):807-9
- 364 Pahuchonma W, Janitschke K. Serodiagnosis of parasitic infections by ELISA with different antigens. *Southeast Asian J Trop Med Public Health* 1987 Jun;18(2):193-6
- "An enzyme-linked immunosorbent assay is described in which four antibodies (amoebalysin, schistosomiasis, echinococcosis and filariasis) can be tested at once. Because of the sensitivity, specificity, reproducibility and practicability this test system can be recommended as a quantitative routine test." (Authors' abstract)
- 365 Pal CH, Shahabadi MS, Ince B. Rapid diagnosis of rotavirus gastroenteritis by a commercial latex agglutination test. *J Clin Microbiol* 1985 Nov;22(5):846-50
- "The Rotalex test, a commercial latex agglutination test for rotavirus, was compared with immunocassay, for detection of rotavirus in stools of children and neonates. For initial stool specimens from 265 children <3 years old with diarrhea, the Rotalex test had a sensitivity of 81.7% and specificity of 99.5% compared with EM results. Positive and negative predictive values were 98 and 94.9%, respectively. The Rotalex test was slightly more sensitive and specific than the Rotazyme test. When daily stool specimens from patients with rotavirus gastroenteritis were examined, the sensitivity of the Rotalex test varied depending on the time of stool collection relative to the onset of symptoms. Sensitivity was 100 (20/20), 96 (23/24), and 54% (7/13) during 1 to 4, 5 to 7, and 8 to 18 days, respectively, after the onset of symptoms. The sensitivity of the Rotazyme test varied similarly with days from onset. We also examined 214 EM-negative stool specimens from asymptomatic newborns. False positivity by the Rotalex test was only 3.3% (7/214) compared with 4.2% (9/215) for the Rotazyme test. The Rotalex test was as sensitive and specific as EM for detection of rotavirus during the acute stage of illness and much faster and cheaper than EM or the Rotazyme test. The test appears to be suitable for routine use in small hospitals, emergency wards, or even the physician's office for rapid diagnosis of rotavirus gastroenteritis." (Authors' abstract)
- 366 Pal CH, Mayock D. Rotazyme test in neonates [letter]. *J Pediatr* 1985 Feb;106(2):343-4

- 367 Pak SG, Voronov AV, Gurskii IUN, Malov VA, Tur'ianov MKh. [Use of immunoenzyme analysis in the rapid diagnosis and study of the characteristics of circulating O-antigen in biological fluids of patients with acute dysentery Sonne]. Zh Mikrobiol Epidemiol Immunobiol 1969 Jun(6):77-81

"The data on the clinical approval of the original enzyme immunoassay system for the determination of somatic O-antigen in the blood serum and urine of patients with acute dysentery are presented. The level of the antigen determined in the biological fluids of patients has been shown to depend on the severity of the disease. Different types of dynamic curves, reflecting the level of O-antigen in the biological fluids of patients with acute Sonne dysentery and characteristic of different clinical forms of the disease, have been established." (MEDLINE)

- 368 Pal T, Emody L, Pasco AS. ELISA detectable virulence marker antigen of enteroinvasive *Escherichia coli* is coded by a 140 megadalton plasmid. Acta Microbiol Hung 1986;33(4):341-4

- 369 Pal T, Echeverria P, Taylor DN, Sethabur O, Hanchalay S. Identification of enteroinvasive *Escherichia coli* by indirect ELISA and DNA hybridisation [letter]. Lancet 1985 Oct 5;2(8458):785

- 370 Pal T, Pasco AS, Emody L, Voros S, Selley E. Modified enzyme-linked immunosorbent assay for detecting enteroinvasive *Escherichia coli* and virulent *Shigella* strains. J Clin Microbiol 1985 Mar;21(3):415-8.

"Immune sera were produced in rabbits with living cells of an enteroinvasive O143 strain derivative of the same strain. In the enzyme-linked immunosorbent assay, absorbed sera reacted specifically with only virulent *Shigella* strains and enteroinvasive *E. coli* strains of different geographical origin, regardless of species or serogroups. The investigation of 83 strains indicated complete agreement between enzyme-linked immunosorbent assay results and those of the keratoconjunctivitis test. It is assumed that the absorbed immune sera reacted with a possible virulence marker antigen. This inexpensive and simple method provides an alternative to other virulence tests. It has a definite advantage for screening large number of isolates within 24 h." (Authors' abstract)

- 371 Panda CS, Riley LW, Kumar SN, Khanna KK, Prakash K. Comparison of alkaline phosphatase-conjugated oligonucleotide DNA probe with the Serey test for identification of *Shigella* strains. J Clin Microbiol 1990 Sep;28(9):2122-4

"We compared an alkaline phosphatase-conjugated oligonucleotide DNA probe with the Serey test to determine the sensitivity and specificity of the probe in detecting virulent *Shigella* strains. The probe hybridized with all 52 Serey-test-positive strains (sensitivity, 100%) and 4 of 21 Serey-test-negative strains (specificity, 81%). The probe did not hybridize with any of the Serey-test-negative *S. dysenteriae* type 1 strains. This nonradioactive, synthetic probe provides a simple, rapid way to test a large number of strains simultaneously in a field setting, which will contribute to an improved understanding of the epidemiologic patterns of shigellosis in developing countries." (Authors' abstract)

- 372 Panigrahi D, Agarwal KC, Dubey ML, Samantarya JC. Comparison of three tests for the detection of rotavirus in feces with standard ELISA. J Diarrhoeal Dis Res 1986 Mar;4(1):26-9

"For the detection of rotavirus in feces three tests -- Rota SPAC and Rota-Wellico -- were compared with a standard double-sandwich enzyme-linked immunosorbent assay (ELISA). Fecal samples from 500 apparently healthy and 250 diarrhoeic children (below 5 years) attending an Indian hospital were first tested by a

double-Sandwich ELISA for the presence of rotavirus. One hundred and four positive and 200 negative control samples were further examined using Rotazyme, Rota SPACE and Rota-Wellico tests. Seventy-eight percent (237/304) samples showed consistent results in all four tests. Rota SPACE showed a specificity and sensitivity similar to that of ELISA. The Rotazyme test showed 14% false positive reactions, while the Rota-Wellico test failed to detect the virus in 37.5% of ELISA-positive samples. It is concluded that ELISA or the Rota SPACE test can be used for rapid detection of rotavirus diarrhoea." (Authors' abstract)

373 Panigrahi D, Burks M, Hartharajan H, Finkelstein RA. Evaluation of immuno-dot-blot assay for detection of cholera-related enterotoxin antigen in *Salmonella typhimurium*. J Clin Microbiol 1987 Apr;25(4):702-5

Twenty-five strains of *Salmonella typhimurium* isolated in India were examined for the presence of cholera/coli-related enterotoxin antigen by a previously described latex particle agglutination test and by a newly developed immuno-dot-blot test, using immunopurified goat antibody against the cholera-related enterotoxin isolated from an *Escherichia coli* strain of human origin. The immuno-dot-blot assay could detect 0.02 ng of purified enterotoxin. The amount of toxin antigen detected varied widely from strain to strain. Fourteen of the 25 polymyxin B-treated extracts of bacteria harvested from 6-hour casamino acids-yeast extract broth cultures gave positive results in both serologic assays as well as in rabbit skin tests for delayed permeability factor. An additional strain was positive only in the immuno-dot-blot. Five of the 6 stool isolates and 6 of the 7 blood isolates tested gave positive reactions. Two isolates of *Salmonella enteritidis* tested were also positive. The immuno-dot-blot test appears to be a simple, rapid, and reliable method for detection of cholera-related enterotoxin antigen in *S. typhimurium*. The demonstration of a cholera-related enterotoxin, even in small amounts, in a facultative intracellular pathogen raises interesting questions regarding its potential role in pathogenesis, both of diarrhoeal disease and systemic infections caused by salmonellae. (Modified authors' abstract)

374 Panigrahi D, Dubey ML, Agarwal KC. Evaluation of three *in vitro* methods for the detection of *Esch. coli* heat-labile enterotoxin. Indian J Med Microbiol 1985 Jul;3(3):201-6

The GM₁ enzyme-linked immunosorbent assay (ELISA) was compared with staphylococcal co-agglutination test and the Biken test for the detection of heat-labile enterotoxin (LT) of *Escherichia coli* isolated from human fecal samples. A total of 650 *E. coli* strains isolated from stools of 260 diarrhoeic children aged below 5 attending an Indian hospital were investigated. Toxins were extracted from all these strains. All the 3 *in vitro* assays were performed on 650 strains. GM₁ ELISA proved to be the most sensitive and specific test. The Biken agar test also gave a result comparable with that of GM₁ ELISA. Co-agglutination was less specific and sensitive in comparison to the other two tests. GM₁ ELISA and co-agglutination, GM₁ ELISA and the Biken agar test gave consistent results in 92.3% (600/650) and 99.1% (644/650) cases respectively. GM₁ ELISA was recommended as a highly effective test for detecting *E. coli* LT in a routine diagnostic laboratory. However, for epidemiological and field studies, the Biken agar test is more appropriate, since the method is simple, inexpensive and does not require any toxin extraction.

375 Pedler SJ, Orr KE. ACP broadsheet 124: examination of faeces for bacterial pathogens. J Clin Pathol 1990 May;43(5):410-5

376 Pena AS, Endtz HP, Offerhaus GJ, Hoogenboom-Verdegaaal A, van Duin W, de Vargass N, den Hartog G, Kreunung J, van der Heyden J, Mouton RP, et al. Value of serology (ELISA and immunoblotting) for the diagnosis of *Campylobacter pylori* infection. Digestion 1989;44(3):131-41

"Fifty-two unselected patients referred to for upper gastrointestinal endoscopy were evaluated in several ways to determine the presence of *Campylobacter pylori*. Antibodies against this microorganism were measured to assess the value of serology for the diagnosis of *C. pylori* infection. Five antral biopsy specimens were taken in each patient for culture and bacteriological determinations, histology [morphology and Warthin-Starry (WS) staining] and the urease test (2, 3 and 24 h). Serum antibodies against a sonicate of 6 strains of microorganisms were assayed by enzyme-linked immunosorbent assay (ELISA) and an immunoblotting technique. In 14 of the 52 patients the histology of the antrum was normal, 18 patients had chronic active gastritis and 20 had chronic gastritis without polymorphonuclear infiltration. In the group with normal histology, only 1 patient was positive for *C. pylori* with all methods, and 1 other subject was positive for IgG and 2 for IgA only with ELISA. In the group with chronic active gastritis, 14 were positive with all methods, 1 was negative by WS only and another was negative for IgA according to ELISA, WS and antibodies. Among the patients with chronic gastritis, 7 were positive and 7 negative with all tests; in the other 6 patients the results obtained with the various tests were divergent. Four serological tests were studied and validated against culture, WS and urease test which were considered to be the reference methods. The serological tests showed high sensitivity and specificity for the detection of *C. pylori*-associated active chronic gastritis of the antrum, and can therefore serve as noninvasive methods to identify individuals with this condition." (MEDLINE)

377 Peralas I, Audicana A. Semisolid media for isolation of *Salmonella* spp. from coastal waters. Appl Environ Microbiol 1989 Nov;55(11):3032-3

378 Pereira HG, Azevedo RS, Leite JP, Andrade ZP, De Castro L. A combined enzyme immunoassay for rotavirus and adenovirus (EIRA). J Virol Methods 1985 Jan;10(1):21-8

"A combined enzyme immunoassay for rotavirus and adenovirus (EIRA) was developed as a double-antibody sandwich assay in which test samples are added to plastic wells coated with rotavirus or adenovirus goat antibody. The presence of antigens is detected by mixed-guinea pig antisera to the same viruses followed by rabbit anti-guinea pig IgG conjugated with peroxidase and subsequently by ortho-phenylenediamine substrate. Titrations of rotavirus (SA11) and adenovirus type 2 tissue culture grown viruses in the presence of separate capture sera and mixed detector sera revealed that the two virus-antibody reactions occurred independently and were not affected by each other. Comparison with another enzyme immunoassay for rotavirus, with immunoelectron microscopy and with polyacrylamide gel electrophoresis showed that EIRA is a sensitive and specific method for detecting rotaviruses and adenoviruses in faeces from children with gastroenteritis." (MEDLINE)

379 Pereira LP, Marques LRM, O'Brien AD. Isolation and characterization of monoclonal antibodies to Shiga-like toxin II of enterohemorrhagic *Escherichia coli* and use of the monoclonal antibodies in a colony enzyme-linked immunosorbent assay. J Clin Microbiol 1988 Oct;26(10):2127-31

"The major obstacle in large-scale epidemiological investigations of the incidence of Shiga-like toxin (SLT)-producing *Escherichia coli* in diarrheal stools is the lack of a rapid, specific test to detect toxin. Enterohemorrhagic *E. coli* produces elevated levels of SLT-I, SLT-II, or both cytotoxins (also called Verotoxins). SLT-I but not SLT-II can be neutralized by antiserum to purified Shiga toxin and by monoclonal antibodies to the B subunit of SLT-I. In this study, monoclonal antibodies were generated against a crude preparation of SLT-II produced by an *E. coli* K-12 strain lysogenized with the 933W toxin-converting phage of enterohemorrhagic *E. coli* 933. Hybridoma culture supernatants were screened for anti-SLT-II antibodies by a cytotoxicity neutralization assay and by an enzyme-linked immunosorbent assay (ELISA). Of 53 ELISA-positive lines, 5 were capable of neutralizing the cytotoxicity of SLT-II but not of SLT-I. Shiga toxin, or a variant of SLT-II produced by *E. coli* that causes edema disease of swine,

All five monoclonal antibodies immunoprecipitated the isolated A subunit of SLT-II but not the B subunit. Of these five neutralizing monoclonal antibodies, four were of the immunoglobulin M class and one belonged to the immunoglobulin G1 subclass. All five lines had k light chains. These neutralizing monoclonal antibodies have been used as probes in a colony ELISA to detect SLT-II-positive bacterial colonies. The colony ELISA with these monoclonal antibodies is a specific, sensitive test with potential diagnostic value." (Authors' abstract)

380 Peterson LR, Hotter JJ, Shanholzer CJ, Garrett CH, Gerdling DN. Detection of *C. difficile* toxin A (enterotoxin). Am J Clin Pathol 1987 Feb;87(2):298-9

381 Peterson LR, Olson MM, Shanholzer CJ, Gerdling DN. Results of a prospective, 18-month clinical evaluation of culture, cytotoxin testing, and culture brand (CDT) latex testing in the diagnosis of *Clostridium difficile*-associated diarrhea. Diagn Microbiol Infect Dis 1988 Jun;10(2):85-91

"An 18-month evaluation of culture, cytotoxin, and latex testing for *Clostridium difficile* was performed between July 1, 1985, and December 31, 1986, on 1,536 specimens from 1,406 patients during evaluation of diarrhea. All cases with at least one test positive were investigated for clinical status. There were 144 *Clostridium difficile*-associated diarrheas (CAD) patients; 139 (97%) were positive by culture, 96 (67%) by cytotoxin, and 98 (68%) by latex testing. In the 1,262 non-CAD patients with diarrheal stool, 89 (7.1%) were positive by culture, 18 (1.4%) by cytotoxin, and 68 (5.4%) by the latex test. No CAD patient was positive by cytotoxin testing only, and two were positive by latex testing only. The culture and cytotoxin positivity were similar to our previous reports of 90-97% and 70-73%, respectively. Latex sensitivity (68%) was comparable to that of cytotoxin testing in this large group of patients ($p < 0.5$). Overall, in the 1,262 patients without clinical evidence of *Clostridium difficile* disease, positive tests by latex testing (5.4%) were intermediate between those of culture (7.1%, $p < 0.1$) and cytotoxin (1.4%, $p < 0.001$). (Authors' abstract)

382 Pinon J-M, Poiriez J, Remy G, Lapan H. Immunological studies in amebiasis: isotypic characterization of specific antibodies by enzyme-linked immunofiltration assay. Am J Trop Med Hyg 1987 Sep;37(2):290-5

"Fifty sera from 17 cases of invasive amebiasis (15 hepatic localizations, 1 ameboma, 1 diffused colic localization) were studied by enzyme-linked immunofiltration assay (ELIFA). IgM and IgE anti-*Entamoeba histolytica* were detected in 10 and 15, respectively, of the 17 patients studied. IgM and IgE antibodies were found simultaneously in 10 cases; these 2 isotypes were only recognized twice in the same serum by the same antigenic components. During post-therapy monitoring, in less than 90 days IgE or IgM-Ab re-gressed completely in half the cases. If they appeared or reappeared after the third month, prognosis was bad. In addition to its value for diagnosis and prognosis, the ELIFA allowed us to detect the functional antigenic components revealed more particularly by some IgG, IgM, IgE, or IgA antibodies." (Authors' abstract)

383 Pitirangs C, Echeverria P, Taylor DN, Johnson S. Media for the isolation of *Shigella* [letter]. J Diarrhoeal Dis Res 1987 Mar;5(1):42

384 Pokrovskii VI, Abalakin VA. [Use of immunological adsorption in a heterologous immunoenzyme analytical system for detecting specific antibodies to serogroup B and D1 *Salmonella* lipopolysaccharides in healthy donors and in persons with a history of salmonellosis]. Zh Mikrobiol Epidemiol Immunobiol 1987 Sep;34(9):108-12

"The sera obtained from healthy donors and from subjects with a history of *Salmonella* infections (B, C1, D1 and E1) of medium severity were studied. In the initial testing the heterologous enzyme immunoassay the levels of antibodies to LPS groups B and D1, were somewhat higher in the subjects with a history of the infections than in healthy

donors. After a single adsorption with homologous LPS the levelling of differences between the two groups was observed. After two adsorptions with heterologous LPS the level of the reaction in both the groups increased, and the differences between these groups augmented. The adsorption of sera with heterologous LPS increased the diagnostic sensitivity of the reaction only to 14-23% in the determination of the serogroup of the causative agent. An increase in the antibody level after adsorption with heterologous LPS was supposedly due to the additional detection of group-specific antibodies with low avidity, which appeared as the result of the elimination of cross-reacting highly avid antibodies, as well as non-immunoglobulin molecules complementary to LPS." (MEDLINE)

385 Prey MU, Lorelle CA, Taff TA, Sonsoucle L, Webb MS, Gardner TD, Aquino TI. Evaluation of three commercially available rotavirus detection methods for neonatal specimens. *Am J Clin Pathol* 1988 May;89(5):675-8

386 Prusak-Sochaczewski E, Luong JH. Utilization of two improved enzyme immunoassays based on avidin-biotin interaction for the detection of *Salmonella*. *Int J Food Microbiol* 1989 Jul;8(4):321-33

"Based on a strong interaction between avidin and biotin, two enzyme immunoassays have been modified and tested for the detection of *Salmonella typhimurium* in foodstuffs. In both assays, the antigen containing sample was first reacted with antibody to *Salmonella* which was precoated on a polystyrene microtiter plate. The bound antigen was then allowed to react with an appropriate amount of biotinylated antibody. In the first procedure, the presence of *Salmonella* was quantified by using peroxidase-labeled avidin. In the latter, avidin acted as a bridge between the biotinylated antibody and the biotinylated peroxidase. Samples containing 10(3) and 10(4) cells/ml of the *Salmonella* virulent strain, respectively, were detectable by these two methods. The results thus compared favorably with the detection limit of the standard ELISA (10(5) cells/ml). The superiority of the modified ELISA's utilizing biotin/avidin interactions was also demonstrated for the detection of *Salmonella* in artificially contaminated food samples inoculated with only 2-5 *Salmonella* cells followed by two incubation steps. No significant interference of *E. coli* (up to $5 \times 10(6)$ cells/ml) was observed in the quantification of *Salmonella* cells." (MEDLINE)

387 Qadri A, Ghosh S, Prakash K, Kumar R, Moudgil KD, Talwar GP. Sandwich enzyme immunoassays for detection of *Salmonella typhi*. *J Immunoassay* 1990; 11(2):251-69

