NPK FERTILIZERS MODULATES THE GROWTH, YIELD AND BIOCHEMICAL PARAMETERS OF BUCKWHEAT PLANTS

A THESIS
BY

MD. NAIMUR RAHMAN

Registration No.: 2205142
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MASTER OF SCIENCE (MS)
IN
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DEPARTMENT OF BIOCHEMISTRY AND MOLECULAR BIOLOGY

HAJEE MOHAMMAD DANESH SCIENCE AND TECHNOLOGY UNIVERSITY, DINAJPUR-5200

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Submitted to the Department of Biochemistry and Molecular Biology Hajee Mohammad Danesh Science and Technology University, Dinajpur in partial fulfillment of the requirement for the degree of

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DECEMBER, 2023

DEDICATED

TO MY

BELOVED PARENTS AND MY UNCLE

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ABSTRACT

This study was carried out to determine the effect of different doses of NPK fertilizers on plant growth, yield and antioxidant enzyme activities of common buckwheat genotype. The LR18 landraces of buckwheat were collected from buckwheat germplasm center of biochemistry and molecular biology department of Hajee Mohammad Danesh Science and Technology University, Dinajpur, Bangladesh. These seeds were sown in the field following the RCBD design. We used different doses of NPK fertilizers (T_0: control, T_1: urea = 36g/8m^2, TSP = 52g/8m^2, MOP = 12g/8m^2, T_2: urea = 44g/8m^2, TSP = 60g/8m^2, MOP = 20g/8m^2, T_3: urea = 52g/8m^2, TSP = 68g/8m^2, MOP = 28g/8m^2, T_4: urea = 60g/8m^2, TSP = 76g/8m^2, MOP = 36g/8m^2) in soils, these fertilizers were used in 20 days interval. The morphological data were recorded and analyzed from one landrace. Among the morphological data, plant height, distant of first node, number of inflorescence, per plant, raceme length, number of branches,1000 grain weight, and grain weight per plot showed the statistical significance. Our study showed that flowering was started about 28 days after sowing (DAS) and 100% germination was recorded 34 DAS. We randomly collected leaves from seedling stages of buckwheat plants for antioxidant enzyme activity analysis. Our study found the significant differences between plant height, raceme length, grain weight per plot and seed weight per plant. The highest plant height was observed in T_4 with mean and standard deviation was 39.77 ± 3.09and these plants showed significant variations control plants and among all treatments. On the other hand, the highest seed weight per plot was observed in T_4 with a mean and standard deviation was 483.33 ± 18.24.The antioxidant enzyme activity showed that moderate differences between four treatments. The lowest catalase activity was observed in T_4 with a mean and standard deviation was 0.72 ± 0.03. The highest peroxidase activity was measured in T_4 with a mean and standard deviation was 8.20 ± 5.50. On the other hand, the highest ascorbate activity was calculated in T_4 with a mean and standard deviation was 4.26 ± 0.82. But the highest hydrogen peroxide activity was measured in T_2 with a mean and standard deviation was 1.74 ± 0.07. The highest MDA was measured in $T_0$with a mean and standard deviation was 3.57 ± 0.10But the lowest MDA was measured in T_4 with a mean and standard deviation was 0.52 ± 0.02. The highest proline was measured in $T_4$with a mean and standard deviation was 2.43 ± 0.22But the lowest proline was measured in T_0 with a mean and standard deviation was 0.72 ± 0.09. According to our results it is said that, T_4 (T_4: urea = 60g/8m^2, TSP = 76g/8m^2, MOP = 36g/8m^2) is a suitable dose between different NPK doses to common buckwheat cultivation.

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LIST OF ABBREVIATIONS

g	=	Gram
ml	=	Mililiter
µl	=	Microliter
BMB	=	Biochemistry and Molecular Biology
BW	=	Buckwheat
BC	=	Black cumin
Mg/ml	=	Miligram per mililiter
µg /µl	=	Nanogram per microliter
°C	=	Degree Celsius
et al.	=	And others
nm	=	Nanometer
Cm	=	per centimeter
MDA	=	Methylenedioxyamphetamine
G	=	Gram
HW	=	Honeyweed
M	=	Mililitre

CHAPTER-I

INTRODUCTION

Buckwheat (*Fagopyrum esculentum*) is a dicotyledonous summer-growing plant in the Polygonaceae family that produces dark-hulled, triangular, starch-filled seeds (Halbrecq *et al*., 2005). Fagopyrum species can be classified into two categories: common buckwheat (*Fagopyrum esculentum*) and Tartary buckwheat (*Fagopyrum tataricum*) (Ali *et al*., 2018*)*. Common buckwheat is the most popular food item in Asian countries including China, Japan and Korea. It has recently been widely grown and consumed in Canada, North America and Europe. Interestingly, it is enriched in many nutritional properties such as protein, natural antioxidants, minerals, vitamins (especially B group) and dietary fiber (Siddiqui *et al.*, 2017). It is a low input plant that provides an important impact antioxidant source. Plain buckwheat proteins can inhibit gallbladder production more effectively than products with soy protein. They may delay breast carcinogenesis by reducing serum estrogen and inhibit colon carcinogenesis by reducing cell proliferation *(Taylor et al.*, 2010). The food crop tartary buckwheat has high anti-tumor and health advantages and it may also lower blood pressure, blood sugar, and lipid levels *(Zhu, 2014)*. Its yield, though, ranges around 1500 to 2400 kg/ha , which is quite low. To advance the buckwheat industry, it is crucial to produce Tartary buckwheat at a high and consistent yield (Ali *et al*., 2018). Buckwheat is widely cultivated in North America and is thought to have originated in North or East Asia. In China, it has been grown since at least 1000 BC. It has a very short growing period and a very high ability to adapt to difficult situations *(*Zhang *et al*., 2021*)*. Around the world, a wide variety is being grown, although primarily in the northern hemisphere. The most popular buckwheat variety right now is *Fagopyrum esculentum* Moench, usually known as common buckwheat or sweet buckwheat, however, *Fagopyrum tartaricum* may also be available in certain areas (Zhang *et al*., 2019). Buckwheat grain is an excellent source of protein and carbohydrate with a high content of unsaturated fatty acids and has a glycemic index that is low. Additionally, it has substances having preventive benefits. The bran fraction has the highest percentage of dietary fiber at 40%. Phytosterols in use can help to decrease blood cholesterol *(*Siddiqui *et al.*, 2017). Compared to other grains, buckwheat is a better supplier of magnesium, potassium, phosphorus, zinc, manganese, and copper. Pyridoxine is the vitamin that is most prevalent *(Erley et al., 2005).* Genetic background, environmental conditions and agriculture treatments are reported to be responsible for variation in starch composition, internal structure and physicochemical properties. Nitrogen is an important nutrient for crop growth and is suitable nitrogen can maintain and improve crop quality. Grain and cooking qualities may decrease under high nitrogen levels. Nitrogen fertilization has a significant effect on protein content and grain yield, and protein content is significantly positively correlated with nitrogen fertilization *(Schulte Auf'm Erley et al., 2005)*. Many studies have also shown that nitrogen fertilization affects the functional properties of starch. It was found that the peak viscosity (PV), cool paste viscosity and breakdown (BD) viscosity of rice starch could decrease significantly with increasing nitrogen levels. Believe that starch grain accumulation during grain filling can be affected by nitrogen fertilizer application (Witkowicz *et al*., 2021). Therefore, understanding the important quality characteristics and selecting appropriate nitrogen levels is essential to improve the quality characteristics and yield of common buckwheat. To date, few studies have reported the effect of different nitrogen levels on agronomic traits and stratch physicochemical properties of common buckwheat. The research was important for its potential use in modern common buckwheat production systems (Siddiqui *et al*., 2017). Phosphorus (P) is considered to be a limiting nutrient for crop production. P shortage results in a decrease in stomatal conductance and a slower rate of photosynthesis, which affects the metabolism of nitrogen and P as well as the buildup of dry matter in crops (Sardans and Peñuelas, 2021). Since P is typically immobilized in soil, most crops find it difficult to recover more P in the short term. However, by producing phosphatase and other compounds to break down phosphate, buckwheat roots may efficiently take P from the soil. Phosphate fertilizer can simultaneously balance nitrogen absence in the soil and encourage root growth, which supports buckwheat roots' great ability to store inorganic P and together determines plant tolerance to harsh conditions (Gairhe *et al*., 2018). The appropriate application rate of phosphate fertilizer is crucial for improving crop photosynthetic efficiency and yield belived that a reasonable application rate of phosphate fertilizer improves light energy utilization efficiency and dry matter accumulation rate, thereby increasing production. further reported that phosphate fertilizer significantly increases chlorophyll content, net photosynthetic rate, and maximum quantum yield (Fv/Fm), thus improving low light-utilization efficiency by enhancing PSII reaction center performance (Witkowicz *et al*., 2021). Nevertheless, there is still considerable room for improvement in promoting the growth and yield of buckwheat using phosphate fertilizer, particularly under varying planting densities (Gairhe *et al*., 2018). The supplies of water and nitrogen to a plant during its critical stages of growth are the main factors that define crop yield. A crop experiences irregular water deficits during its life cycle in rainfed agriculture. An effective anti-stress-oriented approach therefore ought to focus on increasing the units of water productivity (Management, 2021). The main objective of their conceptual review is to confirm that adequate K management can be used as an important tool to alleviate the negative effects of water deficit on plant growth, yield-component formation, and yield. The French and Schultz approach of using the water-limited yield (WLY) was modified in this review into a graphical form and was used to discriminate between yield fractions that depended on the volume of transpired water from those that were induced by K fertilizer. By using this method, it was possible to demonstrate the extent of several crop (winter wheat, spring triticale, maize, sugar beet) responses to the K supply *(Grzebisz et al., 2013).* Yield increases resulting from K application mostly appeared under conditions of mild water deficit. As described for sugar beet, finding the critical period of crop K sensitivity is a decisive step in understanding its impact on water-use efficiency. It has been shown that an insufficient supply of K during crucial stages in the yield formation of cereals (wheat, spring triticale), maize, and sugar beet coincides with a depressed development in the yield components. The application of K fertilizer to plants is a simple agronomic practice used to increase crop tolerance to a temporary water shortage. It may be that the improvement of a plant's access to K during mild water-deficiency stress will increase water uptake by the root cells, which in turn increases their osmotic potential and thereby allows extension growth. This growth in turn promotes access to other mineral elements (including nitrogen) and water, which favor plant growth and yield (Sardans and Peñuelas, 2021). Potassium, mostly as a cation (K^+), together with calcium (Ca^{2+}) are the most abundant inorganic chemicals in plant cellular media, but they are rarely discussed. K^+ is not a component of molecular or macromolecular plant structures, thus it is more difficult to link it to concrete metabolic pathways than nitrogen or phosphorus (Ullah, 2014). Over the last two decades, many studies have reported on the role of K^+ in several physiological functions, including controlling cellular growth and wood formation, xylem-phloem water content and movement, nutrient and metabolite transport, and stress responses. In this paper, we present an overview of contemporary findings associating K^+ with various plant functions, emphasizing plant-mediated responses to environmental abiotic and biotic shifts and stresses by controlling transmembrane potentials and water, nutrient, and metabolite transport (Wang *et al*., 2016). These essential roles of K^+ account for its high concentrations in the most active plant organs, such as leaves, and are consistent with the increasing number of ecological and agricultural studies that report K^+ as a key element in the function and structure of terrestrial ecosystems, crop production, and global food security *(Grzebisz et al., 2013).* We synthesized these roles from an integrated perspective, considering the metabolic and physiological functions of individual plants and their complex roles in terrestrial ecosystem functions and food security within the current context of ongoing global change. However, provide a bridge between studies of K^+ at the plant and ecological levels to ultimately claim that K^+ should be considered at least at a level similar to N and P in terrestrial ecological studies (Sardans and Peñuelas, 2021). Therefore, the objective of this study was to investigate the agronomic characteristics and antioxidant enzyme activities of common buckwheat treated by different NPK fertilizers levels. That's why, the main objectives are the effects of different NPK fertilizers on plant growth, yield and antioxidant enzyme activity related traits.

