

**MONSURS MEDIUM AND DEXTRIN HEART INFUSION AGAR AS ROUTINE
MEDIA FOR THE LABORATORY DIAGNOSIS OF CHOLERA**

Md. Imdadul Huq
Dr. James McLaughlin
Dr. W. F. Verwey

Over the past decade cholera has continued to spread into areas of the world where laboratorians are unfamiliar with the isolation and identification of V. cholerae. For this reason, and because non-cholera diarrheas can give a clinical picture similar to cholera, media giving rapid presumptive identification of V. cholerae are important.

During the last years considerable progress has been made in improving the sensitivity of the techniques of the isolation of Vibrio cholerae. As many as 92% to 96% of clinical cases may be confirmed by presently available bacteriological procedures. A large number of both plating and enrichment media has been described by various workers for the isolation of V. cholerae and are used by different laboratories largely on the basis of personal preferences and individual experience. Cholera Research Laboratory has had the opportunity to evaluate many of them.

A selective medium, TTGA (Taurocholate Tellurite Gelatin Agar, Monsur) and a non selective medium, Gelatin Agar (Goodner, Smith) has been used routinely in the Cholera Research Laboratory. Recently Thiosulphate citrate Bile Salt Sucrose Agar, TCBS, because it is commercially available in dehydrated form, does not require autoclaving and gives a characteristic V. cholerae colony, has been widely recommended for the isolation of V. cholerae. A new non-selective differential medium, DHI (Dextrin Heart Infusion Agar, Schrank, 1973) has recently been described. Because of the large number of cholera cases occurring in the CRL Hospital, the Bacteriological Laboratory has had an excellent opportunity to evaluate and compare these media for diagnostic usefulness.

Materials and Methods

Rectal swabs or stool were collected from patients before therapy was started. Two swabs were taken, one streaked onto Gelatin Agar and DHI and the other onto TTGA and TCBS. All plates were incubated overnight at 35°C and then read. Suspected V. cholerae colonies were confirmed by agglutination with O group I and specific antisera.

The non-selective medium DHI used in this report contains dextrin, beef extract, gelatin, heart infusion broth and neutral red. (Slide I). Vibrio cholerae give a characteristic reddish colony due to dextrin utilization and the uptake of neutral red and are surrounded by a halo demonstrating gelatinase activity.

Originally both BBL and Eiken TCBS was used, however, the BBL brand was unsatisfactory and those results are not in this report.

Results

7631 rectal swab or stool samples were plated on combinations of three or all four of the media, 5070 positive isolations of V. cholerae were made on at least one of the media. Slide 2 shows the results from a series using all four of the media (Gelatin Agar, Dextrin Heart Infusion Agar, Taurocholate Tellurite Gelatin Agar and TCBS) on 1425 cultures. In this series TTGA detected 611 positives and was positive whenever any of the other culture media showed cholera organisms. 2.1% of these were missed on TCBS, 2.9% on Gelatin Agar and 3.3% on Dextrin Heart Infusion Agar. In a larger series, where TCBS was not included, 3326 positive isolations were made, Gelatin Agar or DHI failed to detect 261 (5.9%) of these V. cholerae positives.

In order to directly evaluate the effectiveness of DHI versus Gelatin Agar another series was run using either TCBS or TTGA as the selective medium and DHI and Gelatin Agar as the non-selective medium. 1744 positive isolations were made in which V. cholerae was missed 17 times on DHI and 14 times on Gelatin Agar.

Discussion

The CRL bacteriological laboratory has been using TTGA and Gelatine Agar as routine V. cholerae isolation media. In a comparative study of media in 1963, Finkelstein and Gomes found Gelatin Agar least effective and suggested Meat Extract Agar as an alternative. However in our laboratory Meat Extract Agar was found to be unsatisfactory. We have now tested Dextrin Heart Infusion Agar as an alternative to Gelatin Agar in routine diagnostic work. The isolation rate of V. cholerae was shown to be almost identical on these two media. Though Dextrin Heart Infusion Agar is a bit more costly than Gelatin Agar, it gives very characteristic colonies and in those circumstances where a non-selective medium is used, Dextrin Heart Infusion Agar is superior to Gelatin Agar because of the ease of colony recognition.

It has been described recently and confirmed in CRL that some V. cholerae strains fails to grow on T.C.B.S. Therefore in the central laboratories where skilled technical staff and sterilising facilities are available TTGA is the medium of choice. However under field conditions where minimal facilities and skill may be available, TCBS would probably be preferred because of its ease of preparation and the highly characteristic appearance of V. cholerae colonies.

PROCEEDINGS OF THE 9TH MEETING OF THE SCIENTIFIC
REVIEW AND TECHNICAL ADVISORY COMMITTEE OF
THE CHOLERA RESEARCH LABORATORY

①

and

REPORTS OF THE COLLABORATIVE STUDIES BETWEEN CENTER
FOR MEDICAL RESEARCH AND CHOLERA RESEARCH
LABORATORY

For the
YEAR 1974

Dacca, Bangladesh