"Enzyme immunoassays were developed using monoclonal antibodies raised against somatic (O), flagellar (H) and capsular (VI) antigens of *Salmonella typhi*. The assay based on anti-O monoclonal antibodies could specifically detect *S. typhi* and soluble lipopolysaccharide (LPS) isolated from *S. typhi*. Anti-H MoAbs detected motile *S. typhi* and soluble flagellar antigen. Monoclonal antibodies against capsular polysaccharide could detect VI-containing *S. typhi* as well as soluble VI antigen. The three assays reported here detected *S. typhi* with 100% sensitivity in blood culture broths obtained from bacteriologically confirmed typhoid patients and were negative with blood specimens containing *Salmonella* senftenberg, *E. coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Proteus mirabilis* or *Streptococcus* (alpha-hemolytic) derived from patients with pyrexia. The assays, however, did not demonstrate the presence of soluble antigens in sera and urine samples obtained from typhoid patients." (MEDLINE)

388 Qadri SMH, Akhter J, Ostrowski S, Qadri SGM, Cunha BA. High incidence of false positives by a latex agglutination test for the diagnosis of *Clostridium difficile* associated colitis in compromised patients. *Diagn Microbiol Infect Dis* 1989 Jul/Aug;12(4):291-4

"Detection of *Clostridium difficile* cytotoxin using cell culture assays for the diagnosis of

antibiotic-associated colitis has been used for over a decade. Because the methodology is time consuming and cumbersome, a recently introduced commercial latex agglutination (LA) kit has attracted much attention. We compared the sensitivity and specificity of this method with the cytotoxic assay (CTA) using diarrheal stools from 652 patients at a referral tertiary care center. Specimens from 71 (10.9%) patients were found positive with CTA and 98 (15%) by LA. Of these, 67 stools were positive by both methods. Four specimens showed cytotoxicity but were negative by LA. Of the 31 patient specimens that were positive by LA but negative by CTA, 23 were obtained from leukemic, bone marrow transplant and four from renal transplant patients. Thirteen of these patients had *Giardia lamblia* (three), *Salmonella enteritidis* (three), *Blastocystis hominis* (four), *Rotavirus* (two), and *Shigella boydii* (one) in their stools. No significant organisms were found in the rest of the LA-positive and CTA-negative specimens." (Authors' abstract)

389 Raghubeer EV, Matches JR. Temperature range for growth of *Escherichia coli* serotype O157:H7 and selected coliforms in *E. coli* medium. J Clin Microbiol 1990 Apr;28(4):803-5

390 Rahaman MM, Morshed MG, Aziz KMS, Munshi MMH. Improved medium for isolating *Shigella* [letter]. Lancet 1986 Feb 1;1(8475):271-2

Shigellae require special handling for isolation, and multiple platings on selective media are recommended. Here a modified MacConkey's medium is described which greatly increases the rate of isolation of *Shigella dysenteriae* type 1 and *S. flexneri*. Rectal swabs or fecal specimens from 392 diarrhoeal patients at a field station in Teknaf, Bangladesh, were freshly inoculated on MacConkey's agar, *Salmonella-Shigella* (SS) agar and Teknaf enteric agar (TEA) medium. The final identification of *Shigellae* was made by slide agglutination with known antisera. TEA outperformed MacConkey's medium, SS medium, and in combination the MacConkey's and SS media. The isolation rate of *Shigella* spp. with only one TEA plate per patient gave 42% additional isolates. In contrast to MacConkey plates, TEA plates grew fewer lactose fermenters; some TEA plates showed uniform growth of non-lactose fermenters which turned out to be pure colonies of *Shigella*. The diagnosis was confirmed by serology. The TEA medium increased significantly the isolation rate of both *S. dysenteriae* type 1 and *S. flexneri*.

391 Rahman M, Sack DA, Wadood A, Yasmin M, Latif A. Rapid identification of *Vibrio cholerae* serotype O1 from primary isolation plates by a coagglutination test. J Med Microbiol 1989 Jan;28(1):39-41

"A coagglutination test was developed for identifying suspected colonies of *Vibrio cholerae* serotype O1 directly from primary isolation plates. Visible agglutination occurs when *V. cholerae* O1 antibody attached to cell-wall protein A of *Staphylococcus aureus* reacts with its homologous antigen. From 314 faecal samples from clinically suspected cases of cholera, 210 colonies from thiosulphate citrate bile salts sucrose (TCBS) agar and 222 colonies from taurocholate tellurite gelatin (TTG) agar were tested as suspect *V. cholerae*. In each case 204 isolates were identified as *V. cholerae* O1 by conventional methods and also gave positive results for *V. cholerae* O1 in the coagglutination test; with one partial exception, no other colonies tested gave positive results. The coagglutination test is simple and inexpensive and provides a result 24 h earlier than conventional methods." (Authors' abstract)

392 Rai GP, Zachariah K, Shrivastava S. Comparative efficacy of indirect haemagglutination test, indirect fluorescent antibody test and enzyme linked immunosorbent assay in serodiagnosis of typhoid fever. J Trop Med Hyg 1989 Dec;92(6):431-4

"In the present study three tests, viz. IHA, IFA and ELISA were compared for their sensitivity, specificity and predictive values in serodiagnosis of typhoid fever. One hundred

and eleven sera samples comprising 41 culturally confirmed, 14 clinically suspected and 56 normal controls were tested. Among the three tests, ELISA was found to be the method of choice using single serum sample." (Authors' abstract)

393 Rai GP, Zachariah K, Shrivastava S. Detection of typhoid fever by Widal and indirect fluorescent antibody (IFA) tests: a comparative study. *J Hyg Epidemiol Microbiol Immunol* 1989;33(3):331-6

"Widal test is a conventional method for the detection of typhoid fever. However, it takes 18-24 hours to complete the test. In the present study indirect fluorescent antibody test has been compared with the Widal test using single serum specimens and was found to be rapid, sensitive and specific. Serum specimens from 41 culture proven cases of typhoid fever, 14 clinically suspected cases and 22 normal individuals were collected. Whereas Widal test detected 63.41% positive cases. IFA test detected 87.80% from among culturally proven typhoid cases. Among the clinically suspected cases of typhoid fever, IFA test detected 85.71% (28.57 + 57.14%) while Widal test detected only 57.13% (35.71 + 21.42%) positive cases out of above 14 cases." (Authors' abstract)

394 Raj P, Bhan MK, Prasad AK, Kumar R, Sazawal S, Bhandari N. Evaluation of a new latex agglutination kit for detection of human rotavirus in faecal specimens. *Indian J Med Res* 1989 May;89:165-9

"A commercial latex agglutination (LA) test was compared with ELISA and direct electron microscopy (EM) for detection of rotavirus antigen in 93 stool specimens obtained from as many children with acute gastroenteritis. Seventy one specimens (76.3%) were either positive or negative with all the three techniques, while 22 (23.7%) gave contradictory results. Only 1 sample was positive by LA test but not with ELISA or EM. The sensitivity of LA test and EM was 62.5 per cent (30 of 48) and 75 per cent (36 of 48); the corresponding specificity being 97.7 per cent (44 of 45) and 100 per cent (45 of 45) respectively. ELISA was more sensitive than the LA test and EM for detection of rotavirus antigen. LA test which is highly specific and a rapid method, may be useful in certain situations but its low sensitivity makes its unsuitable for use in routine clinical practice." (Authors' abstract)

395 Rakotozandrindraini R, Hamann HP, Zschock M, Schliesser T. [Thermophilic *Campylobacter* from fecal samples of animals and their behavior in the ganglioside (GM1)-ELISA, Vero cell test and salt aggregation test]. *Zentralbl Veterinarmed [B]* 1990 Mar;37(2):142-7

"From 397 fecal specimens from apparently healthy and from diarrheic pigs, dogs, cats and cattle 59 strains (= 15%) of thermophilic *Campylobacter* (C.) spp. were isolated by culture. 39 strains were identified as *C. coli* and 18 as *C. jejuni* whereas 2 isolates could not be classified. None of the strains was found to be positive for cytotoxic enterotoxin in the GM1-ELISA. In the Vero-cell test 5 isolates showed a cytotoxic effect. The salt aggregation test (SAT) for indicating cell surface hydrophobicity was positive with 24 strains (5 *C. jejuni*, 19 *C. coli*). A correlation of isolation results with clinical manifestation could not be observed." (MEDLINE)

396 Ram S, Khurana S, Sharma S, Vadhwa DV. Evaluation of coagglutination test for serotyping of enteropathogenic bacteria. *Indian J Med Res* 1989 Sep;89: 290-6

"Results of conventional agglutination and coagglutination (COA) tests for serotyping of enteropathogenic *Escherichia coli* (EPEC), *Shigella*, *Salmonella* and *Vibrio cholerae* were compared. Eighty isolates of different serotypes of EPEC falling in the wellcome polyvalent (OK) 2, 3 and 4 antisera, showed good (4+) agglutination with COA reagent up to 1:960 dilution. Out of 50 strains of *Shigella* species tested, 25 of *Sh. dysenteriae* and 5 of *Sh. sonnei* gave good reaction up to 1:1920 dilution and 15 of *Sh. flexneri* up to 1:960 dilution in COA test. Similar reaction (4+) by 5 strains of *Sh. boydii* was seen

up to 1:480 dilution only. All 100 isolates of different *Salmonella* species viz., *S. paratyphi* A(5), *S. typhimurium* (50), *S. typhi* (15), *S. weltevreden* (5) and *S. senftenberg* (25), when serotyped by COA, gave good reaction in 1:480 dilution except *S. typhi* factor 9 and dH antisera, which gave very weak reaction at 1:480 dilution and good reaction at 1:280 dilution. All seven isolates of *V. cholerae* gave good reaction in COA test even up to 1:1920 dilution. No cross reaction with any COA reagent was obtained in the 1206 heterologous isolates tested. Thus due to its higher efficiency, lower cost and good specificity, COA may prove to be a better method for serotyping of enteropathogens." (Authors' abstract)

397 Ramamurthy T, Pal A, Nair GB, Pal SC, Takeda T, Takeda Y. Experience with toxin bead ELISA in cholera outbreak (letter). *Lancet* 1990 Aug 11;336(8711):375-6

398 Rambach A. New plate medium for facilitated differentiation of *Salmonella* spp. from *Proteus* spp. and other enteric bacteria. *Appl Environ Microbiol* 1990 Jan;56(1):301-3

399 Rand KH, Houck HJ, Swingle HM. Rotazyme assay in neonates without diarrhea: results of screening survey and preliminary analysis of false positive specimens. *Am J Clin Pathol* 1985 Dec;84(6):748-51

"One hundred forty-seven stool specimens from 93 infants younger than four months of age in a Neonatal Intensive Care Unit were tested for rotavirus by the Rotazyme ELISA method (Abbott Laboratories, North Chicago, IL). None of the infants had diarrhea at the time of the testing. Ten of 147 (6.8%) specimens were either low or suspect positive. None had rotavirus by electron microscopy. Excluding the suspect positives, which were negative on retesting, the false positive rate was only 6 of 147 (4.1%). Of five specimens with sufficient material and repeatedly positive tests, heat to 56°C for one-half hour eliminated the binding to the Rotazyme bead but had no effect on the rotavirus positive control. One patient was found to have an extremely high positive Rotazyme test, independently of the survey. No virus was found in this specimen by electron microscopy, and the material responsible for the false positive result was not removed by centrifugation (100,000 X g for one hour), heat to 56°C for one-half hour, trypsin, ether/ β -mercaptoethanol, or dialysis. Thus, false positives were encountered, but the overall rate was acceptably low. Such false positives are likely to result from more than one cause and, depending on results of further study, may be confirmed as false positives by loss of reactivity at 56°C for one-half hour and perhaps lack of binding to a control bead." (Authors' abstract)

400 Rao BN. An evaluation of modified Widal test in the diagnosis of enteric fever. *J Indian Med Assoc* 1989 Aug;87(8):179-80

"Serum samples obtained from 50 bacteriologically positive cases of *Salmonella typhi* infection as well as 50 healthy individuals were subjected to conventional and modified Widal tests simultaneously. A 4-fold difference in the titres was noticed in 50 sera of the test group and no change in the titres of the control group. The early rising 'O' antibodies which are of IgM in nature and formed due to recent infection are inactivated by 2-mercaptoethanol while it has no effect on IgG antibodies which are formed due to flagellar 'H' antigens or due to anamnestic response or by non-specific reasons. The use of modified Widal test along with conventional one can therefore help in the diagnosis of enteric fever with certainty." (Authors' abstract)

401 Rattan A, Maheshwari V, Maheshwari NK, Malik A, Malik MA. Application of modified widal test in specific diagnosis of enteric fever. *Indian Pediatr* 1990 Mar;27(3):295-7

402 Ravaoarino M, Coulanges P. [Comparison of the latex technic and the ELISA technic in research on human rotaviruses in the feces]. *Arch Inst Pasteur Madagascar* 1987;53(1):119-22

403 Raybould TJG, Crouch CF, Acres SD. Monoclonal antibody passive hemagglutination and capture enzyme-linked immunosorbent assays for direct detection and quantitation of F41 and K99 fimbrial antigens in enterotoxigenic *Escherichia coli*. J Clin Microbiol 1987 Feb;25(2):278-84

"Production of diarrhea in neonatal calves by enterotoxigenic *Escherichia coli* depends on its ability to attach to the epithelial cells of the intestine via surface adhesins called pili or fimbriae and to secrete enterotoxins. The most important of these fimbriae are designated K99 and F41. We produced and characterized a murine monoclonal antibody specific to F41. This monoclonal antibody and a K99-specific monoclonal antibody were used to develop sensitive and specific passive hemagglutination and capture enzyme-linked immunosorbent assays (ELISAs) for detection and quantitation of F41 and K99 antigens in *E. coli* cultures and culture supernatants. The capture ELISA systems exhibited excellent sensitivity and specificity, whereas the passive hemagglutination systems appeared to be oversensitive. The ability of the capture ELISAs to detect K99 and F41 fimbrial antigens in fecal specimens from calves was evaluated. Fimbrial antigens were detected in six of six specimens from scouring calves but not in four of four specimens from nonscouring calves." (Authors' abstract)

404 Reynolds D, Hughes JH. Comparison of the Rotazyme assay with an avidin-biotin-amplified dot-immunobinding assay for detecting rotaviruses [letter]. J Infect Dis 1985 Sep;152(3):647-8

405 Roberts AP, Talboys CA, O'Neill PM, Azadian BS. Assessment of rapid method for identifying *Escherichia coli* (letter). J Clin Pathol 1989 Apr;42(4): 441

406 Roberts DH, Lucas MH, Swallow C. Comparison of the agar gel immunodiffusion test and ELISA in the detection of bovine leukosis virus antibody in cattle persistently infected with bovine virus diarrhoea virus. Vet Immunol Immunopathol 1989 Oct;22(3):275-81

"When six cattle persistently infected with bovine virus diarrhoea virus (BVDV) were inoculated with lymphocytes infected with bovine leukosis virus (BLV), a depressed antibody response to BLV was observed by ELISA which was due to a decrease in IgG1 synthesis. The ELISA was more sensitive and more reliable than the agar gel immunodiffusion (AGID) test in detecting BLV infection in cattle persistently infected with BVDV. Decreased antibody responses were manifested in the AGID test by negative, inconclusive or weakly positive reactions: only two of the six cattle developed antibodies that generated positive AGID reactions." (MEDLINE)

407 Robins-Browne R, Cianciosi S, Morris JG, Jr. Evaluation of different techniques for detection of virulence of *Yersinia enterocolitica* [letter]. J Clin Microbiol 1990 Sep;28(9):2159

408 Robins-Browne RM, Millotis MD, Cianciosi S, Miller VL, Falkow S, Morris JG, Jr. Evaluation of DNA colony hybridization and other techniques for detection of virulence in *Yersinia* species. J Clin Microbiol 1989 Apr;27(4):644-50

409 Rogers DG. Application of the peroxidase-antiperoxidase technique for the detection of enterotoxigenic *Escherichia coli* K99 pilus antigen in paraffin-embedded tissues. J Histochem Cytochem 1985 Aug;33(8):762-6

"The peroxidase-antiperoxidase (PAP) technique was evaluated as a method for detecting enterotoxigenic *Escherichia coli* (ETEC) K99 pilus antigen in paraffin-embedded tissues of swine. Antigenic reactivity was optimal when tissues were processed in cold methanol according to a modified method of Sainte-Marie. This method of processing adequately preserved tissue morphology. The PAP technique was applicable to tissues where endogenous peroxidase activity was minimal." (MEDLINE)

410 Rojas L, Torres DR, Mediola BJ, Finlay CM. Detection of specific anti-*Giardia* serum antibody by an immunofluorescence test in children with clinical giardiasis. *Am J Trop Med Hyg* 1989 May;40(5):477-9

411 Romla SA, Abou-Zakham AA, el-Kholy ES. Immunodiffusion, immunofluorescence and enzyme-linked immunosorbent assay in the serodiagnosis of giardiasis. *J Egypt Soc Parasitol* 1990 Jun;20(1):209-14

"Circulating anti-*Giardia lamblia* antibodies have been estimated in the sera of 48 patients of parasitologically proven *G. lamblia* infection using the immunodiffusion (I.D.) indirect fluorescent antibody (I.F.A.) and enzyme-linked immunosorbent assay (ELISA) tests. The results showed that ELISA technique was the most simple, sensitive (95.8%) and specific (100%) test. These advantages make it more applicable in seroepidemiology of giardiasis." (MEDLINE)

412 Ronnberg B, Soderlund O, Wadstrom T. Evaluation of a competitive enzyme-linked immunosorbent assay for porcine *Escherichia coli* heat-stable enterotoxin. *J Clin Microbiol* 1985 Dec;22(6):893-6

"A competitive enzyme-linked immunosorbent assay (ELISA) was compared with the conventional suckling mouse assay for the identification of heat-stable enterotoxin (STa) in samples from piglets suffering from diarrhea. A total of 110 *Escherichia coli* isolates, 22 primary cultures, and 26 fecal samples from piglets up to 8 weeks of age with diarrhea were compared in parallel by both assays. Of the 110 isolates tested, all gave consistent results by the ELISA and the suckling mouse assay; 60 strains were negative and 50 strains produced STa by both tests. Identical results were obtained when 22 primary agar cultures were screened for STa production by both methods; 6 were found to produce STa, while 16 did not. When 26 fecal samples were tested for the presence of STa, 10 were negative and 12 were positive by both assays. One of the remaining four samples gave questionable positive results by both the suckling mouse assay and the ELISA, but *E. coli* isolated from this sample gave positive results by both tests. The remaining three samples were negative by the suckling mouse assay, but gave questionable positive results by the ELISA. *E. coli* isolates from these three samples were always negative by both assays. The ELISA used in this study provides a reliable and convenient method for diagnosing STa-producing enterotoxigenic *E. coli* of porcine origin." (Authors' abstract)

413 Rose JB, Landeen LK, Riley KR, Gerba CP. Evaluation of immunofluorescence techniques for detection of *Cryptosporidium* oocysts and *Giardia* cysts from environmental samples. *Appl Environ Microbiol* 1989 Dec;55(12):3189-96

"*Cryptosporidium* and *Giardia* species are enteric protozoa which cause waterborne disease. The detection of these organisms in water relies on the detection of the oocyst and cyst forms or stages. Monoclonal and polyclonal antibodies were compared for their abilities to react with *Giardia* cysts and *Cryptosporidium* oocysts after storage in water, 3.7% formaldehyde, and 2.5% potassium dichromate, upon exposure to bleach, and in environmental samples. Three monoclonal antibodies to *Cryptosporidium parvum* were evaluated. Each test resulted in an equivalent detection of the oocysts after storage, after exposure to bleach, and in environmental samples. Oocyst levels declined slightly after 20 to 22 weeks of storage in water, and oocyst fluorescence and morphology were dull and atypical. Oocyst counts decreased after exposure to 2,500 mg of sodium hypochlorite per liter, and fluorescence and phase-contrast counts were similar. Sediment due to algae and clays found in environmental samples interfered with the detection of oocysts on membrane filters. Two monoclonal antibodies and a polyclonal antibody directed against *Giardia lamblia* cysts were evaluated. From the same seeded preparations, significantly greater counts were obtained with the polyclonal antibody. Of the two monoclonal antibodies, one resulted in significantly lower cyst counts. In preliminary studies, the differences between antibodies were not apparent when used on

the environmental wastewater samples. After 20 to 22 weeks in water, cyst levels declined significantly by 67%. Cysts were not detected with monoclonal antibodies after exposure to approximately 5,000 mg of sodium hypochlorite per liter." (Authors' abstract)