Objectives

a. To investigate the effect of NPK fertilizer doses on plant growth and yield related traits of buckwheat cultivars.
b. To know the activity of antioxidant enzymes (catalase, ascorbate peroxidase, and peroxidase) and hydrogen peroxide under different doses of NPK fertilizer applications.
c. To evaluate and recommend the suitable NPK fertilizer dose (s) for cultivation of the buckwheat genotypes in Bangladesh.

CHAPTER II
REVIEW OF LITERATURE

Review of related literature is necessity in the sense that it provides scope for reviewing the stock of knowledge and information relevant to the proposed research. Despite the fact that a few numbers of works have been done in Bangladesh related to this research, there are some published reports and related activities. However, the limited number of works so far published are mentioned here along with other related works. Plant nutrition and fertilizer management have a long history, and much has been written about the relationships between soil and plant nutrient status and crop development and yields. The majority of research focuses on the antioxidant enzyme activity, growth and yield of common buckwheat. Due to the bunch grass with upright tillers and the many variables involved in determining crop yields (rainfall, temperature, day length, and humidity), literature on NPK effect on buckwheat morphology, antioxidant activity, yield and growth is less prevalent and more complex. The most limiting element restricts plant growth and thus final production. A short description on the available literature relevant to the present investigation has been presented below:

2.1 Buckwheat

Buckwheat belongs to the dicotyledonous grass in the Polygonaceae family, genus Fagopyrum. Buckwheat is the generic term for two agricultural species: esculent buckwheat (*F. esculentum* Moench) and tartary buckwheat (*F. tataricum* Gaertn). Although it is not a triticeae, the agricultural use of buckwheat grains (achenes) is quite comparable to that of graminaceous crops, hence agriculturalists describe it as a triticeae. Different popular names of buckwheat were used to describe its spread across Asia and Europe and continue to be utilized to verify buckwheat's origins. In India, buckwheat is referred to as ogal, in Nepal called mite phapar, in Bhutan as jare, and in Russia as grecicha kul'turnaja. Grechka is a Ukrainian word, while gryka, tatarka gryka, or poganka are Polish word (Zhao *et al.*, 2018). In Sweden, it is called bovete, in Denmark, boghvede, and in Finland, tattari. In Slovenia, it is known as ajda, hajdina, or idina, and in Bosnia, Serbia, Montenegro, and Croatia as heljda. It is known as sarrasin, ble'noir, renoue'e, bouquette in French; gwinizh-du in Breton (Northwestern France); fagopiro, grano saraceno, sarasin, faggina in Italy; and Buchweizen or Heidekorn in German (Natho, 2008). In Korean, it's spelled maemil. Soba is the Japanese term for buckwheat noodle dishes. In Mandarin, common buckwheat is referred to as tian qiao mai, whereas tartary buckwheat is regarded as ku qiao mai.

2.2 History and Spread of Cultivated Buckwheat

De Candolle, (1883) Proposed a theory about buckwheat origin location over a century ago, and suggesting that buckwheat originated in Siberia and the northern part of China. In 1957, Nakao pointed out that De Candolle was incorrect because there were many wild buckwheat species widely distributed in the southern part of China, indicating that the southern part of China may have been the original location of buckwheat rather than Siberia or the northern part of China (Konishi *et al*., 2005). Since the 1980s, Chinese agriculturists and buckwheat specialists have found an enormous number of wild buckwheat species in southern China, resulting in various fresh perspectives. Singh *et al*., (2020) argued that the Daliangshan region is among the sites of origin of Tartary buckwheat based on the large quantity of wild buckwheat, peculiar ecological circumstances, folklore, and practices involving buckwheat in this region. Chen, (1999) argued that buckwheat developed in the southwestern part of China, on the east side of the Himalayas, based on their investigations into buckwheat history, historical events, and Yunnan national history. Buckwheat is thought to have originated in Yunnan, as well as the Daliangshan and Xiaoliangshan regions, which served as borders between Yunnan and Sichuan. According Campbell, (1997), the southwestern region of China was not only a differentiation and spread center, but it was also the genesis of the Fagopyrum Mill. Ohnishi, (1998) confirmed the wild relative species F. esculentum ssp. ancestrale as the ancestor of the cultivated species F. esculentum Moench using morphology, biology of reproduction, isozyme assessment, RAPD, and AFLP. Tsuji and Ohnishi, (2000) investigated the association between the wild related species *F. tataricum* ssp. potanini and the cultivated species *F. tataricum* Gaertn. using isozyme analysis, RAPD, and AFLP. Their research indicates that the joint region of Yunnan and Sichuan, as well as the eastern section of Tibet, are the places where cultivated Tartary buckwheat first appeared. According to studies on the distribution of wild buckwheat species in southwest China, the history of buckwheat cultivation, Chinese history, and molecular systematics, the southwest region of the country was not only a hub for buckwheat allocation and diversity but additionally the source of the domesticated species (common buckwheat and Tartary buckwheat). Today, everyone agrees on these findings (Zhou *et al*., 2018).

2.2.1 Original place of tartary buckwheat

The mountains of southwest China are where tartary buckwheat is most commonly grown. The morphology of different populations varies somewhat. Totally self-pollinating is tartary buckwheat. Its traits, such as nectarines and some stamens with specific degenerations, small, green flowers that frequently do not open and close fertilized, extremely short styles and stamens, and so on, imply that tartary buckwheat transformed from cross-pollination species with heterostyle under lower temperatures that are undesirable to insects' cross-pollination. Wild tartary buckwheat is a weed that thrives in fields, wooded areas, riverbanks, ditch banks, and waste hillside. Since it is primarily found in the cooler, high-altitude regions of the eastern Qing Zang Plateau, Chuan Xi Plateau, and Yun Gui Plateau (frequently > 1500 m above sea level), tartary buckwheat probably evolved in these regions. According to reports, the eastern Qing Zang Plateau, which includes Metuo, Bimi, Chayue, Linmang, Nielamu, and MiEn, as well as Tibet, is home to the majority of the hypothesized progenitor species (F pilus) of F tataricum, which overlaps the region where the wild forms of F tataricum are prevalent. The data suggest that Tibet in southern China may have been the origin of tartary buckwheat. (Zhou, *et al*., 2018).

2.2.2 Original place of common buckwheat

The most widespread buckwheat species worldwide is F. esculentum, which is cultivated. It can be found, among other areas, throughout North America, Europe, and Asia. However, populations indigenous to various places do not differ significantly in form or isozyme (Ohnishi, 1993); (Chen, 1999b). Southwest China's Yunnan province, particularly in Lijiang and Yongsheng, is where wild *F. esculentum* is most commonly found (Ohnishi, 1991). There are some variations among various populations. Several causes, including the following, contribute to the paucity of variation in common buckwheat: (1) A brief account of cultivation that has mostly remained unchanged. The first mention of buckwheat farming is found in the Shen Nong Shu, an ancient Chinese text from the third to the fourth century B.C. (Lin, 1994). The earliest mention of archaeology dates to the west Han Dynasty (206 B.C.–8 A.D.) (Lin, 1994). This shows that buckwheat farming has a recent history compared to wheat. (2) The results of artificial elements like artificial spreading and the cross-pollination reproductive system. These effects can lower divergence among diverse wasteland on hillside and vast farming in rural China, which was reclaiming waste mountain, and enable variation interchange across various communities. These reduce the difference between *F. esculentum* from farms and wild ones. Southwest China is home to a large number of related species, the bulk of which belong to the small-achene group, including the hypothesized progenitor species of F. esculentum. According to how sensitive it is to light and temperature, common buckwheat may be divided into three ecotypes: medium ecotype, summer ecotype, and autumn ecotype. Although this type can grow, bloom, and give fruit in both spring and autumn, autumn is when it performs best. A primordial ecotype is the autumn ecotype. The accessions of *F. megaspartnium* native to Chengdu, Sichuan, have extremely susceptible to temperatures and light within the large-achene category, which means they only distinguish inflorescence and flowers for very little time in autumn (often August to September in Chengdu, Sichuan, China), suggesting that the ecotype of *F. megaspartnium* is rigid autumn ecotype that is more primitive than that of *F. megaspartnium*. The fall ecotype of *F. megaspartnium* is also found in accessions from Yunnan and Guizhou, where the plants and achenes are more similar to F. esculentum than those from Sichuan. Although some Yunnan and Guizhou accessions are purely fall ecotype, others are not.