414 Rosoff JD, Sanders CA, Sonnad SS, deLay PR, Hadley WK, Vincenzi FF, Yajko DM, O'Hanley PD. Stool diagnosis of giardiasis using a commercially available enzyme immunoassay to detect *Giardia*-specific antigen 65 (GSA 65). J Clin Microbiol 1989 Sep;27(9):1997-2002

"A commercially available enzyme immunoassay for the diagnosis of giardiasis was evaluated in a clinical trial. The ProSpecT/*Giardia* diagnostic test (Alexon, Inc., Mountain View, Calif.) was compared with the standard ova and parasite (O&P) microscopic examination. Additionally, several widely used stool fixatives and a commonly used transport medium were assessed for compatibility with the immunoassay. A total of 325 stool specimens were collected and used to evaluate assay performance. Of those, 93 specimens were collected from symptomatic *Giardia* O&P-positive patients and 232 specimens were randomly collected from patients as part of a routine health screening procedure. All 93 *Giardia* O&P-positive stool specimens were strongly positive by visual and spectrophotometric examination using the immunoassay. Of the 232 randomly collected specimens, 16 were positive by O&P examination and immunoassay, 6 were negative by O&P examination but positive by immunoassay, and 1 was positive by O&P examination and negative by immunoassay. There was substantial supportive evidence that indicated that the six immunoassay-positive, O&P-negative specimens were true-positives. When these six specimens were accepted as true-positives, the immunoassay detected almost 30% more cases of *Giardia* infection than did O&P examination. Its sensitivity and specificity were 96 and 100%, respectively, while the sensitivity and specificity of O&P examination were 74 and 100%, respectively. The immunoassay also performed well on specimens treated with 10% neutral formalin, sodium acetate-formalin fixative, and Cary-Blair transport medium. However, the test was not compatible with polyvinyl alcohol-treated specimens. Overall, the ProSpecT/*Giardia* test was a sensitive, specific immunoassay which was easy to run and interpret. It offers a simple solution to traditional difficulties encountered in diagnosing *Giardia* infection." (Authors' abstract)

415 Rotbart HA, Yolken RH, Nelson WL, Davis D, Roe MH, Levin MJ. Confirmatory testing of Rotazyme results in neonates. J Pediatr 1985 Aug;107(2):289-92

416 Roxas DJC, Mendoza M. Assessment of a single Widal test in the diagnosis of enteric fever. J Philip Med Assoc 1989 Oct-Dec;65(2):211-4

"The diagnostic value of a single Widal Test was assessed in our institution. The control population consisted of 40 normal healthy individuals who were tested for Widal titers in October 1986. The study population, on the other hand, consisted of 40 culture-proven cases of enteric fever seen between January 1981 and September 1986. Only the highest H or O titer was considered for each subject. Of the 40 healthy subjects, 17 or 42% had 1:160 titer in either H or O while 585 had 1:80 titers. The sensitivity and specificity of the test was calculated, assuming that the control group had negative culture studies at the time of examination. Results showed that at a titer of 1:320, the sensitivity is 57.5%, while the specificity is 100%. If we consider 1:160 as "diagnostic", which is usual in some centers, the sensitivity would be 72.5%, but the specificity only 57.5%. there was no statistically significant difference between the sensitivity of the test during the first and second week of illness. On the basis of these findings, titers of 1:320 were found of diagnostic significance in this series. The significance and implications of these findings are discussed." (Authors' abstract)

417 Rubin FA, McWhirter PD, Burr D, Punjabi NH, Lane E, Kumala S, Sudarmono P, Pulungsih SP, Lesmana M, Tjaniadi P, Sukri N, Hoffman SL. Rapid diagnosis of typhoid fever through identification of *Salmonella typhi* within 18 hours of specimen acquisition by culture of the mononuclear cell-platelet fraction of blood. J Clin Microbiol 1990 Apr;28(4):825-7

418 Rulzong W. Isolating *Campylobacter jejuni* in an atmosphere made oxygen deficient using pyrogallol in a candle jar. J Diarrhoeal Dis Res 1987 Jun;5(2):107-9

419 Ryan RW. Considerations in the laboratory diagnosis of antibiotic-associated gastroenteritis. Diagn Microbiol Infect Dis 1986 Mar;4(3 suppl):S79-S86

"*Clostridium difficile* has been shown to be the major cause of antibiotic-associated gastroenteritis in both humans and experimental animals. During the past few years an increasing number of laboratories have attempted to detect, isolate, and identify this organism and its toxin from clinical samples. Direct visualization of *C. difficile* in patient specimens using immunofluorescent microscopy has been proposed. The major disadvantage of this method was its lack of specificity due to crossreaction with other clostridial species. Attempts to absorb the antisera with crossreacting strains also failed. Laboratory diagnosis of *C. difficile* in clinical specimens has relied on either culture using one or more selective media or on the detection of specific cytotoxin in stool filtrates. Until recently the cytotoxicity assay was the only procedure available for the routine detection of cytotoxin and, as a result, has limited this test to laboratories with access to tissue culture facilities. As a result, there has been much interest in the development of immunochemical methods for the detection of *C. difficile* toxins. We originally reported on the detection of *C. difficile* toxin in stool filtrates using counterimmunoelectrophoresis. We examined 140 fecal specimens submitted for *C. difficile* toxin assay by counterimmunoelectrophoresis, using both unabsorbed and absorbed antitoxin, tissue culture, and bacterial culture. Using tissue culture assay as the reference method, the sensitivity of counterimmunoelectrophoresis and counterimmunoelectrophoresis-absorbed was 100% and the specificity 63.0% and 77.5%, respectively. Enzyme immunoassay assays for the detection of toxin A from *C. difficile* have also been reported, however, at the present time they do not appear to be as sensitive as the cytotoxicity assay for toxin B (cytotoxin)...." (MEDLINE)

420 Sadallah F, Brighthouse G, Giudice GD, Drager-Dayal R, Hocine M, Lambert PH. Production of specific monoclonal antibodies to *Salmonella typhi* flagellin and possible application to immunodiagnosis of typhoid fever. J Infect Dis 1990 Jan;161(1):59-64

"Four murine monoclonal antibodies (MAbs) to *Salmonella typhi* flagellin were produced. These MAbs did not react with eight other enterobacterial strains tested: *Salmonella enteritidis*, *Salmonella typhimurium*, *Salmonella paratyphi A*, *Escherichia coli*, *Shigella flexneri*, *Shigella sonnei*, *Yersinia enterocolitica*, and *Campylobacter jejuni*. All four MAbs cross-reacted with *Salmonella muenchen* flagellin indicating specificity for d antigenic flagellar epitope. One MAb (C4) was selected to develop a double antibody sandwich enzyme-linked immunosorbent assay (ELISA) to detect *S. typhi* flagellin in serum samples. By use of this assay *S. typhi* flagellar antigen was detected in 95.5% of serum samples from patients with positive hemoculture for *S. typhi*, in 93.6% of samples from patients with positive serodiagnosis of typhoid fever, in 26% of samples collected from patients who were initially hemoculture-positive for *S. typhi* and who had undergone 7-8 d of chemotherapy, in 8.5% of samples from healthy persons from an endemic area, and in no samples from healthy persons from a nonendemic area. The presence of high levels of flagellin antibody titers did not interfere with the antigen detection. The detection of *S. typhi* flagellar antigen in patient serum may have practical value for rapid diagnosis of typhoid fever." (Authors' abstract)

421 Saez-Llorens X, Guzman-Verduzco LM, Shelton S, Nelson JD, Kupersztoch YM. Simultaneous detection of *Escherichia coli* heat-stable and heat-labile enterotoxin genes with a single RNA probe. J Clin Microbiol 1989 Jul;27(7):1684-8

"A single RNA probe was synthesized and used to detect simultaneously the methanol-soluble heat-stable enterotoxin and heat-labile enterotoxin genes in *Escherichia coli* strains. The results with the blot hybridized or radioactive probe correlated 100% with the biological assay results for both toxins. The RNA probe detected the three known heat-stable enterotoxin A alleles." (Authors' abstract)

422 Saha SK, Sanyal SC. Better preservation of *Campylobacter jejuni*/C. coli in a defined medium. Indian J Med Res 1991 Jan;93:26-8

"*Campylobacter jejuni* and C. coli strains were preserved at -10°C in different stock media to determine their efficacy to preserve the organism for a longer period of time. An improved defined stock culture medium was developed for the organism by removal and effective neutralisation of the toxic metabolites. Comparative study revealed that phosphate buffer saline (PBS), pH 6.7 supplemented with 0.2 percent charcoal, 0.025 percent FBP (ferrous sulphate, sodium metabisulphate and sodium pyruvate), 0.1 percent L-cystein and 10 percent glycerol could support survival of C. jejuni/coli strains for as long as 135 days at -10°C followed by George's medium, brucella broth with 15 percent glycerol, fetal calf serum with 50 percent TSYB (trypticase soy yeast-broth) and glycerol transport broth respectively." (Authors' abstract)

423 Salama SM, Bolton FJ, Hutchinson DN. Application of a new phage typing scheme to campylobacters isolated during outbreaks. Epidemiol Infect 1990 Jun;104(3):405-11

424 Samadpour M, Liston J, Ongerth JE, Tarr PI. Evaluation of DNA probes for detection of Shiga-like-toxin-producing *Escherichia coli* in food and calf fecal samples. Appl Environ Microbiol 1990 May;56(5):1212-5

"The use of DNA probes for Shiga-like toxin I (SLT-I) and SLT-II for detection of SLT-producing *Escherichia coli* (SLTEC) in foods and calf fecal samples was evaluated. Enrichment cultures were prepared from food or fecal samples. Colonies formed by plating of enrichment cultures were probed for SLTEC by colony hybridization. Alternatively, enrichment cultures were analyzed for SLTEC presence by dot blot. The lowest detected concentration of SLTEC in sample homogenates inoculated with E. coli O157:H7 corresponded to 1.3 CFU/g of sample. Of the 44 food samples and 28 fecal samples from dairy calves tested by the colony hybridization method, 4 food samples, including ground beef, raw goat milk, blueberries, and surimi-based delicatessen salad, and 9 calf fecal samples were positive with the SLT probes. The dot blot technique yielded results within 48 h and can be used as a fast and sensitive method of detection for SLTEC in foods and calf fecal samples. The colony hybridization technique took 3 to 4 days but permits recovery of the positive colonies when desired." (Authors' abstract)

425 Sambourg M, Goudeau A, Courant C, Pinon G, Denis F. Direct appraisal of latex agglutination testing, a convenient alternative to enzyme immunoassay for the detection of rotavirus in childhood gastroenteritis, by comparison of two enzyme immunoassays and two latex tests. J Clin Microbiol 1985 Apr;21(4):622-5

"During February and March 1984, 207 fecal samples from infants and children with gastroenteritis were tested for rotavirus with four techniques: two enzyme immunoassays (Rotazyme; Abbott Laboratories, North Chicago, Ill., and Enzygnost-Rotavirus; Calbiochem-Behring, La Jolla, Calif.) and two latex agglutination tests (Rotalax; Orion Research, Inc., Cambridge, Mass., and Slidex Rota-Kit; Biomerieux). All stool samples were also tested for yeasts and bacterial pathogens. Electron microscopy was used to investigate discrepant results. We found 47% positive samples with Enzygnost-Rotavirus, 38% with Rotazyme, 37% with Slidex Rota-Kit, and 34% with Rotalax. No specimen was found positive by Rotazyme only or Slidex Rota-Kit only. On the contrary, 12 samples which were positive with Enzygnost-Rotavirus only and 3 which were positive with Rotalax only were not confirmed as positive by electron microscopy. Both enzyme immunoassays gave 6% equivocal results; Slidex Rota-Kit gave significantly fewer equivocal results than did Rotalax: 2.9% versus 9.7% ($p < 0.01$). The sensitivity and specificity of latex tests compared favorably with that of enzyme immunoassays. Latex agglutination tests can be performed by unskilled personnel and are rapid and relatively cheap. They appear to be very suitable for routine laboratory work and may prove useful for large-scale screening in developing countries." (Authors' abstract)

426 Sami Z, Rehman G. Detection and enumeration of fecal coliforms and other micro-organisms in drinking water (a comparison of two techniques). J Pak Med Assoc 1985 Nov;35(11):329-34

Comparative efficacies of two techniques of water analysis based on bacterial counts were investigated in Pakistan. Fifty water samples from different sources were examined by a standard method and 62 water samples were examined by the method of Rand *et al.* with some alterations (improvised method). Results of presumptive coliform counts by the standard method could be obtained per 100 ml. By the improvised method, the exact coliform counter per ml of water sample could be obtained. For confirmatory fecal coliform test, the time consumed by the standard method was at least 4 days, whereas by the improvised method, this result was available after 2 days. Further, the improvised method required less glassware and culture media. Nine different species were identified by the standard method and eleven isolated by the improvised technique. Organism recovery was 13.5% more by the improvised method than the standard method. Thus, the improvised method has proved to be ideal as it provides information on presumptive coliforms, fecal coliforms, and standard plate counts, and confirms the identity of the isolates. This method was less time consuming and comparatively less expensive.

427 Samrejrangroj P, Tharavanij S. Assessment of validity of counterimmuno-electrophoresis and ELISA in the routine diagnosis of amoebiasis. Southeast Asian J Trop Med Public Health 1985 Sep;16(3):365-70

"Counterimmunoelectrophoresis (CIE) was used to detect antibodies against *Entamoeba histolytica* in 99 sera from amoebiasis suspected patients using antigens from the HK-9 and HT-12 strains of axenically cultivated *E. histolytica*. There was no significant difference of seropositive rates using antigens from these two strains suggesting the both strains were equally suitable for use in the routine CIE test for the diagnosis of amoebiasis. In comparison with ELISA, the CIE test was a little less sensitive than ELISA with per cent sensitivity of 93.5 and 100 respectively. Two amoebic liver abscess patients who were CIE negative were ELISA positive. The CIE seropositive rate was related to the period after onset of clinical symptoms with per cent positivity of 66.7, 93.3 and 100 when the sera were tested on ≤ 5 , 6-10 and ≥ 11 days of illnesses respectively." (Authors' abstract)

428 Samuelson J, Acuna-Soto R, Reed S, Biagi F, Wirth D. DNA hybridization probe for clinical diagnosis of *Entamoeba histolytica*. J Clin Microbiol 1989 Apr;27(4):671-6

429 Sanders RC, Campbell AD, Jenkins MF. Routine detection of human rotavirus by latex agglutination: comparison with enzyme-linked immunosorbent assay, electron microscopy and polyacrylamide gel electrophoresis. J Virol Methods 1986 Jul;13(4):285-90

"A commercially available latex agglutination test, RotaScreen (Merck Diagnostics Ltd., West Byfleet, Surrey, U.K.) was evaluated for the detection of human rotaviruses in stool specimens. The results obtained were compared with those from 3 other routine assay systems used in this laboratory: enzyme-linked immunosorbent assay (ELISA), electron microscopy (EM) and polyacrylamide gel electrophoresis (PAGE) of viral ribonucleic acid. 400 stool samples were examined by the 4 assay systems under routine conditions. RotaScreen latex agglutination was found to be more sensitive than EM and PAGE, and highly specific for rotavirus antigens." (MEDLINE)

430 Santogade PJ, Bokkenheuser VD, Faisal MA, Kotler DP, Scholes JV, Holt PR. Evaluation of methods for identification of *Campylobacter pylori* infection. NY State J Med 1990 Jan;90(1):4-7

"Fifty unselected patients undergoing routine esophagogastroduodenoscopy were evaluated for infection with *Campylobacter pylori* (CP). Antral specimens were cultured, and

biopsies from the antrum and the body of the stomach were examined histologically. Specimens of antral brushings were analyzed with Gram stain, and urease testing was performed on gastric aspirates, antral brushings, and antral biopsy homogenates. Twenty-seven (54%) patients were CP-positive by silver stain and/or culture of mucosal biopsies. The simplest and fastest diagnostic method was Gram stain of antral brushing, which was 93% sensitive and 100% specific. CP-negative patients were more likely to have normal histology in antrum and body tissues, while CP-positive patients usually exhibited superficial or chronic gastritis ($p < 0.01$). Using ELISA technique, 67% of all patients and 89% of CP-positive patients had serum antibodies against sonicated CP organisms. We conclude that evidence of gastric CP infection is common, is associated with inflammatory changes of the gastric mucosa, is suggested by finding antibodies to CP in serum, and can be accurately and rapidly diagnosed by staining of endoscopically derived cytology and biopsy specimens." (MEDLINE)

431 Sathar MA, Bredenkamp BLF, Gathlram V, Simjee AE, Jackson TFHG. Detection of *Entamoeba histolytica* immunoglobulins G and M to plasma membrane antigen by enzyme-linked immunosorbent assay. J Clin Microbiol 1990 Feb;28(2):332-5

"Sixty-one serum specimens from 22 patients with clinically diagnosed amoebic liver abscess (ALA), 10 hospitalized patients with a variety of diseases other than amoebiasis, 12 normal healthy controls, and 17 subjects from an amoebiasis-endemic area were assayed by enzyme-linked immunosorbent assay (ELISA). The plasma membrane fraction of axenic cultures of *Entamoeba histolytica* HK9 separated from other subcellular fractions by differential centrifugation was used as the antigen to detect specific immunoglobulin G (IgG) and IgM antibodies. Using a single serum dilution of 1/100 and optical densities at 492 nm of 0.200 and 0.250 as the cutoff values for the IgM and IgG ELISAs, their respective sensitivities in 22 ALA patients were 91% (20 of 22) and 95% (21 of 22). In 22 patients (10 hospitalized and 12 normal healthy controls), the specificities of the IgM and IgG ELISAs were 95% (21 of 22) and 91% (20 of 22), respectively. All five asymptomatic carriers of pathogenic *E. histolytica* were seropositive by the IgG ELISA and the amoebic gel diffusion test (AGDT). The AGDT was positive for three of six culture-negative controls, while the IgG ELISA was positive for all six. For six asymptomatic carriers of nonpathogenic zymodemes, the AGDT was positive for two, and the IgG ELISA was positive for three. There was an excellent correlation ($r = 0.96$) between the IgG ELISA and the AGDT. Only one of six culture-negative controls, none of the asymptomatic carriers of pathogenic *E. histolytica*, and one of six carriers of nonpathogenic *E. histolytica* were seropositive by the IgM ELISA, thus highlighting the specificity of the IgM ELISA in the diagnosis of ALA. It is believed that the use of plasma membrane fractions has improved the diagnostic potential of the IgM ELISA." (Authors' abstract)

432 Scholl DR, Kaufmann C, Jollick JD, York CK, Goodrum GR, Charache P. Clinical application of novel sample processing technology for the identification of salmonellae by using DNA probes. J Clin Microbiol 1990 Feb;28(2): 237-41

433 Scott-Taylor T, Ahluwalia G, Klisko B, Hammond GW. Prevalent enteric adenovirus variant not detected by commercial monoclonal antibody enzyme immunoassay. J Clin Microbiol 1990 Dec;28(12):2797-2801

"A commercial monoclonal antibody enzyme immunoassay for the detection of enteric adenovirus types 40 and 41 (Ad40 and Ad41) in stool specimens was evaluated. Twenty-one stool specimens from children with gastroenteritis, with adenovirus particles visible by electron microscopy, and reference strains Ad40 Dugan and Ad41 Tak were tested by Ad40- and Ad41-specific and adenovirus group-reactive immunoassays. All stool specimens tested positive in the group-reactive immunoassay. However, only six specimens, containing isolates of Ad40 strain Hovi-X, and Ad40 genomic variant, and Ad41 strain Tak, reacted with the specific immunoassay, besides the reference strains. Fifteen stool specimens determined by restriction analysis to contain a genomic variant of Ad41 were negative by specific immunoassay. The positions of restriction site differences from the prototype strain Ad41 Tak were analyzed, and four mutations were mapped

within the hexon gene; two others may occur in the fiber gene. The Ad41 genomic variant not detected by the enteric test is presently the most frequent cause of local adenoviral gastroenteritis. Highly specific monoclonal antibodies can fail to detect genomic variants of enteric adenoviruses, probably because of alteration of external neutralizable epitopes under immunological pressure to vary." (Authors' abstract)

434 Sefcovicova L, Tietzova J, Rusnak M, Sprockova N. [Incidence of rotaviruses in the feces of neonates determined by the ROTA-ELISA-Bloveta Immunoenzyme test kit]. *Cesk Epidemiol Mikrobiol Immunol* 1987 Feb;36(1):10-6

435 Selb B, Baumeister HG, Maass G, Doerr HW. Detection of human rotavirus by nucleic acid analysis in comparison to enzyme-linked immunoassay and electron microscopy. *Eur J Clin Microbiol* 1985 Feb;4(1):41-5

"The results of RNA analysis for the detection of rotavirus were compared with those of a standard enzyme-linked immunosorbent assay (ELISA) and electron microscopy using 212 faecal specimens obtained from 200 children with gastroenteritis. Rotavirus was extracted directly from faecal specimens and RNA segments were made visible by polyacrylamide gel electrophoresis using a silver staining technique. Of the 212 faecal specimens 137 were found to be positive in ELISA, 125 in RNA analysis and 121 in both methods. Forty-nine of the 212 specimens were also investigated by electron microscopy. Thirty-five were positive when examined by electron microscopy, 37 were positive in ELISA and 33 in RNA analysis. RNA analysis of 119 faecal samples in outbreaks and sporadic cases of rotavirus infection yielded 42 different rotavirus electrophoretotypes. The results indicated that no one method was sufficient to detect all positive specimens and that RNA analysis is useful in epidemiological studies." (Authors' abstract)

436 Sen D, Saha MR, Manna B, Pal SC. Xylose lysine desoxycholate agar for recovering *Shigella dysenteriae* 1 and *Shigella flexneri* from faeces. *J Diarrhoeal Dis Res* 1987 Jun;5(2):94-6

"To test the relative efficacy of isolating *Shigella* spp. from faeces, xylose lysine desoxycholate agar (XLD) was compared with MacConkey's agar (MA), desoxycholate citrate agar (DCA) and *Salmonella-Shigella* agar (SSA). Inoculation onto XLD medium alone or onto combinations of plating media which include XLD resulted in a significantly higher ($p < 0.001$) recovery of *Shigella dysenteriae* type 1 and *S. flexneri* than when using MA, SSA, or DCA alone or in combination which exclude XLD. The XLD medium was superior to MA, DCA and SSA for recovering *S. dysenteriae* type 1 and *S. flexneri*. The importance of selecting appropriate combinations of media for isolating *S. dysenteriae* type 1 and *S. flexneri* from faeces was demonstrated." (Authors' abstract)

437 Seriwatana J, Echeverria P, Taylor DN, Sakuldaipera T, Changchawalit S, Chivoratanond O. Identification of enterotoxigenic *Escherichia coli* with synthetic alkaline phosphatase-conjugated oligonucleotide DNA probes. *J Clin Microbiol* 1987 Aug;25(8):1438-41

"Alkaline phosphatase-conjugated (AP) 26-base oligonucleotide DNA probes were compared with the same probes labeled with γ - ^{32}P for the identification of heat-labile (LT) and heat-stable (ST) enterotoxigenic *Escherichia coli* (ETEC). The AP oligonucleotide probes were as sensitive as the radiolabeled (RL) probes in detecting LT and STA-2 target cell DNA, but the AP ST probe, which differed from STA-1 by two bases, was less sensitive than the RL probe in detecting STA-1 DNA (6.25 versus 0.78 ng). Of 94 ETEC that were identified with the RL probe, the AP probes detected 93% (28 of 30) of ST, 73% (25 of 34) of LT, and 67% (20 of 30) of LTST ETEC. When colony lysates of these ETEC were examined, the AP probes identified all 94 ETEC. In examination of stool blots, the RL and AP probes were shown to have sensitivities of 71 and 59%, specificities of 91 and 86%, positive predictive values of 87 and 73%, and negative predictive values of 86 and 74%, respectively. AP oligonucleotide probes to detect ETEC were less sensitive in detecting ETEC by colony or stool blot hybridization

than the RL probes but could be used by laboratories without access to radioisotopes to examine colony lysates." (Authors' abstract)

438 Sethabutr O, Hanchalay S, Echeverria P, Taylor DN, Leksomboon U. A non-radioactive DNA probe to identify *Shigella* and enteroinvasive *Escherichia coli* in stools of children with diarrhoea. *Lancet* 1985 Nov 16;2(8464):1095-7

A non-radioactive biotinylated DNA probe was constructed to detect *Shigella* and enteroinvasive *Escherichia coli* (EIEC). Stools were collected from children with diarrhoea of less than 72 h duration aged below 10 years. Specimens were examined with the biotinylated probe after removing streptavidin-binding glycoproteins with proteinase K. To determine the relative sensitivity of the biotinylated vs radiolabelled probes, decreasing concentrations of multicopy recombinant plasmid containing the 17 kb fragment were examined with both probes by hybridization for 24 h. Both probes detected 125 pg of target-cell DNA after hybridization for 24 h and exposure to indicator dyes or X-ray film for 4 h. Both probes hybridized with 52 EIEC strains examined. They also hybridized with stool blots from 11 of 13 children with culture-proven shigellosis of EIEC diarrhoea. Stools from 43 children from whom *Shigella* and EIEC were not isolated did not hybridize with either probe. Biotinylated DNA probes can be as sensitive as radiolabelled probes, but have the advantage of a longer shelf-life and greater availability. Since biotinylated probes and the assay reagents can be lyophilized and transported at room temperature it is possible for laboratories in the developing countries to examine specimens in DNA hybridization assays.

439 Sethabutr O, Unicomb LE, Holmes IH, Taylor DN, Bishop RF, Echeverria P. Serotyping of human group A rotavirus with oligonucleotide probes. *J Infect Dis* 1990 Aug;162(2):368-72

"Rotaviruses (RV) in stools of children with diarrhea in Thailand were serotyped by monoclonal enzyme immunoassay (MEIA), and RNA extracted from these specimens were tested for hybridization with oligonucleotides constructed from the nucleotide sequences of VP7 of human serotypes 1, 2, 3, and 4. Of 178 specimens that contained RV as identified with a monoclonal antibody to group A RV, 84% (149/178) hybridized with serotype-specific oligonucleotides, and 42% (74/178) were serotyped by MEIA ($p < .001$). Of the 74 specimens that were serotyped by MEIA, 92% (35/38) of type 1, 97% (34/35) of type 2, and the one type 4 RV hybridized with the HuG1Ac, HuG2Ac, and HuG4Ac oligonucleotides, respectively. RV strains identified in children in Thailand in 1987 and 1988 to which a serotype could be assigned by either method were either type 1, type 2, or, less often, type 4. Testing RV for hybridization with oligonucleotides for genes encoding VP7 is an alternate method of determining RV serotypes." (MEDLINE)

440 Shaffer N, do Santos ES, Andreason P, Farmer JJ, 3d. Rapid laboratory diagnosis of cholera in the field. *Trans R Soc Trop Med Hyg* 1989 Jan-Feb;83(1):119-20

441 Shah NM, Jhala VM. Latex agglutination test (Rotalex) for rapid diagnosis of rotavirus in neonatal calf diarrhoea. *Indian J Anim Sci* 1990 Nov;60(11):1311-2

442 Shaw RD, Stoner-Ma DL, Estes MK, Greenberg HB. Specific enzyme-linked immunoassay for rotavirus serotypes 1 and 3. *J Clin Microbiol* 1985 Aug;22(2):286-91

"We prepared monoclonal antibodies against two serotypically distinct rotavirus strains: Wa, a serotype 1 virus of human origin, and rhesus rotavirus, a simian serotype 3 virus. Monoclonal antibodies which react specifically with VP7 of each serotype were identified by hemagglutination inhibition tests, plaque reduction neutralization studies, and solid-phase immunoassays which used wild-type and reassortant strains of rotavirus. An enzyme-linked immunoassay was designed which utilizes two of these antibodies to correctly identify serotype 1 and serotype 3 viruses." (Authors' abstract)

443 Shen DT, Ward AC, Gorham JR. Detection of mink enteritis virus in mink feces, using enzyme-linked immunosorbent assay, hemagglutination, and electron microscopy. *Am J Vet Res* 1986 Sep;47(9):2025-30

"Twenty-five mink were inoculated with mink enteritis virus (MEV). Fecal specimens were collected daily and were simultaneously evaluated for MEV antigen by use of a direct enzyme-linked immunosorbent assay (ELISA), hemagglutination (HA), and electron microscopy. Results of the evaluations indicated that MEV was shed in the feces on postinoculation days 5 and 6. The virus was not detectable by ELISA or HA after postinoculation day 6, although viruses were found in reduced numbers by use of electron microscopy. The ELISA was specific for MEV, and the sensitivity of the ELISA for MEV was comparable with that of HA." (MEDLINE)

444 Shepherd RW, Boreham PFL. Recent advances in the diagnosis and management of giardiasis. *Scand J Gastroenterol* 1989;24(suppl 169):60-4

445 Shetty N, Prabhu T. Evaluation of faecal preservation and staining methods in the diagnosis of acute amoebiasis and giardiasis. *J Clin Pathol* 1988 Jun;41(6):694-9

"The use of a faecal preservative and several staining methods, together with formalin ether concentration, were evaluated for the improved diagnosis of intestinal amoebiasis and giardiasis in 1285 patients with diarrhoea or dysentery and from asymptomatic controls. All samples were screened by three wet mount techniques. Thirty eight specimens of diarrhoeal or dysenteric stool were preserved in polyvinyl alcohol (PVA) and stained by trichrome and Spencer and Monroe short iron haematoxylin stain. Thirty nine preserved faecal samples submitted for routine screening were subjected to formalin ether concentration, wet mount examination, and permanent staining. Saline and buffered methylene blue (BMB) mounts were equally good for detection of trophozoites *Entamoebae* while *Giardia* trophozoites were detected only by the saline mount. The iodine mount was superior to the other mounts for protozoan cyst detection. The concentration procedure enhanced cyst recovery. Faecal preservation and subsequent staining was superior to wet mount examination for detection of the trophozoite stage and avoided the need for fresh specimens. Both the trichrome and the iron haematoxylin stains were comparable for the detection of cysts and trophozoites of the *Entamoebae*. *Giardia lamblia* trophozoites stained better with iron haematoxylin than with the trichrome. Preservation and permanent staining is recommended as the most productive means for the accurate identification of the various protozoan parasites." (Authors' abstract)

446 Shetty P, Dilawari JB, Virk KJ, Sehgal R, Ganguly NK, Mahajan RC. Enzyme-linked immunosorbent assay using excretory/secretory and somatic antigens as a diagnostic test for human hookworm infection. *Trans R Soc Trop Med Hyg* 1988 Sep-Oct;82(5):736-8

447 Shlmada K, Tsuji H. Enrichment for detection of *Campylobacter jejuni*. *J Clin Microbiol* 1986 May;23(5):887-90

This study aimed at the development and evaluation of an enrichment medium for the aerobic detection of small numbers of *Campylobacter jejuni*. Gifu anaerobe-modified (GAM) semi-solid medium, supplemented with vancomycin, polymyxin-B, trimethoprim, and amphotericin-B, can serve as efficient enrichment for the recovery of *C. jejuni* under aerobic conditions. Only 35 faecal samples were used for detection of *C. jejuni* from children with gastroenteritis. *C. jejuni* was isolated from 2 faecal samples by the direct plating method (isolation rate, 5.7%), but enrichment with GAM semi-solid medium containing the antimicrobial agents yielded the bacterium from 6 faecal samples (isolation rate, 17.2%). Thus, it is superior to the direct plating method in its recovery of very small numbers of *C. jejuni* (as few as 3 to cells/g of faeces or millilitres of milk). Blood supplementation, agitation, and elevated temperature are not required for this improved recovery. The antimicrobial agents added have proved useful in suppressing other bacteria found in clinical specimens.

448 Shimada T, Sakazaki R, Fujimura S, Niwano K, Mishina M, Takizawa K. A new selective, differential agar medium for isolation of *Vibrio cholerae* O1:PMT (polymyxin-mannose-tellurite) agar. *Jpn J Med Sci Biol* 1990 Apr;43(2):37-41

449 Shimizu M, Murakami Y, Nishioka N, Satou K, Tsunemitsu H, Tanabe T. Detection of bovine rotavirus by enzyme-linked immunosorbent assay using monoclonal antibody. *Bull Natl Inst Anim Health* 1989 Mar;(93):19-24

450 Shinozaki T, Ushijima H, Tajima T, Kim B, Araki K, Fujii R. Evaluation of four tests for detecting human rotavirus in feces [letter]. *Eur J Pediatr* 1985 Jan;143(3):238

451 Shivananda S, Hordijk ML, Ten Kate FJW, Probert CSJ, Mayberry JF. Differential diagnosis of inflammatory bowel disease: a comparison of various diagnostic classifications. *Scand J Gastroenterol* 1991;26(2):167-73

"Fifty consecutive patients with inflammatory bowel disease of the colon who presented at the University Hospital Rotterdam/Dijkzigt were assessed by four methods: clinical diagnosis, criteria defined by Lennard-Jones and by the Organisation Mondiale de Gastroenterologie (O.M.G.E.) scoring systems, and histologic slide review. All cases were classified into three diagnostic groups: established Crohn's disease (CD), indeterminate colitis, or definite ulcerative colitis (UC). The classifications were compared by kappa analysis. Eighteen of the 50 patients were classified as having established CD by the O.M.G.E. scoring system and Lennard-Jones criteria; 17 were so classified by clinicians, and only 8 by histologic slide review. The agreement among clinician's diagnosis, Lennard-Jones criteria, and the O.M.G.E. scoring system was good (Fleiss-Cohen-weighted kappa; $p < 0.001$). Agreement among histology, Lennard-Jones criteria, and the O.M.G.E. scoring system was less good ($p < 0.05$) and not significantly associated with clinical diagnosis. Histology was less prone to diagnose established CD or established UC and more likely to diagnose indeterminate colitis. This study has shown that the systems of disease definition set out by Lennard-Jones and the O.M.G.E. are comparable and agree well with each other and clinician's diagnosis, but biopsy specimens have a limited diagnostic value in disease differentiation in inflammatory bowel disease." (Authors' abstract)

452 Shockley LJ, Kapke PA, Lapps W, Brian DA, Potgieter LND, Woods R. Diagnosis of porcine and bovine enteric coronavirus infections using cloned cDNA probes. *J Clin Microbiol* 1987 Sep;25(9):1591-6

453 Shukry S, Zaki AM, DuPont HL, Shoukry I, El Tagi M, Hamed Z. Detection of enteropathogens in fatal and potentially fatal diarrhea in Cairo, Egypt. *J Clin Microbiol* 1986 Dec;24(6):959-62

454 Simkins RA, Saif LJ, Weilnu PA. Epitope mapping and the detection of transmissible gastroenteritis viral proteins in cell culture using biotinylated monoclonal antibodies in a fixed-cell ELISA. *Arch Virol* 1989;107(3-4):179-90

"A fixed-cell ELISA was developed using swine testicle (ST) cells infected with the virulent Miller strain of transmissible gastroenteritis virus (TGEV) and purified biotinylated monoclonal antibodies (b-MABs). Five of the b-MABs were specific for the peplomer (E2), five reacted to the nucleocapsid (N), and one reacted to the E1 protein of the Miller strain of TGEV. Protein A-Sepharose purification of MABs yielded protein concentrations ranging from 0.40 to 3 mg per ml of ascites. Separate pools of N-MABs and E2-MABs, and the E1-MAB were used to monitor synthesis of TGE viral antigen in ST cells from 0 to 16 h post-infection at various multiplicities of infection (MOI). Epitopes of N proteins appeared sooner and at a lower MOI than those for the E1 and E2 proteins. The fixed-cell ELISA was also used to examine relative binding affinities of TGEV MABs. Concentrations of b-MABs producing a half-maximal signal ranged from 0.11 to 3.8 microgram/ml for E2-MABs, from 0.05 to 0.82 microgram/ml for N-MABs, and 6 micrograms/ml for the E1-MAB. The assay was used to determine the 50% neutralization concentrations for four neutralizing E2-MABs (0.1 microgram/ml to 6.9

micrograms/ml) and one E 1-MAb (1.2 micrograms/ml). Competition assays between b-MABs and unlabeled competitors indicated that at least two major antigenic sites exist on the E 2-protein and 2 to 3 antigenic sites are present on the N-protein of Miller TGEV." (MEDLINE)

455 Singh V, Broor S, Mehta S. Comparison of reverse passive haemagglutination assay & solid phase agglutination of coated erythrocytes with ELISA for rotavirus antigen detection. Indian J Med Res 1986 Sep;84:327-30

The sensitivity and specificity of reserve passive haemagglutination assay (RPHA) and solid phase agglutination of coated erythrocytes (SPACE) using antibody to simian rotavirus (SA-11) were compared with the enzyme-linked immunosorbent assay (ELISA) for detection of rotavirus antigen in stool samples. A total of 152 faecal specimens from children (0-5 years) suffering from acute gastroenteritis were examined for rotavirus by RPHA, SPACE and ELISA. Twenty-one specimens were positive and 95 negative by all three tests. Twenty-one specimens were positive for rotavirus by RPHA and ELISA, and 9 were positive for rotavirus by ELISA alone. Five false positive results were observed in SPACE and RPHA separately. The false positivity in 6 specimens were confirmed since the agglutination could not be blocked after incubation with hyperimmune serum. As compared to ELISA, RPHA had a sensitivity of 82.3% and a diagnostic accuracy of 90.7%, while SPACE was less sensitive (41%) and less accurate (77%). Both SPACE and RPHA had the same specificity (95%). It is suggested that for higher sensitivity with RPHA and SPACE, antisera to human rotavirus may be used. RPHA, however, is a rapid and economic preliminary screening test for the detection of rotavirus in stool samples.