2.2.3 Distribution of the buckwheat species

Buckwheat is regarded to not be very old, despite being grown in China as early as the second and first centuries BC (Li and Yang, 1992). The earliest Chinese records on buckwheat indicate that there was a sizable output in China between the fifth and sixth century AD (Krotov, 1963). It was probably grown in China for a thousand years or so before traveling through Russia to reach Europe. It was brought to Europe throughout the Middle Ages, perhaps from Siberia, and reached Germany around the beginning of the 15th century (Hughes and Hensen, 1934). It seems that widespread cultivation did not begin in Russia until the 15th century (Krotov, 1963). There is no evidence to suggest that buckwheat was grown in ancient India. It was found in most of Europe, India, and China in the 17th century. Since buckwheat was frequently grown on recently cleared ground, it was brought to North America by immigrants from other countries. In the Himalayas, common buckwheat could only be grown at elevations between 500 and 2500 meters, according to (Ohnishi, 1988). In planting patterns over 2500 m, tartary buckwheat takes its place. He asserts that the recent cultivation of buckwheat in this region could be a feasible explanation after noticing a considerable regularity in the allelic frequencies of isozyme loci. Another, maybe more plausible, hypothesis is that farming in this region has a long history, but that allozyme frequency was uniform due to periodic migration from the surrounding populations. Multiple trade routes that passed through this region from southern China and Tibet may have been the site of migration. Ohnishi found that differentiation took place at the boundary of the distribution, which lacked sufficient genetic resources for migration. Buckwheat may have been introduced to Japan circa 3000 years ago, according to archeological findings (Nagatomo, 1984). Buckwheat had already been widely grown as a catch crop when it first appeared in Japanese chronicles in the eighth century. Buckwheat was introduced to Japan via Northern China via the Korean peninsula, according to Nagatomo (1984) and Murai and Ohnishi (1995). Early Jomon (7000–5000 BP) buckwheat farming is known to have existed in Japan.

2.3 Common buckwheat (*Fagopyrum esculentum*)

Common buckwheat (Fagopyrum esculentum), a member of the Polygonaceae family, is a pseudograin. Its origins are in a province in southwest China. After domestication, which took place around 3000 BCE, this East Asian crop was transported to the Middle East and Far East. Since the latter half of the 15th century, its cultivation in Europe has been documented. The seeds, which are exclusively achenes, are frequently classified as cereal grains even though the crop is not a cereal because of their similar function. The dehulled groats are often used to make porridge, and the flour is used to make pancakes, cookies, noodles, cereals, and other foods (Mahata, 2018). The grain is typically fed to livestock or poultry. According to Takeshima *et al*., (2020), the summer (weakly photoperiod sensitive), autumn (extremely photoperiod sensitive), and intermediate ecotypes of common buckwheat are all considered quantitative plants with short days (Joshi, *et al*., 2019). For adaptive expansion and yield maximization, it is essential to comprehend ecotype diversity. The typical growing season for buckwheat is between 70 and 90 days, and it may flourish in a range of conditions, including cool climates, high elevations, sandy soils with good drainage, marginal areas, and acidic soils (pH 5) (Farooq *et al*., 2016). Its seeds have a high carbohydrate content that is gluten-free, excellent protein that has a well-balanced amino acid profile, as well as anti-oxidative, antihypertensive, and anti-obesity effects (Bonafaccia *et al*., 2003; Suzuki; *et al*., 2020). Future food security may be improved by expanding this priceless orphan crop's production area and productivity. Although buckwheat is cultivated in a wide range of latitudes, the growth regions and seasons of each genotype are strictly regulated. Determining each genotype's genetic adaptation is the first step in extending cultivation areas and raising productivity. According to Morishita and Hara (2020), the differentiation of each cultivar into ecotypes suitable for specific growing environments is thought to be the cause of buckwheat's adaptability. According to growing regions and seasons, buckwheat is classified as an empirical short-day (SD) plant having three ecotypes: the autumn ecotype, which is strongly photoperiod sensitive, the summer ecotype, which is weakly photoperiod sensitive, and the intermediate ecotype. When cultivated in the summer (long-day, LD), autumn-ecotype cultivars feature delayed flowering, a low seed-set ratio, continuous flowering, rapid vegetative growth, late maturation, and a significantly lower yield than when grown in the summer. Contrarily, summer-ecotype cultivars have early flowering, a high seed-set ratio, and early maturation when grown in the summer, but their yield is marginally lower when grown in the fall than when grown in the summer (Matano and Ujihara, 1979; Michiyama *et al.*, 2005). For responsive development and production maximization, it is essential to comprehend ecotype distinction since a mismatch among ecotype and environment drastically limits yield potential. The only genetic research on buckwheat ecotypes, however, has focused on the relationship between day duration and blooming period (Hara *et al.*, 2011; Hara *et al.*, 2020). The relationship between flowering time and photoperiod has frequently been studied to determine the ecotype of buckwheat, but day length also appears to have an impact on flowering time and post-flowering reproductive development. For instance, summer culture decreases pollen fertility rates and the capacity to established seed within autumn ecotype varieties as well as certain intermediate-ecotype cultivars, improves the number of malformed flowers, impedes pistil development, causes unusual embryo sacs to form after pollination, and hinders pistil growth (Sugawara, 1958; Guan *et al.*, 2001). Thus, understanding the genetic basis of ecotype differentiation is required.

2.3.1.1 Flower morphology of common buckwheat

Common buckwheat comprises a polygonal perianth at the anthesis that is 6-7 mm in diameter and orthogonal with the pedicel (Campbell, 1997; Cawoy *et al.*, 2006a). It is constructed of five unconnected petaloid tepals. Two stamen swirls make up the androcium. According to Campbell (1997), the external cycle consists of five stamens with introrse breakdown while the inner cycle consists of three stamens with extrorse dehiscence. Exterior cycles filaments are around 1.2 times larger and hold 7.5 % fewer grains of pollen than interior phase stamens. The superior monocular ovary includes an orthotropous ovule and three distinct styles make up the gynoecium (Campbell, 1997).

2.3.1.1.1 Flowering and seed ripening

According to Alekseyeva and Bureyko (2000) and Halbrecq *et al.*, (2005), blooming persists for four to fifteen weeks and begins approximately four to six weeks from seeding. According to Quinet *et al*. (2004), flowers normally spend just a single day in anthesis. Flowering occurs acropetally (derived the bottom toward the top) following this axis, with the shortest inflorescence appearing on the next or fifth node below the cotyledons that make up the central stem. Under these inflorescences, the axillaries flower acropetally (from the top of the plant to the bottom) while the axillaries flower basipetally (as well as the primary stem). Along the inflorescence scales, flowering starts when the inflorescence anthesises. A second flower begins the anthesis between the cymes shortly after the first one as flowering progress from the bottom to the peak of each inflorescence. The largest number of flowers each day are anthesised during the height of flowering, which takes place 2 to 3 weeks following the first anthesis (Halbrecq *et al.,* 2005). There are seasonal variations in the process of development of the embryo, and it requires the embryo approximately ten days from pollination to achieve its greatest size and another fourteen days towards grain development. The size of each zygote for three days after fertilization is 1.7 times higher in the autumn, and it splits more frequently in the summer than during the fall. A triangular achene with a length of around 5 mm is the fruit (seed). The embryo is found in the centre of the its endosperm which is covered by a fine testa beneath the pericarp (Marshall and Pomeranz 1982). The pericarp, which was green throughout seed filling, inevitably turns brown and becomes grey as the seed ages. It is difficult to choose the best time for harvesting due to the abundance of flowers (tens per plant and per day), the lengthy flowering period, and the lengthy ripening duration (seeds at various stages of ripening interact within crops and inflorescences) (Funatsuki *et al*., 2000). Additionally, even though there isn't a breakdown layer, the pedicels are delicate, and air can cause grain to blow across the soil. By affecting the development and fertility of sexual structures, temperature controls yield during blooming (Björkman, 1995c). Because of its vulnerability to frost, buckwheat production is limited to low elevations in temperate areas, and harvest needs to be finished before first frosts. It thrives at temperatures between 18 and 23°C. Flowers fade early around 10°C, while flowering is hampered at 15°C (delayed onset and decreased blossom quantity) (Tahir and Farooq, 1988). High temperatures (25°C) along with dry winds are responsible for flower dying, abortion of sexual components during development, anomalies in embryo sacs, and fruit dehydration (Slawinska and Obendorf, 2001). Flooding and water restrictions are harmful to endosperm maturation as well as growth during flowering. They may result in lighter mature seeds and also embryo abortion (Sugimoto and Sato, 2000). For pollen to survive, there must be dampness. Pollen loses all viability under one hour while exposed at 23°C in dry conditions. According to, the likelihood of fertilization is reduced by 45 % when pollen from plants cultivated at 25°C instead of 18°C.

2.4 Nutritional properties of common buckwheat

Buckwheat has enormous promise as a food ingredient, particularly in the functional food market. Buckwheat, a pseudo-cereal, includes high-quality protein, fiber, resistant starch, rutin, D-chiro-inositol, vitamins, and minerals. D-chiro-inositol, fagopyritols (galactosyl derivatives of D-chiro-inositol), resistant starch, and buckwheat protein product all had good impacts on rats' health, but further research is needed to determine effects on people. Buckwheat's primary antioxidants, rutin and quercetin, have been linked to the treatment of chronic venous insufficiency. Buckwheat groats (dehulled seeds) have a similar nutritional content to cereals. Buckwheat includes a high level of polyunsaturated essential fatty acids like linoleic acid, as well as starch and fiber in comparable levels. Several vitamins (B1, C, and E) are present, as are minerals in abundance. Buckwheat protein has a higher nutritious quality than cereal protein due to its higher lysine content. On the other hand, reduced digestibility has been observed, probably due to tannins, phytic acid, and protease inhibitors. Some protease inhibitors can potentially cause allergic responses in people. Malting may improve the nutritional and functional qualities of buckwheat by enhancing protein digestibility and the quantity of nutritional and functional components (Wijngaard and Arendt, 2006).

2.4.1 Carbohydrates

2.4.1.1 Starch

The total carbohydrate percentage of common buckwheat groats was 67.8-70.1% (Li and Zhang, 2001); (Steadman *et al*., 2001a), with starch accounting for 54.5% (Steadman et al., 2001a). Because of the presence of the pericarp, buckwheat seeds had a greater carbohydrate percentage of 73.3% but a similar starch percentage (55.8%). Tartary buckwheat had a slightly greater percentage of starch (57.4 0.12%) than common buckwheat, however this could be related to the cultivar studied in this study (Bonafaccia *et al*., 2003b). Although an apparent amylose content of 46.6% has been determined, lower apparent amylose values of 21.1-27.4% have been reported in subsequent publications (Yoshimoto, *et al*., 2004). These ranges correspond to cereal amylose percentages (Qian and Kuhn, 1999b). There were no consistent changes in apparent amylose percentages between common and tartary buckwheat (Li *et al* 1997). Because iodine affinity rises when buckwheat starches are defatted, the presence of amylose-lipid complexes has been proposed (Yoshimoto, *et al*., 2004). (Qian *et al*., 1998) support this assumption. Buckwheat starch contains a high concentration of crude fat and amylose. In addition, differential scanning calorimetry study revealed a second melting transition peak temperature of 84.5 °C. Both findings could indicate the presence of amylose lipid complexes. The development of amylose-lipid complexes may limit swelling power and solubility (Qian *et al.*, 1998). (Yoshimoto, *et al.*, 2004) discovered a significant disparity between the apparent amylose content (25.5-26.5%) and the actual amylose content (15.6-17.9%). This distinction suggests the presence of a high concentration of longer chain amylopectins, which have a proclivity to compound with iodine (Yoshimoto, *et al.*, 2004). (Praznik *et al.*, 1999) and (Noda, *et al.*, 1998) validated the occurrence of long chain branching compounds. The degree of polymerization (DP) of amylopectin varied depending on the botanical origin and therefore across buckwheat starches, but 4% of amylopectin had a DP of 6 and 7, while > 40% had a DP of 10-12 (Noda, *et al.*, 1998). This suggests that buckwheat amylopectins include more long chains than cereal amylopectins. This indicates the presence of a larger level of super molecular glucan structures, which can influence viscosity. Super molecular glucan complexes can breakdown during gelatinization, however they tend to reconstruct following chilling. Buckwheat starch had greater peak viscosities than cereal starches and pasted more like root and tuber starches. Aside from super molecular glucan structures, this high viscosity values can be attributed to buckwheat starches having a larger granule swelling and gelling tendency than cereal starches (Yoshimoto, *et al.*, 2004). Peak gelatinization temperatures varied slightly depending on the buckwheat variety. Peak gelatinization temperatures of buckwheat starches have been observed to range from 57.2 to 66.7°C by (Noda, *et al.*, 1998), however most peak gelatinization temperatures have been characterized within a range of 63.7-70.8°C (Yoshimoto, *et al.*, 2004). It should be noted that the equipment used to assess gelatinization qualities can have an effect on gelatinization curves. Different gelatinization onset temperatures (60, 70, and 80°C, for example) have been documented using differential scanning calorimetry analysis, fast viscoanalysis, and Brabender visco amylography, respective (Qian and Kuhn, 1999a). Buckwheat starch has a higher water-binding capacity than wheat and corn starch at 109.9%. The tiny size (and thus larger surface area) of buckwheat starch granules explains this high value (Qian *et al* 1998).

2.4.1.2 Resistant Starch

Based on the pace and breadth of digestion in vitro, starch is classified into three types: fast digestible starch, slowly digested starch, and resistant starch (RS). Physically inaccessible starch, native granular starch, and retrograded starch are the three types of RS (Englyst *et al.*, 1992). Undigested starch may have favorable nutritional effects similar to those seen with fiber (Skrabanja and Kreft, 1998). The physical structure of starch, the level of retrogradation, the amylose-to-amylopectin ratio, and non-starchy inhibitory components all influence starch digestibility. RS was found in raw buckwheat groats with a total starch level of 73.5%. Processing can have an impact on RS. For example, autoclaving reduced RS from 33.5% to 7.5%. In contrast, autoclaving or boiling can increase the percentage of retrograded starch from 1% to 4-7%. Rats excreted 0% starch from native buckwheat groats versus 1.0-1.6% starch from hydrothermally processed buckwheat. As a result, it was proposed that rats (and possibly humans) can digest native buckwheat starch but not hydrothermally processed buckwheat starch (Becker and Hedtke, 1995).

2.4.1.3 Fiber

Many studies have suggested that dietary fiber (DF) may be beneficial in the prevention of certain diseases (Roberfroid, 1993). Insoluble fiber (IDF) and soluble fiber (SDF) are the two types of DF. IDF typically contains lignin and cellulose, whereas SDF typically contains pectin and gums (Steadman *et al*., 2001a). SDF, in particular, may benefit human health by lowering blood cholesterol levels (Roberfroid, 1993). Furthermore, DF may bind proteins and minerals, inhibit digestive enzymes, and so reduce digestibility or absorption. Buckwheat seeds have a DF of 27.38%, according to (Bonafaccia *et al*., 2003b). DF is mostly found in the seed coat and hull, which explains why buckwheat groats have a much lower DF percentage of 7% (Bonafaccia *et al*., 2003).

2.4.1.4 D-Chiro-Inositol

D-Chiro-inositol is an inositol isomer found in buckwheat seeds in quite high concentrations (Steadman, *et al*., 2000). When D-chiro-inositol was delivered intravenously or orally, it reduced high plasma glucose in spontaneously insulin-resistant rhesus monkeys, streptozotocin-treated hyperglycemic rats, and normal rats (Ortmeyer *et al*., 1993) (Ortmeyer *et al*., 1995). Because buckwheat has a relatively high level of D-chiro-inositol, providing doses of 10 and 20 mg of D-chiro-inositol in the form of natural buckwheat concentrate reduced serum glucose concentrations in streptozotocin-diabetic rats by 12-19%. Although no human studies of D-chiro-inositol have been published, buckwheat concentrate has been proposed as a natural product to aid in the treatment of diabetes (Kawa *et al*., 2003). (Steadman, *et al*., 2000) suggest a free level of D-chiro-inositol in buckwheat groats ranging from 20.7 to 41.7 mg 100/g, dwb, although the majority of D-chiro-inositol in buckwheat is present as fagopyritols (Horbowicz *et al*., 1998). Fagopyritols are galactosyl derivatives of D-chiro-inositol that accumulate in some species' seeds. Buckwheat embryos are distinguished by the accumulation of galactosyl cyclitols rather than raffinose oligosaccharides. D-chiro-inositol, fagopyritol B1, B2, and B3 structures. Fagopyritols, like other soluble carbohydrates like sucrose, are mostly found in buckwheat embryos (71.4%). Fagopyritol B1 is the most abundant fagopyritol in buckwheat seeds, accounting for 41.16% of soluble carbohydrates in embryos (g, dwb). In addition to B1, two digalactosyls and two trigalactosyls have been found in embryos from common buckwheat seeds (Steadma *et al.*, 2001c). They were found at a concentration of 2 mg/g, dwb, of total soluble carbohydrates (Horbowicz *et al.*, 1998). Tartary buckwheat had half the amount of fagopyritols as normal buckwheat. Tartary buckwheat had a soluble carbohydrate not found in ordinary buckwheat varieties. This molecule, which was detected at a level of 31%, was presumably rhamnosyl glucoside (Steadman et al 2000). Malting is one method for increasing fagopyritol levels. Germinating at 18°C increased fagopyritol B1, while germinating at 25°C increased both A2 and B2 levels. It is necessary to explore the effects of D-chiro-inositol and fagopyritols in natural buckwheat concentrate on plasma glucose levels in humans. These substances may have beneficial effects on diabetic people, and products may be designed appropriately.

2.4.1.5 D-chiro-inositol (DCI)

DCI is a functional polysaccharide found in common buckwheat that has insulin-like biological activity and is primarily contained in the germ. In mice, oral administration of DCI-rich Tartary buckwheat bran extracts reduced glucose, C-peptide, glucagon, lipid, and urea nitrogen levels while improving glucose tolerance (Yao, *et al.*, 2008). Common buckwheat DCI contains physiological characteristics such as regulating liver lipid metabolism and acting as an antioxidant, as well as antiaging and anti-inflammatory properties. DCI is found in common buckwheat in both free form and as its galactosyl derivative, which is the major form (Hu, *et al.*, 2015). DCI can be produced by hydrolyzing common buckwheat sugar alcohol with -galactosidase. However, because -galactosidase is not present in human stomach contents, the level of free DCI in foods must be boosted chemically (Yao, *et al.*, 2008).