456 Singh-Naz N, Rodriguez WJ, Kidd AH, Brandt CD. Monoclonal antibody enzyme-linked immunosorbent assay for specific identification and typing of subgroup F adenovirus. J Clin Microbiol 1988 Feb;26(2):297-300

457 Sippel JE, Hanafy HM, Diab AS, Prato C, Arroyo R. Serodiagnosis of typhoid fever in paediatric patients by anti-LPS ELISA. Trans R Soc Trop Med Hyg 1987;81(6):1022-6

"Enzyme-linked immunosorbent assay (ELISA) and immunoblot analysis showed that up to 50% of the anti-typhoid antibody in sera from blood culture positive paediatric typhoid fever patients is directed against lipopolysaccharide (LPS) antigen. Anti-Salmonella typhi LPS ELISA was therefore compared to Widal agglutination for serodiagnosis of typhoid fever in paediatric patients. Sera from 38 paediatric control individuals were ELISA negative for anti-S. typhi LPS IgG; all but 2 of these specimens were negative for anti-S. typhi LPS IgM. Paediatric patients hospitalized with signs and symptoms of typhoid fever were separated into 4 groups and tested by ELISA with the following results: 46 patients negative by both culture and Widal agglutination tests, 48% positive for anti-S. typhi LPS IgG and 35% for anti-S. typhi LPS IgM; 22 negative by culture but with positive Widal titres, 82% and 68% positive respectively; 28 culture positive for S. typhi, 93% and 82% respectively; and 12 culture positive for Salmonella other than S. typhi, 92% and 92% respectively. These data suggest that anti-S. typhi LPS ELISA is a suitable assay for diagnosis of typhoid fever in children." (Authors abstract)

458 Snyder DE. Indirect immunofluorescent detection of oocysts of Cryptosporidium parvum in the feces of naturally infected raccoons (Procyon lotor). J Parasitol 1988 Dec;74(6):1050-2

459 Snyder JW, Cost K. The use of monoclonal antibodies in identification of microorganisms. Clin Microbiol Newslett 1988 Nov 15;10(22):169-71

460 Sommerfelt H, Svennerholm A-M, Kalland KH, Haukanes B-I, Bjorvatn B. Comparative study of colony hybridization with synthetic oligonucleotide probes and

enzyme-linked immunosorbent assay for identification of enterotoxigenic *Escherichia coli*. J Clin Microbiol 1988 Mar;26(3):530-4

"On the basis of the published nucleotide sequences of the genes that code for the heat-labile toxin LTh and the heat-stable toxins STaI and STaII of human enterotoxigenic *Escherichia coli*, a 34-mer and two 33-mer oligonucleotide probes were synthesized. To compare their relative efficacies in the detection and differentiation of enterotoxigenic *E. coli*, a colony hybridization technique using these probes and a GM1 ganglioside enzyme-linked immunosorbent assay using monoclonal anti-LT and anti-ST antibodies were used with 76 strains of *E. coli* with known enterotoxin profiles. For further evaluation of probe specificity, the enterotoxigenic bacteria *Vibrio cholerae* 01 and non-01 and *Yersinia enterocolitica* were examined with the colony hybridization technique. The sensitivity of colony hybridization compared favorably with that of GM1 ganglioside enzyme-linked immunosorbent assay, and the two assays showed a high level of concordance in specific detection and differentiation of *E. coli* with various enterotoxin profiles ($\kappa=0.906$, $P<0.00001$). The probes did not hybridize with DNAs from strains of *V. cholerae* 01 or non-01 or *Y. enterocolitica*." (Authors' abstract)

461 Sommerfelt H, Grewal HMS, Bhan BK. Simplified and accurate nonradioactive polynucleotide gene probe assay for identification of enterotoxigenic *Escherichia coli*. J Clin Microbiol 1990 Jan;28(1):49-54

"The present study describes a colony hybridization setup for identification of enterotoxigenic *Escherichia coli* obviating the need for advanced equipment and radioactive isotopes. With a modest laboratory arrangement, polynucleotide gene probes were produced in large quantities. The probes were labeled with digoxigenin and, after hybridization, detected with an antidigoxigenin alkaline phosphatase conjugate. With an established isotope-based oligonucleotide hybridization assay as reference, a blinded study on a large battery of enterotoxigenic and nonenterotoxigenic bacteria revealed a satisfactory sensitivity and specificity of the nonradioactive assay." (Authors' abstract)

462 Sory M-P, Tollenaere J, Laszlo C, Blot T, Cornelis GR, Wauters G. Detection of pYV+ *Yersinia enterocolitica* isolates by P1 slide agglutination. J Clin Microbiol 1990 Nov;28(11):2403-8

463 Srivastava L, Srivastava VK. Serological diagnosis of typhoid fever by enzyme-linked immunosorbent assay (ELISA). Ann Trop Paediatr 1986 Sep;6(3):191-4

"Serum sample from 22 bacteriologically proved cases of typhoid fever, 41 febrile cases who were culture negative and 70 sick and healthy age-matched controls were tested for enzyme-linked immunosorbent assay (ELISA) IgG and IgM antibodies using *Salmonella typhi* LPS antigen. IgG and IgM antibodies were present in 72.7% and 81.8% respectively as against Widal test which was positive in 40.9% in proved cases. In febrile controls IgG and IgM ELISA antibodies were present in 80.4% and 60.9% respectively as against 53.6% by Widal test. This difference between the two tests was statistically significant $p<0.001$. ELISA test was more sensitive than the Widal test and hence it may be useful in rapid serodiagnosis of typhoid fever and also in circumstances where bacteriological techniques are not available." (MEDLINE)

464 Steinruck H, Samtleben S, Ronnberg B, Wadstrom T. Comparison of a coagglutination test with the GM1-ELISA and the Y-1 adrenal cell assay for the detection of heat-labile enterotoxin from *Escherichia coli*. Zentralbl Bakteriol Mikrobiol Hyg [A] 1985 Dec;260(4):436-42

"A number of different assay methods has been developed for the detection of the heat-labile enterotoxin (LT) of ETEC strains isolated from humans and animals. In the present study 40 *Escherichia coli* strains were subject of a comparative study of the staphylococcal coagglutination (Coa-LT) test, the GM1-ELISA and the Y-1 adrenal cell

test. All but two of 20 "LT-ST" or "LT-only" producing ETEC strains gave identical results in both the Coa-LT test and the bioassay. None of the 14 "ST only" producing ETEC strains gave false positive results. It is concluded that the Coa-LT test is a simple and rapid assay method for the routine diagnosis of LT producing ETEC strains." (MEDLINE)

465 Stibbs HH, Samadpour M, Manning JF. Enzyme immunoassay for detection of *Giardia lamblia* cyst antigens in formalin-fixed and unfixed human stool. J Clin Microbiol 1988 Sep;26(9):1665-9

466 Stibbs HH. Monoclonal antibody-based enzyme immunoassay for *Giardia lamblia* antigen in human stool. J Clin Microbiol 1989 Nov;27(11):2582-8

"A visually readable monoclonal antibody-based antigen-capture enzyme immunoassay for the detection of *Giardia lamblia* antigen in human stool specimens was developed and found to be 97% (30 of 31 stool specimens) sensitive for formalinized stools and 82% (49 of 60 stool specimens) sensitive for unfixed stool specimens by visual reading. The storage of specimens in 10% formalin resulted in increased absorbance in 20 of 26 *G. lamblia*-positive specimens tested as both formalinized and unfixed specimens; the increase averaged 1,336%. The assay was specific for antigens of this organism and for antigens derived from the cyst, as opposed to the trophozoite, stage. The assay could detect the antigens of five cysts per well, but could not detect antigen in in vitro-cultured trophozoites. A mouse monoclonal antibody of the immunoglobulin G1 (IgG1) subclass, which was prepared against cysts of *G. lamblia*, was used as the solid-phase capture antibody. The antibody was reactive with the cyst wall, as determined by immunofluorescence. Polyclonal rabbit anti-cyst IgG was used as the secondary antibody, and peroxidase-labeled goat anti-rabbit IgG was used as the tertiary antibody in the assay format. Maximal capture of antigen from stool specimens occurred by 30 min. Optimal dilution of specimens was in the range of 1:60 to 1:600. Preliminary characterization of affinity-purified antigen recognized by the monoclonal antibody showed that it is heat stable (100°C, 12 min) and resistant to sodium periodate treatment and that it may exist in multiple molecular weights from 45,000 to 110,000." (Authors' abstract)

467 Strat E, Tansanu I, Gotia S, Moraru E, Combiescu AA, Persu A, Boisteanu DH, Diaconu G, Dusa MF. Detection of group rotavirus antigen with Romanian ELISA kits in the acute gastroenteritis of the infant and young child. Rev Med Chir Soc Med Nat Iasi 1986 Apr-Jun;90(2):323-5

468 Sugiyama J, Gondalra F, Matsuda J, Soga M, Terada Y. New method for serological typing of *Vibrio cholerae* 01: using a monoclonal antibody-sensitized latex agglutination test. Microbiol Immunol 1987;31(4):387-91

A new method for serological identification of *Vibrio cholerae*, using a monoclonal antibody-sensitized latex agglutination test, is described. *V. cholerae* strains belonging to serogroup 0:1 were known to have caused epidemic cholera in Japan. The serogroup had three serotypes, Ogawa, Inaba, and Hikojima. For the identification of *V. cholerae* 0:1 and its serotypes, diagnostic antisera produced by immunisation of rabbits were available. Of the three monoclonal antibodies, viz. anti-a, anti-b and anti-c tested, anti-a agglutinated both type Inaba and Ogawa, anti-b agglutinated only type Ogawa and anti-c agglutinated only type Inaba. These monoclonal antibodies did not agglutinate 11 strains of non-*V. cholerae* 01. The latex agglutination test was done with 68 strains of *V. cholerae* 01 (52 strains of type Ogawa, 15 of type Inaba, and 1 of type Hikojima), and 11 strains of non-*V. cholerae* 01, 10 of *Escherichia coli*, 10 of *Salmonella choleraesuis*, and 10 of *Shigella flexneri*. All Ogawa strains showed positive agglutination of latexes A and B, whereas all Inaba strains showed positive agglutination of latexes A and C. No agglutination was found with non-*V. cholerae* 01, *E. coli*, *S. choleraesuis*, and *S. flexneri* tested with Hikojima. Sensitivity of each sensitised latex

showed that latex A gave detectable agglutination reactions at a concentration of 5×10^6 colony-forming units per ml for both type Ogawa and Inaba, latex B at a concentration of 1×10^7 colony-forming units per ml for type Ogawa only, and latex C at a concentration of 3×10^7 colony-forming units per ml for type Inaba. It is concluded that the sensitivity of the latex reagents is sufficient for the detection of *V. cholerae* 01 in stools of cholera patients.

469 Svennerholm A-M, Wikstrom M, Lindblad M, Holmgren J. Monoclonal antibodies against *Escherichia coli* heat-stable toxin (STa) and their use in a diagnostic ST ganglioside GM1-enzyme-linked immunosorbent assay. *J Clin Microbiol* 1986 Oct;24(4):585-90

"Seven monoclonal antibodies (MAbs) against heat-stable enterotoxin (ST) from a human *Escherichia coli* isolate were prepared and evaluated for their usefulness in an ST immunodetection assay, the ST ganglioside GM1-enzyme-linked immunosorbent assay (ELISA). This assay is based on the ability of STa, as present in, for example, culture filtrates from ST-producing *E. coli*, to inhibit specific anti-ST antibody from binding to solid-phase-bound ST ganglioside (GM1-bound ST-cholera B subunit). Four of the MAbs were of immunoglobulin G1 (IgG1), one was of IgG2b, and two were of IgM isotype. All the IgG1 MAbs could be completely inhibited by addition of free ST; 0.2 to 0.4 ng of purified ST inhibited binding of these MAbs by 50%. The non-IgG1 MAbs were, in contrast, not inhibited by 200-fold-higher amounts of purified ST, probably because they were directed against linkage epitopes or were of low affinity or both. When the IgG1 MAbs were tested in the ST GM1-ELISA, ST could be detected in culture filtrates from stock human *E. coli* isolates with 100% sensitivity and specificity. ST in filtrates from fresh stool cultures was demonstrated with higher sensitivity with the MAb ST GM1-ELISA than with the conventional infant mouse test. Both subtypes of STa, STaI and STaII, could be detected by the ST GM1-ELISA by using either IgG1 MAb in the immunodetection step, whereas infant-mouse-active ST from *Yersinia enterocolitica* failed to react." (Authors' abstract)

470 Svoboda I, Rodak L, Franz J, Stepanek J, Valicek L. [Differential diagnosis of viral diarrheas in calves] *Vet Med (Praha)* 1986 Aug;31(8):497-504

"There is a description of the enzyme-immunologic method (ELISA), which was used for demonstration of rotaviruses and coronaviruses in the samples of excrements of the calves suffering from diarrheas. It is shown by a comparison with the results obtained by electron microscopy that the ELISA method provides by up to 50% higher capture rate and the reaction is highly specific. The method can also be applied to a detection of human rotavirus in the children's stool." (MEDLINE)

471 Tabouret M, DeRycke J. Detection of cytotoxic necrotising factor (CNF) in extracts of *Escherichia coli* strains by enzyme-linked immunosorbent assay. *J Med Microbiol* 1990 Jun;32(2):73-81

"An enzyme-linked immunosorbent assay (ELISA) consisting of a double sandwich technique with rabbit and sheep antibodies, was developed for the detection of cytotoxic necrotising factor (CNF) in extracts of *Escherichia coli* strains. The assay was evaluated by comparison with the results obtained with an assay based on toxicity for HeLa cell cultures. In a study of extracts of 27 CNF+ and 45 CNF- strains obtained by ultrasonic disintegration, no false positive and only three false negative results were recorded; the latter were obtained with strains that produced less CNF than any of the others examined. Frozen-thawed extracts contained about four times more CNF cytotoxic activity than extracts prepared ultrasonically; the testing of 54 CNF+ and 68 CNF- frozen-thawed extracts resulted in no false positive and only one false negative response. Whichever type of extract was used, no significant cross-reaction was observed with heat-labile (LT) or heat stable (ST) enterotoxin, verotoxin (VT1, VT2), haemolysin, or Vlr cytotoxin." (Authors' abstract)

472 Tadese T. Survey of rotavirus infections in acute gastroenteritis in Addis Ababa: a comparison between enzyme linked immunosorbent assay (ELISA) and Immunoelectroosmophoresis (IEOP). East Afr Med J 1985 Aug;62(8):560-4

"A total of 217 (181 acute gastroenteritis cases and 36 non-gastroenteritis controls) stool samples were collected. A comparison of two methods (ELISA and IEOP) was made for the detection of rotavirus antigen from the stool samples. The validity of the tests was corroborated by the finding of rotavirus infections at various age groups and specific peak months of the study period. ELISA was shown to be more sensitive and could be of high value for routine diagnosis of rotavirus in acute gastroenteritis." (Author's abstract)

473 Takiff HE, Seidl M, Krause P, Rooney J, Brandt C, Rodriguez W, Yolken R, Straus SE. Detection of enteric adenoviruses by dot-blot hybridization using a molecularly cloned viral DNA probe. J Med Virol 1985 Jun;16(2):107-18

"Enteric adenoviruses (EAd) (candidate adenoviruses 40 and 41, subgroups F and G) have been implicated in the etiology of gastroenteritis in infants, but their clinical significance has been unclear because a rapid test to distinguish these agents from other adenovirus (Ad) types has not been available. We developed a dot-blot hybridization assay for EAd DNA using a cloned DNA fragment that has little homology to non-EAd DNAs. The dot-blot system detected less than 20 pg of EAd DNA, while showing minimal cross hybridization to representative strains from all other Ad groups. There was no detectable hybridization to extracts of samples known to contain other enteric viruses. It was further shown that low levels of EAd in specimens could be amplified by culturing for 1 day in 293 cells. Stool samples and tissue culture lysates prescreened by electron microscopy, cell culture or ELISA were tested in a blind fashion. Using endonuclease analysis as the standard for typing the isolates, we found the dot-blot system to have a 91% sensitivity and 71% specificity for detecting EAd and distinguishing them from other Ads. False-positive and equivocal dot-blot results appeared to be caused by other Ads." (Authors' abstract)

474 Talwar V, Kaur I, Gupta HC. Counter Immunoelectrophoresis (CIEP) for serological diagnosis of typhoid fever. Indian J Med Res 1986 Oct;84:353-7

Ultrasonic lysate and veronal-buffer extract antigens of *Salmonella typhi* were used for detection of antibody by counter-immunoelectrophoresis (CIEP) in sera of 49 *S. typhi* culture-positive patients (group I), 50 febrile controls (group II) where causes other than typhoid were established and 96 afebrile healthy controls (group III) with no history of TAB vaccination or typhoid fever in the last one year. Positivity of 87.7% using ultrasonic lysate antigen and 69.3% with veronal-buffer extract antigen (VBE) in CIEP was seen as compared to 69.3% by Widal test, in 49 *S. typhi* culture-positive patients. The sensitivity of the CIEP test using ultrasonic lysate was 87.7% and specificity was 91% as compared to 69.3% and 87.6% with VBE antigen. The sensitivity and specificity of Widal test was 69.3% and 89% respectively. Attempts to detect antigen from serum by CIEP using commercial *S. typhi* 'O' antiserum, and the hyperimmune antiserum prepared in the laboratory revealed negative results in the culture-positive patients. Although CIEP gives quicker results, it requires special apparatus and antigen; hence it cannot score much over the conventional Widal test for the diagnosis of typhoid fever.

475 Taniguchi K, Urasawa T, Morita Y, Greenberg HB, Urasawa S. Direct serotyping of human rotavirus in stools by an enzyme-linked immunosorbent assay using serotype 1-, 2-, 3-, and 4-specific monoclonal antibodies to VP7. J Infect Dis 1987 Jun;155(6):1159-66

"Four serotypes of human rotaviruses that can be differentiated by neutralization tests have been described so far. We prepared serotype 1-, 2-, 3-, and 4-specific, neutralizing monoclonal antibodies to human rotaviruses. All were directed to VP7, a

glycoprotein that carries a major serotype specificity. An enzyme-linked immunosorbent assay using these monoclonal antibodies has been developed for serotyping human rotavirus isolates. The sensitivity and specificity of this method were verified by using various strains of human rotavirus that were adapted to cell culture. Furthermore, it was shown that the method was applicable to serotyping human rotaviruses directly in stool specimens." (Authors' abstract)

476 Tantivanich S, Suwannasopone N, Chaicumpa W, Wattanavijarn W, Leksomboon U. Comparison of ELISA with electron microscopy, and PAGE for the detection of human rotavirus in stools. *J Med Assoc Thai* 1988 Aug;71(8):436-42

"A total of 363 diarrheic stool specimens were collected from May 1985 to April 1986 from children aged 5 days to 12 years and 4 months who had acute diarrhea and came to Bangkok children's hospital for treatment. Rotaviruses were found in 27.27 per cent of the patients by enzyme-linked immunosorbent assay (ELISA). The highest incidence of the rotavirus diarrhea was in the month of January which was cool and dry and was found among the children aged between 6 months and 2 years old. The incidence between males and females was 1.54:1. Most of the rotavirus diarrheic patients were from low income families. Nine different RNA electropherotypes were found among the rotaviruses isolated. Seventy per cent were of long pattern of which the electropherotype A predominated throughout the year course of study. ELISA is the most sensitive method for the clinical diagnostic purpose while the electropherotyping is suitable for epidemiological study" (Authors' abstract)

477 Taylor DE, Chang N. Immunoblot and enzyme-linked immunosorbent assays of *Campylobacter* major outer-membrane protein and application to the differentiation of *Campylobacter* species. *Mol Cell Probes* 1987 Sep;1(3):261-74

"The major outer-membrane protein (MOMP) from *Campylobacter jejuni* NCTC 11168 was purified by solubilization in Triton X-100. Whole-cell proteins of *Campylobacter* species and the MOMP were subjected to electrophoresis on sodium dodecyl sulfate-polyacrylamide gels. Immunoblotting on nitrocellulose paper and enzyme-linked immunoblot assay (ELISA) analyses using polyclonal antiserum to the *C. jejuni* MOMP. Purified MOMP from *C. jejuni* NCTC 11168 contained a single major protein of 46 kDa. Whole-cell preparation of *C. jejuni* NCTC 11168 also showed a major band of 46 kDa which was recognized in immunoblots by the anti-MOMP serum. Other *C. jejuni* strains, as well as *C. coli* and *C. lariidis* strains showed similar MOMP bands at 45-46 kDa. Other *Campylobacter* species (*C. fetus* ss. *fetus*, *C. hyointestinalis* and *C. pylori* (new nomenclature) did not react in immunoblots with antiserum to the *C. jejuni* MOMP. ELISAs showed that antiserum raised against the *C. jejuni* MOMP reacted with *C. jejuni*, *C. coli* and *C. lariidis* strains. Other *Campylobacter* species displayed only a very low degree of cross-reactivity. The distinct antigenic relationship demonstrated between the MOMP of *C. jejuni*, *C. coli* and *C. lariidis* is consistent with the close degree of relatedness of these species as determined by DNA homology studies. The anti-MOMP serum appeared to be useful in the rapid differentiation of *C. jejuni*, *C. coli* and *C. lariidis* from other *Campylobacter* species." (MEDLINE)

478 Tenover FC. Diagnostic deoxyribonucleic acid probes for infectious diseases. *Clin Microbiol Rev* 1988 Jan;1(1):82-101

479 Thanikachalam Y, Santhanakrishnan BR, Lakshminarayana CS. Evaluation of coagglutination test for rapid diagnosis of typhoid. *J Trop Pediatr* 1986 Dec;32(6):301-3

"Twenty-nine children with typical clinical picture of typhoid and 15 with atypical manifestations were investigated for typhoid by conventional tests like blood culture, Widal test, urine culture, and the results were compared with coagglutination tests of urine, serum, and cerebrospinal fluid. Coagglutination of urine was positive in all cases, earlier than significant titre by serology. It was negative in control subjects and in the urine of

patients suffering from urinary tract infections due to *E. coli* and other common organisms which cause urinary tract infection. The test became negative earlier in patients treated with multiple drug therapy than those treated with single drug therapy." (Authors' abstract)

480 Thomas EE, Puterman ML, Kawano E, Curran M. Evaluation of seven immunoassays for detection of rotavirus in pediatric stool samples. *J Clin Microbiol* 1988 Jun;26(6):1189-93

"The performance of seven commercially manufactured rotavirus assays was evaluated with 144 pediatric stool specimens and compared with electron microscopy (EM) findings. The four enzyme-linked immunosorbent assays used were Rotazyme II, Pathfinder, IDL rotavirus immunoassay, and Enzygnost (Behring) rotavirus assay. The three latex tests were Meritec rotavirus detection test, Virogen Rotatest, and Bartels rotavirus latex test. Test outcomes were compared with EM on the basis of sensitivity, specificity, positive-negative predictive value, and the kappa statistic. Relative to EM, Meritec had the highest specificity (97%), followed by Virogen (95%), IDL (91%), Pathfinder (85%), Behring (81%), Bartels (72%), and Rotazyme (71%). The sensitivities were as follows: Rotazyme (92%), Pathfinder (89%), Bartels (86%), Virogen (86%), Behring (82%), Meritec (71%), and IDL (75%). Patient age and sex did not influence test results. Owing to the absence of a true standard, the tests were also compared with each other on the basis of the kappa statistic, the frequency of positive test results, and the frequency of samples in which a test differed from all other tests. Using these measures, the assays could be classified into three groups with progressively decreasing utility: group 1 (Virogen, Meritec, IDL, and EM), group 2 (Pathfinder and Behring), and group 3 (Rotazyme and Bartels). Laboratory criteria were also compared. Latex tests were faster and required less equipment than enzyme-linked immunosorbent assays. The Virogen latex assay showed the best overall performance, which made it our choice for rapid and accurate rotavirus diagnosis. However, in children who have gastrointestinal symptoms with negative rotavirus test results, EM will be useful until such time as immunological tests for other enteric viruses are available." (Authors' abstract)