2.4.2 Protein

2.4.2.1 Storage Proteins

By solubility, all proteins can be classified into four categories: Albumins (soluble in water and dilute buffers at neutral pH); globulins (soluble in salt solutions but insoluble in water); glutelins (soluble in dilute acid or alkali solutions); and prolamins (soluble in aqueous alcohols of 70-90%) (Bewley and Black, 1985). Prolamins are the most common storage proteins in cereals, whereas globulins are mostly found in buckwheat. According to Ikeda *et* *al*. (1991), common buckwheat seeds contain 64.5% globulin, 12.5% albumin, 8.0% glutelin, and a tiny amount of prolamins (2.9%) (Radovic *et al*., 1996). reported similar ranges. The principal storage proteins, globulins, are divided into two families based on molecular weight (MW) and sedimentation coefficients (S values) of 7 (average 7-8) and 11 (11-13) during ultracentrifugation. Both are made out of regular subunits (Bewley and Black, 1985). 13S globulin and 8S globulin have been found in buckwheat. 13S globulin has a MW of 280,000. It is a legumin-like storage protein made up of nonidentical subunits made up of one acidic and one basic polypeptide bound together by disulfide bonds. 13S globulin is a key storage protein in buckwheat, accounting for 33% of total seed proteins (Radovic *et al*., 1996). Trimer is the smaller 8S globulin. It is made up of subunits with MWs ranging from 57,000 to 58,000 and is related to vicilin-like storage proteins. In buckwheat, 8S globulin accounts for 7% of total seed proteins (Milisavljevic, 2004). Buckwheat has 2S albumins in addition to globulins. These water-soluble proteins were single-chain polypeptides with molecular weights ranging from 8,000 to 16,000. The majority of the protein in buckwheat is found in protein bodies. Protein bodies are unusual cellular organelles with dimensions ranging from 1 to 10 m and are surrounded by a single membrane. The 13S protein was discovered solely in the cotyledons, but the 8S globulin was discovered in both the cotyledons and the endosperm.

2.4.2.2 Resistant proteins

Buckwheat resistant proteins are also efficient in decreasing blood cholesterol (Tomotake, *et al*., 2000). According to Huff and Carroll (1980), the ratios of lysine/arginine (Lys/Arg) and methionine/glycine (Met/Gly) are the most important determinants in determining the cholesterol-lowering effects of proteins. Buckwheat has much lower Lys/Arg and Met/Gly ratios than other plant proteins. Nutritional studies have also revealed that buckwheat proteins have the strongest cholesterol-lowering effects of any plant protein known to science (Huff and Carroll, 1980).

2.4.2.3 Active proteins

The protein composition of typical buckwheat ranges from 8% to 19%; the amino acids are well balanced, and the bioavailability is high. Albumin, globulin, and gliadin are the proteins found in common buckwheat, in descending order of concentration. Albumin, globulin, and gliadin make up 51.52%, 13.69%, and 8.83% of the total (Sytar et al., 2016).

2.4.2.4 Protease inhibitors

Serine protease inhibitors and trypsin inhibitors are abundant in common buckwheat. According to (Ruan *et al*., 2015) Tartary buckwheat trypsin inhibitors can limit the growth of plant pathogenic fungal hyphae as well as spore propagation (Ruan *et al*., 2011). These researchers also discovered that Tartary buckwheat trypsin inhibitors had a substantial inhibitory effect on the growth of *Mamestra brassicae* (Ruan *et al*., 2015). Germination can diminish the inhibitor's activity, allowing for greater use of amino acids in common buckwheat proteins (Ren, *et al*., 2014).

2.4.2.5 Allergenic protein

Buckwheat includes allergenic proteins as well (Chen *et al*., 2011). The most common allergic reactions to Tartary buckwheat products are eczema or urticarial, acute bleeding, and anaphylactic shock due to a rapid drop in blood pressure (Zheng *et al*., 2011). Immunochemical experiments revealed that the allergic protein derived from Tartary buckwheat seeds has a molecular mass of 20-60 kDa, whereas the most sensitive allergenic protein has a molecular mass of 24 kDa (Cho, *et al*., 2015).

2.4.2.6 Amino Acids

Buckwheat is not a cereal grain and contains very little or no prolamins. This is why Coeliac patients can eat buckwheat (Fasano and Catassi, 2001). In cereal grains, where the percentage of prolamins is high, lysine is scarce, tryptophan is scarce, and threonine levels are deficient. Buckwheat protein is mostly made up of globulins and albumins, but it also contains a variety of amino acids. Pomeranz and Robbins (1972) investigated the amino acid content of ten different buckwheat seed samples. A protein proportion of 13.7% was discovered, with 17 amino acids identified. The first nine amino acids in Table II are required by the organism and must be consumed (Insel, 2004). Buckwheat protein has a high quantity of lysine (6.1% of 100 g of amino acid recovered) when compared to cereal grains. Furthermore, when compared to cereals, buckwheat contains significant levels of arginine (9.7%) and aspartic acid (11.3%), but low levels of proline (3.9%) and glutamic acid (18.6%). (Javornik *et al*., 1981); (Eggum, 1981) observed similar findings. The fact that prolamins, which are abundant in proline and glutamine, are found in extremely low amounts in buckwheat explains the comparatively low quantity of proline and glutamic acid. The amino acid makeup of the buckwheat kernel was identical in all sections. The sole difference between the two is the amino acid makeup.

2.4.2.7 Thiamine-binding protein (TBP)

TBP was first identified in Tartary buckwheat seeds by (Mitsunaga *et al*., 1986) Competitive inhibition tests revealed that protein binding to thiamine is extremely selective (Mitsunaga *et al*., 1986). TBP from Tartary buckwheat seeds can withstand only modest concentrations of denaturing chemicals like 2 Murea, which can stabilize thiamine by generating a protein-thiamine complex. This compound can be degraded by protease after intake, releasing the thiamine (Watanabe *et al*., 1998). TBP can thus be used to treat persons who are unable to store or do not have enough thiamine. Although the subunit structure and immunological properties of Tartary buckwheat TBP differ from those of rice and sesame, the thiamine binding method is identical.

2.4.3 Lipids

2.4.3.1 Fat-soluble substances

The content of free lipids in common buckwheat flour is larger than that of bound lipids, however after extrusion, the opposite result is found (Janeš *et al*., 2012). Some fat-soluble chemicals, particularly flavonoids, polypyrrolidols, phenolic compounds, volatile compounds, triterpenes, and phytosterols, are also important biologically active substances in Tartary buckwheat. Five phytosterols were identified from the seeds of *F. tataricum*, including stigmsat-4-en-3, 6-dione, -sitosterol palmitate, ergosterol peroxide, stigmasterol, and -sitosterol.

2.4.3.2 Fatty acids

Tartary buckwheat includes a high concentration of unsaturated fatty acids (approximately 79.3% of total fatty acids), primarily C18:1, C18:2, C18:3, and C20:1, with a quantity as high as 36.6% of the human essential polyunsaturated fatty acid linoleic acid (C18:2). Tartary buckwheat contains unusually high levels of oleic acid (C18:1) and linoleic acid, and linoleic acid is the most significant fatty acid in the human body, where it cannot be generated, and is the only essential fatty acid (Ren, *et al*., 2014). It is known as vitamin F. It is beneficial to the growth and development of young children. It can help adults avoid coronary heart disease (Ren et al., 2013). For regular physiological functioning and metabolism, cholesterol must be paired with fatty acids. In the absence of linoleic acid, cholesterol binds to saturated fatty acids, causing metabolic disturbances, deposition on blood vessel walls, and, finally, atherosclerosis, which leads to cardiovascular and cerebrovascular illnesses (Zhu, 2016). As a result, linoleic acid has earned the moniker "vascular scavenger," preventing and treating atherosclerosis, cardiovascular disease, and senile obesity.

2.4.4 Polyphenolic compounds

2.4.4.1 Phenolic acid

Caffeic acid, 6-coumaric acid, p-coumaric acid, ferulic acid, tannic acid, p-hydroxybenzoic acid, syringic acid, and gallic acid are the most prevalent phenolic acids in tartary buckwheat (Włoch, *et al*., 2016). Tartary buckwheat has a greater p-hydroxybenzoic acid concentration than normal buckwheat. Furthermore, protocatechuic acid accounts for 65.54% of the phenolic acids in Tartary buckwheat hulls, while p-hydroxybenzoic acid accounts for 58.61% and 78.82% of the total phenolic acid content in Tartary buckwheat bran and core powder, respectively (Sadauskiene, *et al*., 2018).

2.4.4.2 Lectin

Lectin is a glycoprotein found in a variety of buckwheat species (Bai *et al*., 2015a). Lectin has a number of positive biological properties, including anti-insect, antifungal (Herre *et al*., 2004); (Dhuna, *et al*., 2005), anticancer, antiviral (Balzarini, *et al*., 1992), and immunomodulatory (Rubinstein et al., 2004). Purified lectins must be characterized in order to explore the biological activities of lectins obtained from Tartary buckwheat grain. Tartary buckwheat lectin can stimulate the development and proliferation of dendritic cells (DCs) in the peripheral circulation. It efficiently stimulates T-cell immune response. In a cell, lectin induces the synthesis of proinflammatory factors such as interleukin-10 and interleukin-12. The helper T-cell 1 or T-cell 2 immunological response will be affected by the production of these cytokines for maturing peripheral blood DCs. Furthermore, Tartary buckwheat lectin proteins can trigger apoptosis in human leukemia U937 cells. As a result, Tartary buckwheat-derived lectins appear to be promising dietary supplements for the treatment and prevention of solid and blood malignancies.

2.4.4.3 Trypsin Inhibitor

Although there is evidence that protease inhibitors can trigger apoptosis in several tumor cell lines, the underlying processes are unknown. Buckwheat trypsin inhibitor has been shown in vitro to strongly limit the proliferation of two blood cancer cell lines: K-562 chronic myelogenous leukemia and IM-9 multiple myeloma (Wang, *et al*., 2007); (Zhang *et al*., 2007). Recombinant buckwheat trypsin inhibitor (rBTI) has been shown to suppress cell growth and proliferation of H22 hepatic cancer cells in vivo and in vitro (Bai, *et al*., 2015b). This apoptotic induction is mediated by caspase-9 in the mitochondrial pathway in a dose- and time-dependent manner. Furthermore, (Wang *et al*., 2015) reported that rBTI has a similar activity that can trigger apoptosis in cancer cells. The anti-mitophagy impact of rBTI on human liver cancer cell line (Hep G2) was implicated in mitochondrial dysfunction by raising intracellular total reactive oxygen species (ROS), which resulted in mitophagy by triggering subcellular degradation. It is worth noting that rBTI appears to target a subunit of the mitochondrial outer membrane complex's translocase. Taken together, the findings suggest that rBTI may play a role in cancer medication development by targeting mitochondria penetrating peptides for cancer treatment.

2.4.4.4 Rutin

Rutin, orientin, isoorientin, vitexin, isovitexin, and quercetin are the primary flavonoids found in buckwheat (Liu *et al*., 2008). The basic structure of buckwheat flavonoids such as rutin, isoquercetin, and quercetin. Rutin is recognized as the finest health-preserving flavonoid among antioxidant chemicals, and it is also known to be highly protective against inflammation and carcinogenesis (Nile and Park, 2013). Tartary buckwheat seeds and seed sprouts have 47 times more rutin than normal buckwheat seeds and seed sprouts (Kim *et al*., 2008). Rutin, as a flavonoid glycoside, may absorb UV-B light and so protect plants from solar radiation. Rutin glucosidase, the enzyme that destroys rutin, had the maximum enzyme activity in buckwheat leaves 28 days after planting (Suzuki *et al*., 2005). Buckwheat rutin therapy was reported to decrease cardiomyocyte hypertrophy (Chu *et al*., 2014). Angiotensin II-induced hypertrophy can significantly increase intracellular Ca^{2+} levels, calcineurin activity, protein expression, and protooncogene c-fos mRNA transcripts. Buckwheat rutin treatment inhibited calcineurin activity in a concentration-dependent manner in a hypertrophy model system. However, the regulation mechanism of buckwheat rutin in signal transduction pathways in cardiac myocytes remains unknown.

2.4.4.5 Quercetin

Other flavonoid chemicals found in buckwheat seeds and brans, in addition to rutin, include quercetin, isoquercetin, and fagopyrin, which have cytotoxic effects on human hepatoma HepG2 cells (Li, *et al*., 2014). Among the flavonoids, quercetin has the highest antioxidant capacity against the HepG2 cell line. In addition, quercetin inhibited hepatocellular carcinoma growth in a time- and dose-dependent manner. Furthermore, quercetin greatly raised intracellular ROS levels and apoptotic cell death, resulting in cell cycle arrest in the G2/M phase. Thus, buckwheat-derived quercetin is projected to be an efficient antitumor agent, and it demonstrated a synergistic impact in a clinical trial employing combination therapy with a known anticancer medication.

2.4.4.6 Anthocyanin

Buckwheat leaves are dried and sold as herbal tea in Europe under the brand name "Fagorutin." Buckwheat also contains a high concentration of water-soluble anthocyanin, the pigment that determines the color of flowers (Suzuki, *et al*., 2007). The Tartary buckwheat cultivar "Hokkai 10" was modified by applying a chemical mutagen to produce "Hokkai 8," a variety with high quantities of the anthocyanins cyanidin-3-O-glucoside and cyanidin-3-Orutinoside in the sprout (Kim *et al*., 2014).

2.4.4.7 Other Phenolic Compounds

During crop seed germination, nutritional quality and functioning improve. Buckwheat seeds contain a variety of phenolic chemicals, including phenolic acid, rutin, and C-glycosyl flavones. Over 72 hours of seed germination, the nutritional effect of buckwheat seed demonstrates high antioxidant activities as well as different chemical compositions (Zhang, *et al*., 2015).

2.4.4.8 Tatariside F

Traditional medicines have long been used to cure diseases in northeast Asian countries. Novel natural product molecules, in particular, have received attention as potential new sources for chemotherapy adjuvants or anticancer medicines with reduced toxicity. Tatariside F, a new small molecule isolated from the root of Tartary buckwheat, has a significant anticancer impact against human hepatocellular carcinoma (H22 cell line) (Peng, *et al*., 2015).

2.4.4.9 Fagopyrin

Fagopyrin is a buckwheat plant naturally occurring chemical. Fagopyrin's chemical structure is similar to hypericin's naphthodianthrone skeleton, which induces the phototoxic action in humans. Fagopyrin is a red-colored anthraquinone derivative found in the cotyledon of buckwheat (Kreft *et al*., 2013).

2.4.4.10 Inositol derivatives

Fagopyritols are d-chiro-inositol adduct derivatives with one to three galactosyl adducts that accumulate in buckwheat seed. Fagopyritol is utilized as a food supplement and is vital for seed germination (Steadman *et al*., 2000).

2.4.5 Minerals and vitamins

Buckwheat is a good supplier of a variety of minerals. Ikeda et al. investigated the content of eight important minerals in Japanese and three Chinese common buckwheat flours (Ikeda *et* *al*., 2006). The calcium concentration was rather low, but the other seven minerals accounted for 10% to 80% of the RDA. Because of the varying fluidities of the vascular tissues in different regions of Tartary buckwheat seeds, the mineral content of each component varies, with the bran having the highest mineral concentration. The peel and seed coat have the highest concentrations of iron, zinc, manganese, copper, molybdenum, lithium, and aluminum. Furthermore, Tartary buckwheat is high in natural organic selenium, which is deficient in other cereals. Selenium can produce a "metal-selenium-protein complex" in the human body, which aids in the elimination of harmful substances and the regulation of the immune system. Common buckwheat is also high in trivalent chromium; the "glucose tolerance factor" generated in the body by chromium improves insulin action and thus glucose tolerance. The B vitamin content of common buckwheat kernels is approximately 0.78 mg/100 g, and it is high in vitamins B1, B2, and B6, as well as vitamin P, which is not present in other grains. Some fat-soluble vitamins, particularly vitamin E, are also important bioactive components in Tartary buckwheat. Tartary buckwheat kernels have a vitamin E concentration ranging from 2.0 to 45.8 mg/kg (Jiang, *et al*., 2018).

2.5 Medicinal properties of common buckwheat

Buckwheat has been found in Traditional Chinese Medicine to be useful in the treatment of diabetes, as well as having anticancer, antihypertensive, anti-inflammatory, anti-fatigue, antioxidant, hypoglycemic, cholesterol-lowering, and hepatoprotective qualities (Fabjan, *et al*., 2003). According to recent study, polysaccharides in buckwheat are the key active components responsible for its varied pharmacological actions. Polysaccharides extracted from buckwheat have been extensively researched for their hypolipidemic, anticancer, immunoregulatory, antioxidant, and neuroprotective properties (Cho, *et al*., 2015), as well as other health advantages.

2.5.1 Antioxidant activity

Natural products are a promising source of antioxidants, and many bioactive elements of plants, fungi, and animals, particularly polysaccharides, have antioxidant properties (Zhang *et al*., 2018). Buckwheat polysaccharides have been shown to have significant antioxidant activity by a combination of in vitro experiments that assessed their ability to scavenge 2,2-diphenyl-1-picrylhydrazyl (DPPH), hydroxyl and superoxide radicals, reducing power, and ability to chelate ferrous ions (*Nam, et al., 2017).*

2.5.2 Immunomodulatory activity and antitumor activity

Anti-tumor medications with low toxicity and good efficacy have been the focus of modern biomedicine. Bioactive polysaccharides derived from medicinal plants and other natural resources have been shown to be nontoxic and free of negative effects (Liu et al., 2008); (Ramberg *et al*., 2010). Natural polysaccharides' most important biological functions include anticancer and immunoregulatory effects. The anticancer effects are most likely due to their ability to operate as immune modulators in vivo, resulting in an indirect effect on tumor resistance by boosting the body's immunological function (Meng *et al*., 2016). In immuno deficient mice, a low dose of golden buckwheat polysaccharides (50 mg/kg) greatly improved the thymus index, spleen index, phagocytic index, phagocytic count, half hemolytic value, and toe thickness. A medium dose of polysaccharides (100 mg/kg) repaired thymus and spleen immune function, boosted nonspecific immunity, improved specific humoral and cellular immune function, and improved overall immune function (Gu *et al*., 2015); (Yan, *et al*., 2011). Although buckwheat polysaccharides did not significantly prevent tumor growth in S180 sarcoma-bearing mice, they did extend survival time, boost survival rate, and improve mouse quality of life. Buckwheat polysaccharides inhibited tumor cell proliferation in vitro by accelerating apoptosis, the mechanism of which may involve increased antioxidant enzyme activity, a decrease in free radical levels, and an improvement in immunological function.

2.5.3 Hypoglycemic activity and hypolipidemic activity

Buckwheat has been demonstrated to possess anti-diabetic polysaccharides that reduce blood sugar by blocking -glucosidase (Wang, *et al*., 2016). Buckwheat polysaccharides, according to Zhu *et al*., can impair pancreatic lipase catalytic activity by modifying its shape (Zhu *et al*., 2015). Buckwheat polysaccharides may thus be a source of pancreatic lipase inhibitors, which could help with obesity treatment.