481 Thomas JE, Whatmore AM, Barer MR, Eastham EJ, Kehoe MA. Serodiagnosis of *Helicobacter pylori* infection in childhood. *J Clin Microbiol* 1990 Dec; 28(12):2641-6

482 Thompson JS, Hodge DS, Smith DE, Yong YA. Use of tri-gas incubator for routine culture of *Campylobacter* species from fecal specimens. *J Clin Microbiol* 1990 Dec;28(12):2802-3

"We evaluated a tri-gas incubator for *Campylobacter* isolation to be used instead of an anaerobic jar. Fecal specimens were cultured in duplicate onto charcoal selective medium and incubated at 43°C for 48 h in two different environments: a tri-gas incubator (Forma Scientific) adjusted to provide an atmosphere of 10% CO₂, 10% O₂, and the balance N₂; and evacuated anaerobic jars with a replacement gas mixture of 10% CO₂, 5% O₂, and 85% N₂. A total of 106 *Campylobacter jejuni* and 8 *Campylobacter coli* isolates were obtained from 2,348 stool specimens. Of the positive specimens, 113 isolates came from the incubator and 111 isolates came from the anaerobic jars. An additional 32 previously positive specimens were replated onto charcoal selective medium and retested by both methods. We recovered 27 *C. jejuni* isolates, 26 isolates by each method. The isolates from the incubator typically produced discrete colonies, while the isolates from the anaerobic jar showed some degree of swarming in colony formation. The tri-gas incubator provided a cost-effective method for culturing *Campylobacter* spp." (Authors' abstract)

483 Thompson MR, Jordan RL, Luttrell MA, Brandwein H, Kaper JB, Levine MM, Giannella RA. Blinded, two-laboratory comparative analysis of *Escherichia coli* heat-stable enterotoxin production by using monoclonal antibody enzyme-linked immunosorbent assay, radioimmunoassay, suckling mouse assay, and gene probes. *J Clin Microbiol* 1986 Nov;24(5):753-8

"Heat-stable enterotoxin (ST) - producing enterotoxigenic *Escherichia coli* (ETEC) can be identified by a variety of assays, including the suckling mouse assay (SMA), radioimmunoassay (RIA), polyclonal or monoclonal antibody enzyme-linked immunosorbent assay (ELISA), and DNA hybridization with STh and STp gene probes. To compare the sensitivity and reliability of these assays, 100 coded ETEC and non-ETEC isolates were blindly tested in two independent laboratories. SMA, RIA, and monoclonal ELISA were performed in Cincinnati, Ohio, while gene probe analysis was performed in Baltimore, Md. The method of storage of organisms had a profound effect on the stability of plasmids in certain strains. Hybridization experiments to determine the presence or absence of the enterotoxin gene showed that strains stored on Dorset egg medium at room temperature better retained their plasmids than strains stored frozen in skim milk. Forty-four of the 100 organisms obtained from the skim milk stock were found to produce STa in liquid culture by the RIA, SMA, and monoclonal ELISA (100% agreement). However, 50 of 54 of the strains stored on Dorset egg medium which were originally classified as STa+ or ST+ LT+ (positive for both heat-stable and heat-labile [LT] enterotoxins) were found to produce STa and retain the plasmid by each of these assays. Three additional strains were found which harbored the plasmid but did not elaborate STa by any of the assays (3% discrepancy). The monoclonal antibody ELISA appears to be highly reliable for determination of STa production by ETEC and can be easily scored visually even by untrained personnel. Furthermore, when this STa assay is coupled with a polyclonal antibody assay, it is possible to predict the genotype of STh- and STp-producing organisms." (Authors' abstract)

484 Thorns CJ, Sojka MG, Chasey D. Detection of a novel fimbrial structure on the surface of *Salmonella enteritidis* by using a monoclonal antibody. *J Clin Microbiol* 1990 Nov;28(11):2409-14

485 Todd LS, Roberts D, Bartholomew BA, Gilbert RJ. Assessment of an enzyme immunoassay for the detection of salmonellas in foods and animal feeding stuffs. *Epidemiol Infect* 1987 Jun;98(3):301-10

"The *Salmonella* Bio-EnzaBead Screening Kit, in its modified form with both the MOPC 467 and the 6H4 antibodies, was used for the detection of salmonellas in naturally contaminated foods and animal feeding stuffs in parallel with a traditional cultural procedure. Initial results showed an 82% agreement between the enzyme immunoassay (EIA) and cultural methods when using the criterion recommended by the manufacturer as a cut-off for all types of foods. By adjusting the cut-off for each type of food, the number of EIA positive, culture negative samples was reduced although the number of EIA negative, culture positive samples increased. The EIA may be more sensitive than the cultural methods as in many cases the EIA positive, culture negative results could be real positives which were not detected by the cultural methods. The screening kit provides a simple and convenient method for the detection of *Salmonella* in foods and feeds and a presumptive positive result can be reported within 48 h. The advantages and disadvantages of the method are discussed." (Authors' abstract)

486 Toranzo AE, Santos Y, Nieto TP, Barja JL. Evaluation of different assay systems for identification of environmental *Aeromonas* strains. *Appl Environ Microbiol* 1986 Mar;51(3):652-6

487 Toth I, Barrett TJ, Cohen ML, Rumschlag HS, Green JH, Wachsmuth IK. Enzyme-linked immunosorbent assay for products of the 60-megadalton plasmid of *Escherichia coli* serotype O157:H7. *J Clin Microbiol* 1991 May;29(5):1016-9

"Eighty strains of pathogenic *Escherichia coli*, representing each of the major diarrheal disease-causing groups, were examined by direct enzyme-linked immunosorbent assay (ELISA) for the presence of proteins associated with a 60-MDa plasmid from *E. coli* serotype O157:H7. Antiserum specific for plasmid-encoded proteins was prepared by immunizing a rabbit with a wild-type *E. coli* O157:H7 strain (strain 7785) and absorbing

the serum with a plasmid-cured derivative (strain 2-45). Use of this antiserum in Western immunoblot analysis detected two proteins of 82 and 92 kDa in strain 7785 but not in strain 2-45. All 16 wild-type *E. coli* 0157:H7 strains and all 10 Shiga-like toxin (SLT)-producing *E. coli* strains of serotypes other than 0157 were ELISA positive. Thirteen of 14 enterotoxigenic and all of 24 enteroinvasive *E. coli* strains were ELISA negative, as were all of 16 *E. coli* strains isolated from healthy persons. Of 16 traditional enteropathogenic *E. coli* (EPEC) serotypes, 10 were ELISA positive, including 10 of 12 strains carrying the EPEC adherence factor gene. Absorption of the serum with an EPEC adherence factor-positive EPEC eliminated EPEC reactivity. This study demonstrates that two plasmid-mediated proteins are common to *E. coli* 0157:H7 strains and to SLT-producing strains of other serotypes. Detection of these proteins by ELISA provides a sensitive and specific screening test for identifying SLT-producing *E. coli* of both 0157 and non-0157 serotypes. Identification of the cross-reactive proteins found in EPEC could provide the basis for a single assay to detect both EPEC and SLT-producing *E. coli*." (Authors' abstract)

488 Treanor JJ, Madore HP, Dolin R. Development of an enzyme immunoassay for the Hawaii agent of viral gastroenteritis. *J Virol Methods* 1988 Dec;22(2-3):207-14

"The Hawaii agent is a Norwalk-like virus of acute gastroenteritis in humans which is antigenically distinct from the prototype Norwalk agent. We established a solid phase sandwich type microtiter enzyme immunoassay (EIA) for Hawaii antigen employing sera and stools from experimentally challenged volunteers as reagents. This assay detected the Hawaii agent in stools from 3 of 8 volunteers who were ill after oral challenge with the Hawaii agent, including one specimen which was positive to a dilution of 1/320. Virus shedding occurred on days 3 to 7 after challenge. The Hawaii-positive stools did not react in the EIAs for Norwalk and Snow Mountain agent (SMA), nor did Norwalk or SMA-positive stools react in the Hawaii EIA. Human rotavirus, enteric adenovirus, feline calicivirus, and several enteroviruses also did not react in the Hawaii EIA. A blocking EIA to detect serum antibody to the Hawaii agent was also developed employing a diarrheal stool containing Hawaii as a source of antigen. Serum antibody rises were detected in 15 of 16 individuals with experimentally induced illness after challenge and in 3 of 5 individuals who remained well after challenge. The EIA for the Hawaii agent should permit epidemiologic studies of the Hawaii agent to be carried out as well as allow further characterization of the Hawaii virion." (MEDLINE)

489 Tregub AV, Rubtsov IV, Etkin AF, Iankina ZK. [Comparison of the diagnostic value of methods for determining antibodies against the *Salmonella* group-B O-antigen]. *Zh Mikrobiol Epidemiol Immunobiol* 1986 Jan;33(1):24-9

"The analysis of serum samples from 124 patients with the bacteriologically confirmed diagnosis of group B salmonellosis has revealed that the specific neutralization variant of the enzyme immunoassay (EIA) makes it possible to detect IgA, IgG and IgM more effectively than the indirect EIA variant and the passive hemagglutination test." (MEDLINE)

490 Tregub AV, Pokrovskii VV. [Immunoenzyme analysis of the serological activity of the structural modifications of a synthetic antigen simulating the O-determinant 4 of salmonellae]. *Zh Mikrobiol Epidemiol Immunobiol* 1985 Jul;21(7):39-44

"The comparison of the copolymer of 2-O-(alpha-abequosyl)-alpha-aryl-alpha-mannopyranoside and acrylamide (AMA), used as a synthetic antigen, with its natural prototype, *Salmonella typhimurium* lipopolysaccharide, in the enzyme immunoassay (EIA) has revealed that the serological activity of AMA is related to the specific content of antigenic determinants in its molecule. The analysis of the serological specificity of AMA indicates that this specificity corresponds to factor 4 of *Salmonella* O-antigen. The high activity and monofactor specificity of the synthetic antigen AMA suggest that the use of this antigen in EIA as a diagnostic preparation holds good promise." (MEDLINE)

491 Tregub AV, Etkin AF. [Quantitative determination of the soluble O antigen of *Salmonella* serogroup B and of specific antibodies in different classes by using immunoenzyme analysis]. Zh Mikrobiol Epidemiol Immunobiol 1987 Sep;34(9):67-71

"The results of measurements of *S. typhimurium* O-antigen and specific IgA-, IgG- and IgM-antibodies in 115 serum samples from patients with salmonellosis induced by group B *Salmonellae* are analyzed. As determined in this study, the concentrations of IgA-antibodies ranged 0-15 micrograms/ml, the minimal diagnostically significant value being 3.6 micrograms/ml; the concentrations of IgG-antibodies ranged 0-13 micrograms/ml, the minimal diagnostically significant value being 1.3 micrograms/ml; and the concentrations of IgM-antibodies ranged 0-50 µg/ml, the minimal diagnostically significant value being 5.0 µg/ml. The minimal diagnostically significant concentration of O-antigen, determined in selected serum samples obtained from healthy donors, was 0.1 µg/ml. There is a significant correlation between the concentrations of IgA-, IgG- and IgM-antibodies, but the concentrations of these antibodies do not correlate with the concentration of soluble O-antigen. The study showed that the simultaneous determination of *S. typhimurium* O-antigen and specific antibodies in the material under test significantly enhances the effectiveness of the serodiagnosis of the disease." (MEDLINE)

492 Triasuchev IO, Davydov AG, Burkin VS, Pokrovskii VI, Pukhova TV. [Development and use of solid-phase test-systems for the detection of O-antigen of *Shigella sonnei* and *Shigella flexneri*]. Zh Mikrobiol Epidemiol Immunobiol 1989 Jun(6):74-7

"The possibility of the diagnosis of dysentery caused by *S. sonnei* and *S. flexneri*, as well as the determination of the dynamics of the distribution of specific O-antigen in the patient's body, by means of the enzyme-linked immunosorbent assay system developed on the basis of antibody preparations obtained by immunosorption has been studied. The study has shown that for better diagnosis the use of fecal extracts is preferable in assays; when used in combination with bacteriological analysis, these assays make it possible to increase the confirmation of the diagnosis of dysentery by several fold." (MEDLINE)

493 Trofimov EN, Efremenko VI, Narbutovich NI, Pushkar' VG. [Use of ganglioside-containing magnetic polyacrylamide sorbents for the immunoenzyme method of determining cholera enterotoxin]. Zh Mikrobiol Epidemiol Immunobiol 1987 Feb;109(2):57-60

"A variant of EIA techniques for the determination of cholera enterotoxin is proposed. This method is based on the selective sorption of the toxin on ganglioside-containing magnetic granules with its subsequent detection by means of immune serum and antispesific immunoperoxidase conjugate. The proposed method permits the detection of 0.052 +/- 0.02 ng of protein of the purified toxin." (MEDLINE)

494 Tsukamoto T, Otsu K, Kinoshita Y, Onaka T, Honda T, Miwatani T. [Outbreaks of food poisoning caused by enterotoxigenic *Escherichia coli* and detection of heat-labile enterotoxin in stools by an enzyme-linked immunosorbent assay]. Kansenshogaku Zasshi 1986 Oct;60(10):1119-24

495 Ungar BLP, Nash TE. Cross-reactivity among different *Giardia lamblia* isolates using immunofluorescent antibody and enzyme immunoassay techniques. Am J Trop Med Hyg 1987 Sep;37(2):283-9

"Rabbit antisera to 11 *Giardia lamblia* isolates were reacted with 8 *G. lamblia* isolates using single antibody ELISA and indirect immunofluorescent antibody techniques (IFA). Using living trophozoite organisms, IFA showed marked surface fluorescence with homologous antisera-organism pairs while heterologous pairs showed reduced or no reactivity. Using formalin-fixed trophozoites, the pattern of fluorescence changed to

include diffuse internal fluorescence with both homologous and heterologous pairs. In contrast to the variability in surface fluorescence, similar reactivity was noted for homologous and heterologous antisera-isolate pairs with the ELISA. In addition, a double antibody enzyme immunoassay using rabbit antisera prepared to 2 *G. lamblia* isolates was performed with 8 different isolates as test antigen. All 8 were equally well detected. These data confirm previous findings that *G. lamblia* isolates have both different and common antigens. The different antigens appear to be on the surface of the organism while the common antigens appear to be internal or somatic." (Authors' abstract)

496 Ungar BLP, Soave R, Fayer R, Nash TE. Enzyme immunoassay detection of Immunoglobulin M and G antibodies to *Cryptosporidium* in Immunocompetent and Immunocompromised persons. J Infect Dis 1986 Mar;153(3):570-8

"*Cryptosporidium* is a parasite of the human gastrointestinal tract and has a worldwide distribution. We developed a sensitive and reproducible enzyme immunoassay for detection of serum IgG or IgM to *Cryptosporidium*. For IgG, 13 of 15 patients with cryptosporidiosis and 26 of 26 patients with cryptosporidiosis and AIDS were positive, whereas 57 of 60 presumably uninfected individuals were negative. All three IgG-positive presumably uninfected individuals had been potentially exposed. Sensitivity and specificity of this assay was 95%. Patients without AIDS showed an early rise and fall of IgM and later elevation of IgG; some patients with AIDS produced IgM, and all produced IgG. Sera from 9 (20.9%) of 44 Ecuadorian children with diarrhea were positive for both IgM and IgG antibodies; 106 sera from persons with other parasitic illnesses showed a normal distribution for IgG antibody. These ELISA data show that patients without and with AIDS have serum antibody response to *Cryptosporidium* and suggest that exposure to or infection with *Cryptosporidium* is common." (Authors' abstract)

497 Ungar BLP. Enzyme-linked immunoassay for detection of *Cryptosporidium* antigens in fecal specimens. J Clin Microbiol 1990 Nov;28(11):2491-5

"*Cryptosporidium* sp. is a ubiquitous 4- to 6- μ m protozoan parasite infecting the intestinal tract of humans. It causes mild to fulminant diarrhea in patients, especially immunocompromised persons, and it may be hard to detect by microscopic fecal examination. An indirect, double-antibody enzyme-linked immunosorbent assay (ELISA) was developed using specifically produced goat and rabbit antisera to detect *Cryptosporidium* antigens in human feces. Of 62 frozen stools from patients with cryptosporidiosis, as detected by at least two microscopic diagnostic techniques, 51 were positive by ELISA; all ELISA-negative specimens came from patients with fewer than five oocysts per 0.01 ml of concentrated fecal sample examined after modified acid-fast or fluorescent monoclonal antibody staining. A total of 182 specimens from persons without *Cryptosporidium* infection were negative by ELISA in 176 instances; 3 ELISA-positive specimens came from patients with cryptosporidiosis diagnosed earlier. The sensitivity of the assay was 82.3%, and specificity was 96.7%. The predictive value of a positive ELISA was 89.5%, and the predictive value of a negative ELISA was 94.2%. The ELISA was not affected by the presence of eight other intestinal parasites but was sometimes affected by repeated freezing and thawing of fecal specimens. All fecal specimens were heated to 100°C for 2 min to reduce proteolytic enzyme activity, although the necessity of this step needs further evaluation. This first-generation ELISA is a simple, rapid, easily standardized test for *Cryptosporidium* antigens in stool samples which will be useful for diagnosis and for large-scale epidemiologic studies." (Author's abstract)

498 Ungar BL, Yalken RH, Quinn TC. Use of a monoclonal antibody in an enzyme immunoassay for the detection of *Entamoeba histolytica* in fecal specimens. Am J Trop Med Hyg 1985 May;34(3):465-72

"An indirect enzyme-linked immunosorbent assay (ELISA) was developed for the detection of *Entamoeba histolytica* in human feces, using both a monoclonal antibody

and rabbit antisera. It detected from less than 1 to 57 trophozoites of 6 *E. histolytica* strains. Stool specimens were positive by ELISA in 18 of 22 (82%) patients with *E. histolytica* and in 3 of 186 (2%) of patients without demonstrable *E. histolytica* in their stools. The latter included one from a child living near an asymptomatic cyst carrier and another from a traveler with giardiasis who had recently taken antibiotics. One hundred eight of 183 microscopy- and ELISA-negative specimens contained other parasites including *Giardia* (49 specimens), *Endolimax nana* (24), *Entamoeba coli* (21), *Iodamoeba butschlii* (2), and *Entamoeba hartmanni* (1). This ELISA for *E. histolytica* is a simple, sensitive and specific diagnostic tool." (MEDLINE)

499 Urasawa S, Urasawa T, Taniguchi K, Wakasugi F, Kobayashi N, Chiba S, Sakurada N, Morita M, Morita O, Tokieda M, Kawamoto H, Minekawa Y, Ohseto M. Survey of human rotavirus serotypes in different locales in Japan by enzyme-linked immunosorbent assay with monoclonal antibodies. *J Infect Dis* 1989 Jul;160(1):44-51

"To investigate the relative frequency of individual human rotavirus serotypes prevailing in Japan, 562 stool specimens collected from patients with rotavirus gastroenteritis between November 1986 and March 1988 in seven districts were examined by an enzyme-linked immunosorbent assay (ELISA) with serotype 1-, 2-, 3-, and 4-specific monoclonal antibodies. Serotype 1 was the predominant serotype in the winter of 1986-1987; however, both serotypes 1 and 2 were detected frequently in the winter of 1987-1988. The results showed the relative frequency of individual serotypes by locale and the yearly change in the prevalence of each serotype in the same area. The result of subgroup specificity of rotavirus obtained by using ELISA with subgroup I- and II-specific monoclonal antibodies confirmed the general finding that rotavirus strains having subgroup I specificity are serotype 2 and those having subgroup II specificity are either serotype 1, 3, or 4. Unusual strains having both subgroup I and II specificity or neither specificity and strains presumed to represent new serotypes were also found." (Authors' abstract)