2.5.4 Inhibition of human complement

Using a microtiter plate approach, the ability of the honey samples to block human complement, triggered via the classical pathway, was assessed. 30 honey samples were serially diluted in Veronal buffer enriched with calcium and magnesium ions and treated for 30 minutes with human pooled serum as the source of complement. Sheep erythrocytes were subsequently introduced, having been sensitized by amboceptor incubation, because classical route complement activity can only be induced by particles or cells covered with immunoglobulins. Complement activation eventually leads to the creation of a membrane assault complex, which causes erythrocyte lysis. The amount of hemoglobin released, which was measured spectrophotometrically at 405 nm using an automated ELISA reader, served as a proxy for classical route complement activity (van den Berg *et al*., 2008).

2.5.5 Detoxification and slimming

Buckwheat is high in fiber and offers the benefits of eliminating heat, soothing the bowels, and moistening the intestines. Constipation can be relieved by drinking buckwheat tea for 7 to 10 days (Gao, *et al*., 2016). Buckwheat is also a fantastic appetizer and spleen; it can be used to cure gingival bleeding, mouth ulcers, and anemia. The selenium in common buckwheat, which is low in other cereals, can bond with metal in the human body to form a "metal-selenium-ovalbumin" complex, which aids in the excretion of hazardous chemicals (such as lead, mercury, cadmium, and so on) (Ikeda *et al.*, 2006). The niacin in common buckwheat boosts metabolism, improves detoxification, widens tiny blood vessels, and decreases blood cholesterol.

2.5.6 Anti-AIDS effect

An immunopotentiator was discovered in an acidic polysaccharide isolated from common buckwheat. This polysaccharide may increase immune function and modulate immunological organs in mice with subacute aging (Gao, *et al*., 2016). It has the ability to boost the body's vitality, prevent allergic reactions, and greatly raise the quantity of white blood cells in human peripheral blood. Many mineral elements that are useful to humans are found in common buckwheat, including cadmium, selenium, and zinc. According to research, increasing selenium consumption can assist boost immunological function. Quercetin, morin, and camphenanthrol, bioflavonoids found in Tartary buckwheat, have been shown in studies to have bacteriostatic and antiviral properties. Buckwheat includes a range of trace elements, including selenium, chlorophyll, dietary fiber, amino acids, and minerals, in addition to bioflavonoids and phenolic compounds. These minerals, bioflavonoids, and phenolics, can boost human immunity by providing vitality to human cells and allowing for higher resistance to diseases like AIDS (Jing, *et al*., 2016).

2.5.7 Prevention and treatment of cancer

Metal ions can be complexed by bioflavonoids, thereby preventing the lipid peroxidation process catalyzed by trace metal ions. It can efficiently prevent cell damage caused by lipid peroxidation, potentially avoiding or inhibiting cancer's effects, because it is an antioxidant and a free radical quencher. Buckwheat contains cadmium, selenium, zinc, and other mineral elements that are healthy to the human body and can boost immunity and anticancer activity. Proliferation of leukemia cells is inhibited by common buckwheat protease inhibitors (Wang *et al*., 2015). Protein isolated from Tartary buckwheat was administered to rats with colon cancers generated by 1, 2-dimethylhydrazine; the protein reduced colon cancer incidence by 47% through a significant reduction in cell proliferation (p.05).

2.6 Effect of nitrogen fertilizer on common buckwheat cultivation

Nitrogen fertilization and planting density are two important factors that affect the yield of common buckwheat (*Fagopyrum esculentum* M.). Youqiao2, a common buckwheat cultivar with high photosynthetic capacity and Local production is widely planted to investigate the effects of nitrogen fertilization and planting density Leaf photosynthetic characteristics, agronomic traits and grain yield in common buckwheat by a split plot Design main plots were assigned three nitrogen fertilizer rates: 0, 45, and 90 kg ha^{-1}, and subplots Three planting densities were assigned: 60, 90 and 120 plants/m^2 (Ep and Koin, 2001).The results showed that the grain yield was Significantly and positively related to net photosynthesis rate (Pn), stomatal conductance (Gs), transpiration rate (Tr), stomatal limitation value (Ls), chlorophyll content (SPAD value), leaf area index (LAI), vegetation. height, stem diameter, number of branches, number of internodes, number of grains per plant and 1000-grain weight, while significantly and negatively correlated with intracellular CO2 concentration (Ci) and water-use efficiency (WUE). Pn, Gs, Tr, Ls, SPAD, LAI, grain yield, stem diameter, branch number, internode number, number per grain plant, and 1000-grain weight increased and then decreased with nitrogen fertilization and planting growth density, and their maximum value is nitrogen fertilization and planting density of 45 kg ha^{-1} 90 plants/m^2 treatment. Ci and WUE decrease and then increase with increasing nitrogen fertilization and planting density, and their minimum values are observed at nitrogen fertilization and planting of 45 kg ha^{-1} .90 plant density m^2 treatments. Plant height increases with increasing nitrogen fertilization Decreased with increasing planting density. These results suggest nitrogen fertilization and planting. Density had significant effects on leaf photosynthetic capacity, agronomic traits and general grain yield. buckwheat, and a nitrogen fertilization of 45 kg ha^{-1} and a planting density of 90 plants/m^2 is Recommended for planting common buckwheat. Nitrogen fertilizer and planting density had significant effects on the leaf photosynthetic capacity, agronomic traits, and grain yield of common buckwheat. The leaf photosynthetic capacity, agronomic traits (except plant height), and grain yield increased and then decreased with the increase of nitrogen fertilizer and planting density, and the combination of nitrogen fertilization of 45 kg ha^{-1} and planting density of 90 plants/ m^2 is recommended for common buckwheat planting. Nitrogen is an essential nutrient for tartary buckwheat (*Fagopyrum tataricum* (L.) Guyrton.) because this substance Tartary affects the yield and quality of buckwheat. Physicochemical properties of starch represent important parameters of tartary buckwheat quality. Effects of nitrogen application at different rates at different times levels (0, 45, 135, and 225 kg/ha in 2015 and 0, 90, 180, and 270 kg/ha in 2017) were studied on the physicochemical properties of tartary buckwheat starch. Amylose content, particle size and its reversibility Tatary buckwheat starch containing nitrogen was lower than that without nitrogen. structure complexity, The pasting temperature, gelatinization enthalpy, relative crystallinity, light transmittance and solubility of the former were higher than those of the latter.(Halbrecq e*t al*., 2005). Nitrogen application did not change the 'A'-type crystal pattern Tartary Buckwheat Starch. This study indicated that interaction between nitrogen level and year and nitrogen. Fertilizer levels and years significantly affect the physicochemical properties of Tartary buckwheat starch. It also provides information on management of fertilization conditions to obtain these integrated results Starches with special properties for applications in the food or non-food industry (Taylor *et al*., 2010). The productivity of buckwheat was just slightly affected by the N rate. Joshi and Rana (1995) reported a positive yield response at a N fertilization rate of 50 kg ha^{-1} and Baburková *et al*. (2000) found a yield increase with N rates up to 60 kg ha^{-1}. The lack of response might have been caused by the high degree of grain scattering, that occurred during the maturation of buckwheat as a consequence of heavy rainfalls. Obviously, these losses happened predominantly in the fertilized plots, in which lodging was an increasing problem at higher rates of N supply. The decline in harvest index at high N rates, as well as the N losses, that seemed to have occurred shortly before maturity (data not shown), also support this assumption. Grain scattering as a major problem of buckwheat grain production was also stressed by Aufhammer *et al*. (1994) and Lee *et al*. (1996). The absence of an enhancement of grain yield with N fertilization is due to both, an only very slight increase in N uptake and a decreasing NUtE because of a lowered harvest index at high N rates. N uptake as well as harvest index might be enhanced through genotypes less prone to seed scattering and lodging in order to achieve a higher grain yield and NUE (Siddiqui *et al*, 2017; Erley *et al*., 2005). Nitrogen (N) fertilization affects grain quality in common buckwheat (*Fagopyrum esculentum* Moench). But the effects of N fertilizer on various buckwheat protein parameters are not fully understood. This study aimed to investigate the synthesis, accumulation, and quality of buckwheat protein under four N application rates in the Loess Plateau, China (Li *et al*., 2014). Optimal N application (180 kg

N/ha) improved yield, agronomic traits, and N transport and increased protein yield and protein component accumulation. Prolamin and glutelin accumulation first increased and then decreased with increasing N application. The relationships between the contents of protein components and the amount of applied N generally followed quadratic functions. Nitrate reductase and glutamine synthetase activities first increased and then decreased with increasing N levels. Optimal N fertilizer increased the water holding capacity and thermal stability of buckwheat protein and reduced its emulsification capacity, but negligibly changed its oil-absorption capacity. Hydrophobic amino acids and glutelin content were the main factors affecting protein quality. Crop Science Society of China and Institute of Crop Science, CAAS. Production and hosting by Elsevier B.V. on behalf of KeAi Communications Co., Ltd. This is an open access article under the CC BY-NC (Li *et al*., 2014). Optimal N fertilization markedly improved yield, agronomic traits, and N transport in common buckwheat and increased protein yield and accumulation of protein components. Albumin content increased with grain filling. Prolamin and glutelin accumulation first increased and then decreased with increasing N application. The relationship between the content of protein components and the amount of applied N generally followed a quadratic function. NR and GS activities both increased at first and then decreased with increases in N level. Maximal values were obtained with the N_1 (90 kg N/ha) and N_2 (180 kg N/ha) treatments. Moderate application of N fertilizer increased the WHC and thermal stability of the protein and reduced its emulsification capacity, without affecting the OAC of the protein. HAA and glutelin contents were the main factors affecting protein quality. Overall, an N fertilizer application of 180 kg N/ha was most beneficial to common buckwheat protein quality in the northern area of the Loess Plateau (Rasel *et al*., 2019).