500 Urasawa S, Urasawa T, Taniguchi K, Morita Y, Sakurada N, Saeki Y, Morita O, Hasegawa S. Validity of an enzyme-linked immunosorbent assay with serotype-specific monoclonal antibodies for serotyping human rotavirus in stool specimens. *Microbiol Immunol* 1988;32(7):699-708

"We recently developed a method for serotyping human rotavirus (HRV) by an enzyme-linked immunosorbent assay with HRV serotype-specific neutralizing monoclonal antibodies (ELISA serotyping). In the present study this method was compared with the fluorescent focus neutralization test with serotype-specific rabbit antisera (NT serotyping) in the sensitivity and specificity of the test. Direct serotyping of HRVs which were contained in stool specimens indicated that while only 37% of the samples were successfully serotyped in NT, 78% of the samples could be serotyped in ELISA. Regarding the samples whose serotype could be determined in the two tests, the assigned serotypes were identical in both tests. The results obtained indicated the utility of ELISA serotyping in clinical and epidemiologic studies of HRV infection." (Authors' abstract)

501 Urasawa T, Morita Y, Urasawa S, Taniguchi K. Use of microplate cell culture and enzyme immunoassay in titration of serum neutralizing antibody against Hock strain of serotype 4 human rotavirus. *Jpn J Med Sci Biol* 1986 Aug;39(4):163-7

"The tube neutralization test read by enzyme immunoassay developed by Wyatt et al. (1983) for serotype determination of human rotavirus was modified so as to use stationary cultures of MA104 cells in a microtiter plate instead of roller tube cultures. Sera obtained from different age groups were titrated for neutralizing antibody against serotype 4 human rotavirus Hock strain by this test and the results were compared with those obtained by the plaque neutralization test. There was a good correlation between the titers obtained by the two tests and the age distribution pattern of serotype 4 neutralizing antibody was similar to those of serotype 1 and 3 antibodies previously reported." (MEDLINE)

502 Urban RG, Pilpper EM, Dreyfus LA, Whipp SC. High-level production of *Escherichia coli* STb heat-stable enterotoxin and quantification by a direct enzyme-linked immunosorbent assay. *J Clin Microbiol* 1990 Nov;28(11):2383-8

"A convenient and sensitive enzyme-linked immunosorbent assay (ELISA) for the STb heat-stable enterotoxin of *Escherichia coli* was developed and used to quantify STb production by strains with a high level of expression. Based on an antigenic profile of the secreted form of STb, a synthetic peptide (STb3-27) spanning the major predicted epitope was synthesized, coupled to keyhole limpet hemocyanin, and used to immunize rabbits. Anti-STb3-27 antibodies were affinity purified on a synthetic peptide-sepharose 4B column and used in a direct-binding STb ELISA. Based on a highly purified form of toxin as a standard, the ELISA detected as little as 1 to 2 ng of STb from crude culture filtrates. ELISA data revealed that natural STb-producing strains elaborate little STb in defined-medium cultures relative to that elaborated by a recombinant strain harboring a cloned copy of the *estB* gene. Replacement of the endogenous STb promoter with any of several highly active promoters, including a bacteriophage T7 promoter, a β -galactosidase promoter, and a tryptophan- β -galactosidase hybrid (*tac*) promoter, increased the yield of STb 10- to 20-fold over levels obtained by an *E. coli* strain harboring the recombinant *estB* gene. The high level of STb antigen detected by the ELISA correlated with intestinal secretory activity. The combination of a convenient assay and effective hyperproduction of STb will serve as a basis for a large-scale toxin purification strategy." (Authors' abstract)

503 Ushijima H, Honma H, Ohnoda H, Mukoyama J, Oyanagi H, Araki K, Shinozaki T, Morikawa S, Kitamura T. Detection of anti-rotavirus IgG, IgM, and IgA antibodies in healthy subjects, rotavirus infections, and immunodeficiencies by immunoblotting. *J Med Virol* 1989 Jan;27(1):13-8

"Immune responses to individual simian rotavirus (SA 11) structural polypeptides were studied with emphasis on specific IgG, IgM, and IgA in paired sera from four children with rotavirus infections. Responses to simian rotavirus (SA 11) were also studied in 103 healthy children, 10 patients with primary immunodeficiency who received intravenous immunoglobulin, and 11 human immunodeficiency virus antibody-positive patients. All samples were immunoblotted for two major polypeptides--VP2 and VP6--of IgG. Immunoblotted IgA and IgM were elevated 2-4 weeks after the onset of the disease (stage II) in patients with rotavirus infection. Immunoblotted IgG remained almost the same at the onset of the disease and stage II. The scores for primary and secondary immunodeficiency patients were lower than for healthy children. The score obtained by totaling all polypeptides found in each individual was compared with the optical density (OD) value obtained by enzyme immunoassay." (Authors' abstract)

504 Ushijima H, Shinozaki T, Fujii A. Detection of *Clostridium difficile* enterotoxin in neonates by latex agglutination. *Arch Dis Child* 1985 Mar;60(3):252-71

Clostridium difficile enterotoxin (D-1) was detected in 37% (22 of 59; 13 symptomatic and 9 asymptomatic) of neonates in the intensive care unit by latex agglutination test. The presence of enterotoxin cannot explain why some asymptomatic neonates have *C. difficile* cytotoxin in stools. The enterotoxin was not found in 18 healthy control neonates in two other nurseries for newborns. Environmental contamination in the intensive care unit may have been the cause. Latex agglutination test was found sensitive to 15 mg/ml of enterotoxin. This can be useful in detecting diarrhoea.

505 Ushijima H, Kohno H, Kim B, Shinozaki T, Araki K, Fujii R. A new latex agglutination test kit for detecting rotavirus in stool from children with gastroenteritis. *Pediatr Infect Dis* 1986 Jul-Aug;5(4):492-3

506 Vadivelu J, Dunn DT, Feachem RG, Drasar BS, Cox NP, Harrison TJ, Lloyd BJ. Comparison of five assays for the heat-labile enterotoxin of *Escherichia coli*. *J Med Microbiol* 1987 May;23(3):221-6

"Enzyme-linked Immunosorbent assay (ELISA), DNA-DNA hybridisation, Vero cell assay, the Biken test and a new membrane-filter method were compared in the detection of heat-labile enterotoxin (LT) of *Escherichia coli*. Six subcultures of each of 50 strains of *E. coli* from the Biken collection were evaluated "blind" in the laboratory. The combined results of the most reproducible tests (ELISA and DNA-DNA hybridisation) were used to calculate the sensitivity and specificity of the other assays. The Vero-cell assay had a high sensitivity (98%) but a lower specificity (91%). The Biken and membrane-filter assays had sensitivities of 58-71% and 77-84% respectively, depending on the type of antiserum used. Only one false positive result was obtained with the Biken test; specificity of the membrane-filter assay was 94-95%. The membrane-filter assay, with anti-cholera toxin, is specific and reasonably sensitive. It has particular advantages over DNA-DNA hybridisation and the Biken test, and it may prove suitable for screening large numbers of *E. coli* isolates in epidemiological studies in developing countries." (Authors' abstract)

507 Vadivelu J, Parasakthi N, Puthucherry SD. Detection of labile enterotoxin of *E. coli* by the Biken assay and the GM₁-ELISA. Singapore Med J 1988 Feb;29(1):17-9

"One hundred and twelve *E. coli* strains from diarrhoeic children were tested for labile toxin production (LT) by the Biken assay kit (Institute Virion) and a GM₁-ELISA (monosialoganglioside GM₁). Ninety four of the 112 strains agreed on both assays -- 93 being classified as negative and one strain being classified as positive. A further 18 strains were identified as positive by the GM₁-ELISA. With a specificity of 97.5%, the GM₁-ELISA was more sensitive than the Biken assay kit. However, the GM₁-ELISA is not suitable for routine screening of *E. coli* isolates but an improved and more sensitive test similar to the Biken kit may be appropriate." (Authors' abstract)

508 van Nieuwstadt AP, Cornelissen JBJ, Zetstra T. Comparison of two methods for detection of transmissible gastroenteritis virus in feces of pigs with experimentally induced infection. Am J Vet Res 1988 Nov;49(11):1836-43

509 Van Poucke LSG. Salmonella-TEK, a rapid screening method for *Salmonella* species in food. Appl Environ Microbiol 1990 Apr;56(4):924-7

"A micro-enzyme-linked immunosorbent assay (micro-ELISA) using the Salmonella-TEK screen kit was tested for the detection of *Salmonella* spp. in pure cultures as well as in 30 artificially contaminated food samples and in 45 naturally contaminated food samples. Different raw, fleshy foods and processed foods were used as test products. The artificially contaminated minced meat samples were pre-enriched in buffered peptone water, and after incubation, different selective enrichment broths were tested. The micro-ELISA optical density values after enrichment and isolation of the different broths were very analogous. The quickest method to detect *Salmonella* spp. in different foods is to enrich them with *Salmosyst* broth, which reduces the total analysis time to 31 h. The Salmonella-TEK kit for *Salmonella* spp. provides a promising test for the detection of *Salmonella* antigens in food even when they are present at a low concentration (1 to 5 CFU/25 g). The cross-reaction of the anti-*Salmonella* antibodies, especially to other gram-negative bacteria, is nil." (Authors' abstract)

510 Varga FJ, DiPersio JR. Use of the RIM *Escherichia coli* Kit for rapid identification of *Escherichia coli*. Am J Clin Pathol 1986 Dec;86(6):761-4

"The Rapid Identification Method (RIM) *Escherichia coli* Kit (Austin Biologicals Laboratories Inc., Austin, TX) was evaluated as a means to rapidly differentiate at reduced cost clinical isolates of *Escherichia coli* from other members of the *Enterobacteriaceae*. A group of 408 gram-negative rods that were isolated on primary isolation plates were tested with the use of both the kit and complete bacterial identification. The RIM *E. coli* Kit identified 92.5% (272 of 294) of the *E. coli* isolates. Only 1 of 114 non-*E. coli* isolates was misidentified as *E. coli* (positive predictive value equal to 99.6). Although the RIM *E. coli*

Kit was sufficiently rapid (less than one hour), any supply cost savings would depend upon the proportion of true *E. coli* in the group of organisms tested. Savings derived from the lower cost of using the RIM *E. coli* Kit would be reduced by the extra cost of screening non-*E. coli* isolates, which would then have to be completely identified." (Authors' abstract)

511 Verbrugh HA, Mekkes DR, Verkooyen RP, Landheer JE. Widal-type serology using live antigen for diagnosis of *Shigella flexneri* dysentery. *Eur J Clin Microbiol* 1986 Oct;5(5):540-2

512 Vial PA, Kotloff KL, Tall BD, Morris JG, Jr., Levine MM. Detection of immune electron microscopy of 27-nm viral particles associated with community-acquired diarrhea in children. *J Infect Dis* 1990 Mar;161(3):571-3

513 Vinayak VK, Kum K, Chandna R, Venkateswarlu K, Mehta S. Detection of *Giardia lamblia* antigen in the feces by counterimmunoelectrophoresis. *Pediatr Infect Dis* 1985 Jul;4(4):383-6

The presence of *Giardia lamblia* antigen in stools of symptomatic and asymptomatic giardiasis patients was studied by counterimmunoelectrophoresis (CIE) using anti-*G. lamblia* (portland strain I) trophozoite antibody. Of the 20 giardiasis patients with gastrointestinal symptoms and diarrhea, 18 (90%) had detectable antigen by CIE. Seven patients with gastrointestinal symptoms, but without diarrhea, and with stool examinations positive for *G. lamblia* had antigen in the feces. Five of the asymptomatic patients with *G. lamblia* in the stool also had antigen in the feces. Thus among 32 patients, either symptomatic or asymptomatic, with stool examination positive for *G. lamblia*, 30 (94%) were positive for *G. lamblia* antigen. CIE failed to differentiate between symptomatic and asymptomatic *Giardia* infection. There were no false-positive in children with other diseases. Antigen in the stools disappeared within 1 to 2 weeks after eradication of infection with specific treatment. The sensitivity of CIE test in detecting *G. lamblia* antigen was 94%, and the specificity 95%. The diagnostic accuracy in assessing the patients were 93.6%. Since the test is rapid, easy to perform and capable of standardization, its use as a diagnostic tool for giardiasis was recommended.

514 Vinayak VK, Shandil RK, Bansal V, Singh K, Bhasin DK, Kaur U. Uses and limitations in the demonstration of specific circulating immune complexes in patients with amoebiasis. *J Med Microbiol* 1990 Jun;32(2):87-91

"A micro-enzyme linked immunosorbent assay (micro-ELISA) has been evaluated as a diagnostic test to detect amoebic antigen in polyethylene glycol (PEG) precipitated circulating immune complexes (CIC) in sera from patients with amoebiasis. The immune complexes were captured on rabbit anti-amoebic IgG-coated wells of microtitration plates and the complexed antigen was detected by enzyme linked antihuman immunoglobulins. A titre of >160 for the immune complexes was considered to be of clinical significance. The immunoassay detected amoebic, antigen-specific CIC in 35 (94.5%) of 37 patients with confirmed amoebic liver abscess. Twenty (55.5%) of 36 clinically suspected cases of amoebic liver abscess had amoebic antigen-specific CIC and responded favourably to anti-amoebic chemotherapy. Only two (20%) of 10 cases of non-dysenteric symptomatic intestinal amoebic infection had amoebic antigen-specific CIC. One (10%) of 10 patients with non-amoebic intestinal disorders also had amoebic antigen in CIC. However, none of 15 cases of non-amoebic hepatic disorders that included hydatid disease, metastatic adenocarcinoma, hepatocellular carcinoma, cholecystitis and choledocal cyst, 13 cases of rheumatoid arthritis and 25 apparently healthy subjects had amoebic antigen in CIC. The levels of the amoebic antigen-specific CIC did not correlate ($p>0.05$) with either the number of abscess(es) or lobe(s) of the liver involved. However, the levels of antigen-specific CIC were higher ($p<0.01$) in patients with a liver size of more than 5 cm below the right costal margin. Antigen-specific CIC levels tended to decline or disappear during 3-6 months following

completion of therapy. In spite of the limitations for diagnosing symptomatic intestinal amoebic disease, the demonstration of specific CIC is recommended as an immuno-diagnostic procedure for patients with suspected amoebic liver diseases." (Authors' abstract)

515 von Wulffen H, Grote HJ. Enzyme-linked immunosorbent assay for detection of Immunoglobulin A and G antibodies to *Campylobacter pylori*. Eur J Clin Microbiol Infect Dis 1988 Aug;7(4):559-65

516 Vonderfecht SL, Miskuff RL, Eiden JJ, Yolken RH. Enzyme immunoassay inhibition assay for the detection of rat rotavirus-like agent in intestinal and fecal specimens obtained from diarrheic rats and humans. J Clin Microbiol 1985 Nov;22(5):726-30

"An enzyme immunoassay inhibition assay was developed to detect rat rotavirus-like agent (RVLA) or antigenically related viruses in intestinal washings and homogenates obtained from diarrheic rats and in fecal specimens obtained from diarrheic humans. In this assay, RVLA antigens in a test sample inhibited the binding of a biotin-labeled anti-rat RVLA antibody preparation to rat RVLA antigens bound to the solid phase. Intestinal washings and homogenates obtained 1 day after RVLA infection of suckling rats inhibited the binding of the biotin-labeled antibody to the solid-phase rat RVLA antigens by 76 to 100%. The inhibition was blocked by RVLA immune rat serum but not by nonimmune rat serum. Of 27 fecal specimens obtained from diarrheic humans, 6 produced disease characteristic of rat RVLA infection when inoculated into suckling rats. Four of these six specimens produced greater than 50% inhibition in the enzyme immunoassay. Fecal specimens obtained from diarrheic humans that were determined to be negative for RVLA produced an average inhibition of 9.2%. This enzyme immunoassay appears to be a useful diagnostic and research tool for the study of infections with at least one of the antigenically distinct rotaviruses." (Authors' abstract)

517 Voronov AV, Malov VA, Pak SG, Tur'ianov MKh, Gurskii IuN, Serebriakov Mlu, Gramotkina EA, Bengerov Iula. [An immunoenzyme method for determining the O-antigen of Sonne's *Shigella* with the use of affinity isolated antibodies]. Lab Delo 1989;(9):66-70

"The sensitivities of ELISA systems for assays of *Shigella sonnei* O-antigen, developed on the basis of specific rabbit antibodies isolated from antisera by different chromatography techniques have been compared. The highest specificity and sensitivity of O-antigen determination (0.5-1 ng/ml) is achieved when using affinity-isolated antibodies to O-antigen. Clinical trials of this ELISA system have been carried out." (MEDLINE)

518 Wahlquist SP, Eberhard ML. Sodium azide: ineffective as a faecal preservative for parasitological diagnosis. Annl Trop Med Parasitol 1991 Jun;85(3):365-8

"Sodium azide was compared with 10% formalin to evaluate sodium azide's effectiveness as a faecal preservative for intestinal helminths and protozoa. Faecal specimens collected from Haiti were preserved in sodium azide and in 10% formalin and analysed after 1.5, 6.5 and 11.5 weeks by examining direct wet-mount preparations. Sodium azide did not preserve the morphology of either helminths or protozoa as well as 10% formalin did. However, sodium azide prevented embryogenesis of helminth eggs, while some helminth eggs in 10% formalin contained living larvae. Biosafety guidelines regarding the toxicity, reactivity, and disposal of sodium azide were strictly followed. Use of 10% formalin is a significantly better choice than sodium azide for preserving parasites when accurate identification of parasites and biosafety are the main concerns." (Authors' abstract)

519 Walker RC, Ruane PJ, Rosenblatt JE, Lyerly DM, Gleaves CA, Smith TF, Pierce

PF, Jr., Wilkins TD. Comparison of culture, cytotoxicity assays, and enzyme-linked immunosorbent assay for toxin A and toxin B in the diagnosis of *Clostridium difficile*-related enteric disease. *Diagn Microbiol Infect Dis* 1986 May;5(1):61-9

"*Clostridium difficile* culture, test tube, and microtiter cytotoxicity assays, and enzyme-linked immunosorbent assays (ELISAs) for toxin A and toxin B, were simultaneously performed on 113 fresh diarrheal stool specimens randomly selected from those submitted to our clinical laboratory for routine *C. difficile* testing. The performance of these tests in diagnosing *C. difficile*-related enteric disease (CDRED) was based on a clinical assessment of the likelihood of CDRED as determined by a systematic review of case histories blinded from the test results. Among 61 antibiotic recipients, both the microtiter cytotoxicity assay and the toxin A ELISA were highly specific for CDRED (95% and 100%, respectively). Specificities for the other procedures were much lower (tube cytotoxicity assay, 79%; culture, 74%; and toxin B ELISA, 56%). The high sensitivities of the culture (89%) and toxin B ELISA (83%) were somewhat negated by their low specificities. The only test that was both specific and had acceptable sensitivity (78%) was the microtiter cytotoxicity assay. This study indicates that ELISAs for detection of *C. difficile* toxins are not as reliable as the cytotoxicity assay in the laboratory diagnosis of CDRED, and that clinical correlation is essential in the evaluation of any new test for CDRED." (Authors' abstract)

520 Walmsley SL, Karmali MA. Direct isolation of atypical thermophilic *Campylobacter* species from human feces on selective agar medium. *J Clin Microbiol* 1989 Apr;27(4):668-70

521 Weber R, Bryan RT, Bishop HS, Wahlquist SP, Sullivan JJ, Juranek DD. Threshold of detection of *Cryptosporidium* oocysts in human stool specimens: evidence for low sensitivity of current diagnostic methods. *J Clin Microbiol* 1991 Jul;29(7):1323-7

"To determine the minimum number of *Cryptosporidium* oocysts that can be detected in stool specimens by diagnostic procedures, stool samples seeded with known numbers of *Cryptosporidium parvum* oocysts were processed by the modified Formalin-ethyl acetate (FEA) stool concentration method. FEA concentrates were subsequently examined by both the modified cold Kinyoun acid-fast (AF) staining and fluorescein-tagged monoclonal antibody (Immunofluorescence [IF]) techniques. Oocysts were more easily detected in watery diarrheal stool specimens than they were in formed stool specimens. For watery stool specimens, a 100% detection rate was accomplished at a concentration of 10,000 oocysts per g of stool by both the AF staining and IF techniques. In formed stool specimens, 100% of specimens seeded with 50,000 oocysts per gram of stool were detected by the IF technique, whereas 500,000 oocysts per g of stool were needed for a 100% detection rate by AF staining. Counting of all oocysts on IF slides indicated a mean oocyst loss ranging from 51.2 to 99.6%, depending on the stool consistency, as determined by the FEA concentration procedure. Our findings suggest that the most commonly used coprodiagnostic techniques may fail to detect cryptosporidiosis in many immunocompromised and immunocompetent individuals." (Authors' abstract)

522 Westenbrink F, Middel WG, Straver PJ, de Leeuw PW. A blocking enzyme-linked immunosorbent assay (ELISA) for bovine virus diarrhoea virus serology. *Zentralbl Veterinarmed [B]* 1986 Jun;33(5):354-61

523 Westmoreland D, Ashley CR, Caul EO. Rapid and reliable routine diagnosis of rotavirus using a commercial monoclonal antibody based immunoassay. *J Clin Pathol* 1987 Nov;40(11):1384-6

524 Wlonecka J, Olding-Stenkvis E, Schroder H, Hultdt G. Detection of *Giardia* antigen in stool samples by a semi-quantitative enzyme immunoassay (EIA) test. *Scand J Infect Dis* 1989;21(4):443-8

"A semi-quantitative enzyme immunoassay (EIA) test for the detection of *Giardia* intestinalis antigen in faeces was developed. In order to avoid unspecific reactions due to anticalf serum activity, IgG fractions of anti-*giardia* rabbit and sheep sera were purified from antbovine antibodies by immunoadsorption. Faecal specimens tested in the assay were mixed with normal horse serum to avoid unspecific and proteolytic effects of stool components. Out of a range of bacterial and parasitic antigens tested, only high concentrations of *Ascaris suum* egg antigen and *Shigella sonnei* gave slight unspecific reactions. During an outbreak of waterborne giardia infection faecal samples from 49 individuals were tested. *Giardia* cysts were found by microscopy in 22 individuals and specimens from 24 persons were positive by EIA. The estimated amount of antigen found in the positive samples ranged from 50 to 5,000 ng giardia protein/ml. After filtration through a 0.45 µm filter antigenic activity was found in the filtrate of 10 samples. In 3/6 samples with no cysts detected by microscopy, mainly soluble giardia antigen was found." (Authors' abstract)

525 Wilde J, Yolken R, Willoughby R, Eiden J. Improved detection of rotavirus shedding by polymerase chain reaction. *Lancet* 1991 Feb 9;337(8737):323-6

"To improve identification of children excreting rotavirus a method for the amplification of rotavirus RNA by the polymerase chain reaction (PCR) was developed. The assay was compared with a solid-phase enzyme immunoassay in the detection of rotavirus shedding by infants in hospital during the winter peak of rotavirus infections. Forty children were studied in an intermediate care unit after transfer from intensive care units. Only two were admitted primarily because of diarrhoea; the other thirty-eight were admitted for management of various other disorders. Rotavirus shedding was detected by enzyme immunoassay in twenty of the infants, and nine of these (aged 1 week to 8 months) remained in hospital for more than 5 days after the initial detection of rotavirus and could be studied long term. Of 103 faecal samples from the nine infants, 60 (58%) contained rotavirus RNA detected by reverse-transcriptase (RT)/PCR, whereas only 37 (36%) were positive for rotavirus antigen by the immunoassay ($X^2=10.3$, $p<0.002$). The geometric mean time of rotavirus shedding was 9.5 (range 1-19) days as detected by RT/PCR and 5.7 (range 1-17) days by the immunoassay ($p<0.018$). In five of the nine children, RT/PCR detected rotavirus shedding for 2-7 days longer than the immunoassay and in four children RT/PCR was positive 1 or more days before rotavirus antigen was detected. Further studies should attempt to find out whether infected infants are capable of spreading wild-type virus during periods when they are not shedding antigen as detectable by enzyme immunoassay." (Authors' abstract)

526 Wilde J, Eiden J, Yolken R. Removal of inhibitory substances from human fecal specimens for detection of group A rotaviruses by reverse transcriptase and polymerase chain reactions. *J Clin Microbiol* 1990 Jun;28(6):1300-7

"A method was developed for the purification of rotavirus RNA from fecal extracts in order to permit the sensitive identification of group A rotavirus in fecal specimens by the polymerase chain reaction. Sequential reactions with reverse transcriptase and *Taq* polymerase with directed primers from rotavirus gene 6 yielded characteristic 259-base-pair fragments that were then visualized by silver stain on a polyacrylamide gel. As few as 500 genomic copies of purified rotavirus RNA could be detected in this manner. However, when the method was applied to fecal samples with added rotavirus virions, inhibition was noted in many of the fecal extracts which were tested. The inhibition could be reversed by dilution of the fecal extract, but sensitivity was also reduced by a corresponding dilutional factor. The inhibition was quantitatively removed by an added step in the extraction process that utilized chromatographic cellulose fiber powder (CF11 powder) to purify the rotavirus RNA during a series of rapid washing and elution steps. After CF11 purification, rotavirus RNA could be detected in experimental fecal samples at dilutions 1,000- to 10,000-fold beyond the detection limits of standard techniques such as enzyme immunoassay and the direct visualization of RNA following polyacrylamide gel electrophoresis. Furthermore, following purification by CF11, rotavirus RNA could be

detected in all of seven enzyme-linked immunosorbent assay-positive fecal samples obtained from a child with rotavirus gastroenteritis; when CF11 purification was not performed, rotavirus RNA could be detected in only four of these samples, even after the removal of inhibitors by dilution of the extracts. Large-scale identification of rotavirus in fecal specimens may be possible by use of CF11 purification of viral RNA prior to sequential reaction with reverse transcriptase and *Taq* polymerase in a modified polymerase chain reaction." (Authors' abstract)

527 Wimsatt JC, Harmon SM, Shah DB. Detection of *Clostridium perfringens* enterotoxin in stool specimens and culture supernatants by enzyme-linked immunosorbent assay. *Diagn Microbiol Infect Dis* 1986 Apr;4(4):307-13

"An enzyme-linked immunosorbent assay was developed to detect and quantitate *Clostridium perfringens* enterotoxin A in culture supernatants and in stool specimens from cases of diarrhea in which high numbers of enterotoxin-producing *Clostridium perfringens* were isolated. To analyze for enterotoxin A, polyvinyl chloride microtiter plates were coated with dilute immune whole rabbit serum. Enterotoxin A standards and samples were allowed to react with sensitized wells. The presence of the immobilized antigen in the wells was detected by the binding of immune rabbit immunoglobulin conjugated with peroxidase. Nanograms of enterotoxin were detectable. Four enterotoxin-positive and seven enterotoxin-negative cultures grown in Duncan-Strong medium gave expected results. Eighteen of 23 diarrheal stool specimens obtained after a food-poisoning outbreak at a state hospital were found to contain microgram quantities of enterotoxin per gram of stool, whereas five control diarrheal specimens contained <0.6 ng enterotoxin per gram of stool. These results indicate that the enzyme-linked immunosorbent assay technique is useful for differentiating enterotoxigenic strains and for diagnosing diarrhea caused by enterotoxigenic *Clostridium perfringens*." (Authors' abstract)

528 Wood PK, Morris JG Jr., Small PLC, Sethabutr O, Toledo MRF, Trabulsi L, Kaper JB. Comparison of DNA probes and the Sereny test for identification of invasive *Shigella* and *Escherichia coli* strains. *J Clin Microbiol* 1986 Sep;24(3):498-500

"Forty-two *Shigella* and 29 *Escherichia coli* strains were screened for invasiveness in the Sereny test and for hybridization with two recently described DNA probes for the invasiveness plasmid. Both probes produced identical results. All Sereny-positive strains hybridized with both DNA probes. Three Sereny-negative strains also hybridized with the probes, suggesting that there are strains containing the invasiveness plasmid that are not pathogenic in animal models." (Authors' abstract)

529 Woods GL, Iwen PC. Comparison of a dot immunobinding assay, latex agglutination, and cytotoxin assay for laboratory diagnosis of *Clostridium difficile*-associated diarrhea. *J Clin Microbiol* 1990 May;28(5):855-7

530 Wren B, Clayton C, Tabaqchali S. Rapid identification of toxigenic *Clostridium difficile* by polymerase chain reaction [letter]. *Lancet* 1990 Feb 17;335(8686):423

531 Wren BW, Tabaqchali S. Detection of pathogenic *Yersinia enterocolitica* by the polymerase chain reaction [letter]. *Lancet* 1990 Sep 15;336(8716):693

532 Wren BW, Clayton CL, Castledine NB, Tabaqchali S. Identification of toxigenic *Clostridium difficile* strains by using a toxin A gene-specific probe. *J Clin Microbiol* 1990 Aug;28(8):1808-12

533 Wu B, Mahony JB, Simon G, Chernesky MA. Sensitive solid-phase immune electron microscopy double-antibody technique with gold-immunoglobulin G complexes for detecting rotavirus in cell culture and feces. *J Clin Microbiol* 1990 May;28(5):864-8

"A new solid-phase immune electron microscopy double-antibody colloidal-gold

technique (SPIEMDAGT) was developed and compared with direct electron microscopy, direct immune electron microscopy, and enzyme immunoassay for detecting rotavirus. Guinea pig and rabbit antirotavirus antisera were used as capture and detector antibodies, respectively, and goat anti-rabbit immunoglobulin G-gold complexes were employed as a label. Animal rotavirus in cell culture media and human virus in stool specimens were detected by this method. On average, SPIEMDAGT detected 800 times more virus particles than direct electron microscopy and 45 times more particles than direct immune electron microscopy and yielded 20% more positives than enzyme immunoassay. SPIEMDAGT could detect not only viral antigen associated with morphologically recognizable particles but also antigen present when whole virus particles were not visible." (Authors' abstract)

534 Wu HL. [A study on the rotavirus antibody in the normal human sera using ELISA]. *Chung Hua Liu Hsing Ping Hsueh Tsa Chih* 1986 Aug;7(4):203-5

535 Yadav SK, Jain AK, Srivastava VK, Gupta JP. Comparison of stool microscopy and serology (enzyme linked immunosorbent assay) in epidemiology of amebiasis. *Indian J Gastroenterol* 1990 Jan;9(1):25-6

"Stools from 634 individuals from Varanasi were examined for *Entamoeba histolytica* (EH). Serology was done in these subjects by enzyme linked immunosorbent assay (ELISA) employing filter paper technique. Stools were positive for EH in 16.9%, and serology in 15.9%. Both the tests were positive in only 5.2%. In 72.4% both the tests were negative. In 11.7% of stool positive cases, serology was negative, and in 10.7% with positive serology stool examination did not reveal EH. A majority (92.5%) of stool positive subjects had only cysts. Additional parasites were detected in 15.3%." (Authors' abstract)

536 Yam WC, Lung ML, Ng MH. Evaluation and optimization of the DNA filter assay for direct detection of enterotoxigenic *Escherichia coli* in the presence of stool coliforms. *J Clin Microbiol* 1986 Jul;24(1):149-51

The DNA filter assay was compared with conventional tests for direct detection of enterotoxigenic *Escherichia coli* (ETEC) in mixed cultures containing non-toxigenic *E. coli* or stool coliforms. The DNA filter assay was clearly more sensitive and less influenced by strain-to-strain variation. Direct detection of ETEC in mixed cultures by DNA filter assay was affected by the presence of other bacteria in the cultures. By shortening incubation to 6 hours, bacterial overgrowth was minimised, allowing direct detection of ETEC in stools or mixed cultures initially containing 0.2 to 3% ETEC. The 6-h DNA filter assay was further evaluated with stool specimens from 136 diarrhoeal patients and 245 patients with other diseases. Four of the diarrhoeal patients, but none of the other patients, produced positive results. The findings suggest that the modified DNA filter assay affords a convenient and reliable method for clinical and epidemiological studies of ETEC-related diarrhoeas.

537 Yamakawa K, Oyamada H, Nakagomi O. Identification of rotaviruses by dot-blot hybridization using an alkaline phosphatase-conjugated synthetic oligonucleotide probe. *Mol Cell Probes* 1989 Dec;3(4):397-401

"We have evaluated a recently-developed dot-blot hybridization assay for the detection of human rotaviruses using an alkaline-phosphatase conjugated oligonucleotide probe. The lower detection limit of this assay was 1 ng (approximately 5×10^7) copies of the double-stranded (ds) RNA, when a purified preparation from serotype 1 human rotavirus was used but appeared to be much higher when applied on clinical specimens. This assay could detect dsRNA from rotavirus strains belonging to serotypes 1 through 6, 8 and 9. A total of 235 stool specimens were used to evaluate the oligonucleotide probe assay in comparison with polyacrylamide gel electrophoresis and latex agglutination assay (Rotalex). When polyacrylamide gel electrophoresis was used as a gold standard, sensitivity, specificity and positive and negative predictive values of the probe assay were

84, 100, 100 and 89%, respectively. These values were slightly better than those of Rotalex assay which is commonly used in clinical laboratories in Japan. Although the probe assay requires more hands-on time than the immunoassays, the high specificity of this probe assay recommends it as a confirmatory test in the clinical laboratory setting." (MEDLINE)

538 Yolken RH, Ungar B. ELISA detection of *Giardia lamblia* [letter]. Lancet 1985 Nov 16;2(8464):1120

539 Yolken RH, Leggiadro RJ. Immunoassays for the diagnosis of viral enteric pathogens. Diagn Microbiol Infect Dis 1986 Mar;4(3 suppl):S61-S69

"The accurate diagnosis of infectious diseases is important for the optimal management of infected patients as well as for the prevention of disease transmission to susceptible individuals. Because viral gastroenteritis constitutes an important cause of morbidity in children living in developed countries and of mortality in children living in developing countries, there has been a great deal of interest in the development of effective methods for the diagnosis and study of this disease. While there are a number of assay systems that are capable of accurate detection of the agents of viral gastroenteritis in the intestinal contents of infected individuals, solid phase enzyme immunoassays have been used widely for this purpose. The widespread utilization of enzyme immunoassays is based on the advantages inherent in the use of enzymatic markers. These advantages include the stability of enzyme-immunoglobulin conjugates, the high degree of sensitivity inherent in the magnifying nature of enzyme-substrate interactions, and the low cost of assays performed in simple reaction formats. Enzyme immunoassays have thus been developed and widely used for the detection and epidemiologic study of rotaviruses, adenoviruses, coxsackie viruses, hepatitis A virus, and other agents that replicate in the human gastrointestinal tract. In addition, latex agglutination assays and nucleic acid hybridization techniques have been applied to the rapid detection of enteric viruses. It is likely that the application of a number of assay systems will promote accurate identification of a wide range of viral pathogens under different clinical, epidemiologic, and laboratory situations." (MEDLINE)

540 Yolken RH, Miotti P, Viscidi R. Immunoassays for the diagnosis and study of viral gastroenteritis. Pediatr Infect Dis 1986 Jan-Feb;5(1 suppl):S46-S52

541 Yolken RH. New methods for the diagnosis of enteric infections. World J Microbiol Biotechnol 1991 Mar;7(2):150-6

"Gastrointestinal disease remains a major cause of mortality and morbidity throughout the world. Recently, a number of viral, bacterial, and protozoan agents have been identified which can cause a range of gastrointestinal disorders. The effective management of these diseases requires the prompt identification of the infecting microorganism and the early institution of preventive and therapeutic interventions. The detection of infecting microorganisms in fecal and intestinal fluids presents a particular challenge to the diagnostic microbiologist. Cultivation can be difficult due to the fastidious nature of the microorganisms and the presence of cytotoxic materials in the specimen. In the past, immunoassays have been used for the detection of some microorganisms. However, immunoassays have limited sensitivity and cannot detect all infecting microorganisms. Recently, nucleic acid amplification techniques have been developed for the direct detection of pathogenic microbial DNA and RNA in human body fluids. We have found that these methods can be applied for the accurate detection of intestinal viruses provided that inhibitors of enzymatic amplification are removed from the sample. Using affinity binding purification and non-isotopic DNA measurement techniques, we have developed sensitive and specific assays for the quantitation of a wide range of infecting microorganisms in intestinal fluids. Nucleic acid amplification provides a unique tool for the study of enteric pathogens and for the development of strategies for their eventual elimination from the human environment." (Author's abstract)

542 Yolken RH, Eiden J, Leister F. Self-contained enzymic membrane immunoassay for detection of rotavirus antigen in clinical samples. *Lancet* 1986 Dec 6;2(8519):1305-7

"A self-contained enzymic membrane immunoassay (SCEMIA) system has been developed for the detection of viral antigens in clinical samples. The assay system makes use of antiviral antibodies bound to a nylon membrane, a flow-through washing procedure, and a clearly visible endpoint of the enzymic reaction. A SCEMIA system with antibodies against rotavirus detected rotavirus antigen, within 15 min, in all faecal samples from children with gastroenteritis that were positive for antigen in a standard microplate enzyme immunoassay, which took 4 h to complete. In addition, the SCEMIA could detect rotavirus in faecal samples collected from infected individuals both before and after antigen could be detected by a standard immunoassay system. Rotavirus antigen was not detectable in control children who did not have evidence of rotavirus infection. SCEMIA systems are an accurate, rapid, and inexpensive means for the practical diagnosis of viral infections in human beings." (Authors' abstract)

543 Young DM, Biao J, Zheng Z, Hadler J, Edberg SC. Isolation of *Campylobacter jejuni* in Hunan, The People's Republic of China: epidemiology and comparison of Chinese and American methodology. *Diagn Microbiol Infect Dis* 1986 Jul;5(2):143-9

"The incidence of infection with *Campylobacter jejuni* was determined in individuals with and without diarrheal disease seen at a hospital in Changsha, China. Stool specimens were cultured by two methods: one developed by the Chinese and a second control method used commonly in the United States. Among people with diarrhea, 18.7% of the 48 children and 8.7% of 104 adults were infected with *Campylobacter jejuni*. In the group without diarrhea, 8.6% of 105 children but none of 76 adults tested had the organism in their stool. Of the 27 total positive cultures, the Chinese method was positive in 26, compared with 21 for the control method. The Chinese culture method was at least as sensitive and specific as the American method. The biochemical characteristics and antibiotic sensitivities of Chinese isolates were similar to organisms isolated in other parts of the world. This study shows that *Campylobacter jejuni* is a common enteric pathogen in China and that the asymptomatic carrier state of this organism is significant in children." (Authors' abstract)

544 Zavate O, Avram G, Ivan A, Haimovici M, Felea D. [Frequency of the rotavirus antigen, detected by the ELISA method, in the feces of patients with infantile enteritis]. *Rev Med Chir Soc Med Nat Iasi* 1986 Jan-Mar;90(1):77-8

545 Zentner BS, Margalith M, Galil A, Halevy B, Sarov I. Detection of rotavirus-specific IgG antibodies by immunoperoxidase assay and enzyme-linked immunosorbent assay. *J Virol Methods* 1985 Jul;11(3):199-206

"An indirect immunoperoxidase assay (IPA) has been developed for determination of IgG antibodies to rotavirus. The technique employed as antigen, SA-11 infected MA 104 cells, which were air-dried on glass slides and acetone-fixed. In parallel, rota-specific IgG antibodies were determined by enzyme-linked immunosorbent assay (ELISA). Specific IgG antibodies to rotavirus were determined in sera of healthy children and in sera of patients suffering from gastroenteritis. A good correlation ($r = 0.92$) and ($r = 0.98$) for healthy children and patients, respectively, was found between IPA and ELISA techniques. The IPA technique is rapid and simple and positive results, because of the intensive staining, are easily read by low-power light microscope. The potential application of IPA and ELISA methods in serodiagnosis of rotavirus infections is discussed." (MEDLINE)

546 Zenyov H. [Application of immunologic methods to the rapid diagnosis of bacterial infections]. *Rinsho Byori* 1987 Nov;35(11):1216-20

547 Zikmundova L, Hejna M, Sramkova L, Korych B, Kucharska Z. [Possibilities of detecting rotavirus diseases in clinical practice. Comparison of the ELISA, CIEP and latex agglutination methods for the detection of viruses by the determination of an increase in antibodies]. *Cesk Epidemiol Mikrobiol Imunol* 1986 Jul;35(4):217-22

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