Buckwheat is an important winter season crop for hilly areas but is cultivated in rainfed marginal lands and has low productivity. There is a need to increase its yield through better cultivation practices with scientific know-how. To obtain higher yield under rainfed conditions, the crop needs priming with water for 18 hr and manage the nitrogen at the rate of 30-45 kg/ha. Excess nitrogen causes lower grain yield even though the biomass yield is higher. The phosphorus consumption rate was interestingly higher in buckwheat plots with nitrogen application. Moderate consumption of potassium was also observed while nitrogen content in the soil remains apparently unchanged and a similar case for soil reaction and organic matter content. The application of phosphatic fertilizer would be beneficial for buckwheat (Rasel et al., 2019). To evaluate the effect of different levels of nitrogen on growth and yield of wheat a field experiment was conducted in 2017-18 at research area of cereals and pulses section, Ayyub agricultural research institute, Faisalabad. Eight levels of nitrogen i.e. 0, 29, 58, 87, 116, 145, 174, 203 kg ha-1 were evaluated. Experiment was laid out under randomized complete block design (RCBD) with three replications with a net plot size of 10×5m. Data were recorded for growth and yield parameters like number of tillers, plant height, spikelets per spike, seeds per spike, biological yield, 1000 grain weight, grain yield and harvest index. Different levels of nitrogen significantly increased all the growth and yield parameters. Maximum number of tillers, highest plant height and biological yield was recorded from the treatment where nitrogen was applied 203 Kg ha-1 while 1000 grain yield, seeds per spike and grain yield was achieved highest from where nitrogen applied of 145 Kg ha-1(Ali *et al*., 2018).

2.7 Effect of Phosphorus fertilizer on common buckwheat cultivation

The performance of common buckwheat (*Fagopyrum esculentum* Moench) was evaluated using various nitrogen and phosphorus levels at Mountain Agricultural Research Sub-Station, Skardu (Baltistan), Pakistan during summer 2009. Three N levels (0, 50 and 100 kg ha-1) and four P levels (0, 40, 80 and 120 kg ha-1) were used. The experiment was conducted in randomized complete block design using four replications. NP combinations were allotted to plots of size 1.8 m x 5 m, having 6 rows with row to row spacing 0.3 m. Statistical analysis of the data revealed that higher N doses delayed maturity and increased lodging while the higher P doses hastened maturity and decreased lodging. Generally, plant height, branches plant-1, heads m-2, grains head-1, 1000-grain weight, grain yield, biological yield and harvest index increased with increase in N and P levels. The interaction of nitrogen and phosphorus was significant for lodging score, 1000-grain weight, grain yield and biological yield. It was observed that higher doses of nitrogen and phosphorus (100-120 kg ha-1, respectively) used in combination enhanced yield and all yield components of common buckwheat. It is concluded that buckwheat crop, the NP requirements of which are generally said to be very low, shows active response to NP fertilization. Two experiments were carried out in a growth chamber to investigate the phosphorus (P)-uptake efficiency of *Fagopyrum esculentum* Moench (buckwheat) and *Triticum aestivum* (spring wheat) from a Ca-bound form. The first experiment was based on a sand-culture system with either rock phosphate (RP) or $CaHPO_4$ (CaHP) as the P source and nitrate or ammonium nitrate as nitrogen source. A highly calcareous soil was used in the second experiment. Buckwheat was shown to be highly efficient in taking up Ca-bound P compared to spring wheat. When plants were supplied with nitrate, the total P uptake by buckwheat from RP was nearly 10-fold higher than that of spring wheat (20.1 compared with 2.1 mg P pot−1). Changing nitrogen source from nitrate only to ammonium nitrate increased P uptake by spring wheat substantially, but not buckwheat. High P-uptake efficiency of buckwheat was also demonstrated using the field soil, but to a lesser extent, which may be related to the difference in Zn supply between sand culture and field soil. It is suggested that buckwheat may be included in intercropping or crop rotation systems to activate P sources in calcareous soils. The principal mechanism of P uptake efficiency of buckwheat may be its ability to acidify the rhizosphere; however, further study is needed to unravel the regulation of root excretion of H+ and its molecular basis in order to exploit buckwheat's genetic capability to utilize sparingly soluble P from soil (Peñuelas, 2021). Phosphorus must be available for crop uptake very early in growth because it is needed by the crop from the first stages of germination for energy reactions, cell division and growth. Phosphorus deficiency early in the growing season can reduce crop productivity more than P restrictions later in the season. Therefore, effective 4R management must provide an adequate amount of P in an available form when and where the plant can access it early in the growing season. Early in the growing season, the roots of the young seedling are small and can explore only a small amount of soil. This is especially true for spring-planted crops in the Northern Great Plains, where cold soil conditions during the early spring can slow root growth. Cold soils will also reduce the solubility and mobility of soil P. The combination of lower P availability and reduced root growth will limit the plant's ability to take up P from the soil when temperatures are low and increase the need for placement of fertilizer P in or near the seed-row. The optimum timing and placement of P fertilizer are strongly interconnected. The ability of the plant to access fertilizer P early in the growing season will be improved by placing the fertilizer in a position where the roots will contact it soon after germination. Phosphorus is relatively immobile in soil and will remain close to the site of application. Placing the fertilizer in or near the seed-row puts the P in a position where the plant root will contact the fertilizer reaction zone early in growth. Placing the fertilizer in a concentrated band will reduce the contact between the soil and the fertilizer, reducing retention and keeping the fertilizer in an available form for longer in the season. Many crops can increase root density when they contact an area of high P concentration such as a fertilizer band, increasing the ability to take up the nutrient. For crops with the ability to proliferate their roots in the band, a high proportion of the P they accumulate early in growth will come from a fertilizer band. Later in the season, as the plant roots grow, a greater proportion of the P that the plant takes up will come from the bulk soil. Placing the P in or near the seed-row at planting may be especially important for crops that have limited early-season root development, such as flax. However, soluble fertilizer may lead to seedling damage if excess amounts are placed in or too near to the seed-row. Damage is more likely with ammonia forming sources such as diammonium phosphate, as the ammonia contributes to seedling toxicity. Banding the fertilizer below or below and slightly to the side of the seed-row may reduce the risk of seedling damage in sensitive crops while maintaining the benefit of banding for early-season crop access to P. Placement in or near the seed-row at planting is most important in low-P soils where the plant cannot access enough P from the soil to meet its early-season growth requirements.(Rasel *et al*., 2019). Therefore, benefits from starter P are greatest and most frequent where soil test P concentrations are low. Reduced tillage may also increase response to P applied in or near the seed-row at planting, because soils may be slightly denser and cooler in the spring when undisturbed rather than cultivated. If soils are not extremely deficient in P, application of P as a dual band, deep-placed away from the seed-row with N fertilizer may be effective. If the soil test P concentration in the soil is high, the plant may be able to access enough P from the soil early in the season to satisfy its P demand. Building the soil P reserves through large applications of fertilizer P or manure can increase early-season and late-season P supplies and satisfy crop requirements. However, yield responses due to starter applications in or near the seed-row may still occur even when soil P is high, especially with early seeding into cold soils. An early supply of P can have long-lasting impacts of final crop yield potential, but a supplemental supply of P later in crop growth may also be important, particularly if the plant has not had the opportunity to store surplus P reserves. As the plant root system grows, it will access more P from the bulk soil and less from a fertilizer band. Uptake of P from the soil will continue during later growth stages if environmental conditions permit, and this late-season P supply may be important, depending on the initial P status of the plant. On severely depleted soils, the inability to take up adequate P later in the season may mean that maximum yield will not be obtained, even with high rates of seed-placed P. Some studies indicate that foliar applied P fertilizer may provide a benefit as a top-up treatment for wheat or corn if P from seed-placed P applications or uptake from the soil is severely restricted because of moisture stress or low soil P levels. However, benefits of foliar application appear to be rare under conditions experienced on the Northern Great Plains. In summary, under cold soil conditions as are often experienced in the Northern Great Plains during early plant growth, plant access to soil P tends to be reduced because of slower diffusion, less root growth, and lower availability of native soil P. Under these conditions, fertilizer P may be more necessary to ensure adequate crop growth and may be more available for crop uptake because of slower retention reactions. Band application in or near the seed-row at planting will .place the fertilizer in a position where the crop can access it early in the season and when it is required for optimum yield (Taylor *et al*., 2010).

2.8 Effect of potassium fertilizer on common buckwheat cultivation

Potassium, mostly as a cation (K^+), together with calcium (Ca^{2+}) are the most abundant inorganic chemicals in plant cellular media, but they are rarely discussed. K^+ is not a component of molecular or macromolecular plant structures, thus it is more difficult to link it to concrete metabolic pathways than nitrogen or phosphorus. Over the last two decades, many studies have reported on the role of K^+ in several physiological functions, including controlling cellular growth and wood formation, xylem–phloem water content and movement, nutrient and metabolite transport, and stress responses.(Rasel *et al*., 2019) In this paper, we present an overview of contemporary findings associating K^+ with various plant functions, emphasizing plant-mediated responses to environmental abiotic and biotic shifts and stresses by controlling transmembrane potentials and water, nutrient, and metabolite transport. These essential roles of K^+ account for its high concentrations in the most active plant organs, such as leaves, and are consistent with the increasing number of ecological and agricultural studies that report K^+ as a key element in the function and structure of terrestrial ecosystems, crop production, and global food security. We synthesized these roles from an integrated perspective, considering the metabolic and physiological functions of individual plants and their complex roles in terrestrial ecosystem functions and food security within the current context of ongoing global change. Thus, we provide a bridge between studies of K^+ at the plant and ecological levels to ultimately claim that K+ should be considered at least at a level similar to N and P in terrestrial ecological studies. (Sardans and Peñuelas, 2021). The global tea (*Camellia sinensis* L.) output has been in decline despite the continued increases in global tea consumption amount and cultivation area. Application of potassium (K) fertilizer is a common way to enhance tea yield and quality, given the widespread deficiency of soil-available K in tea plantations. However, the specific effects of K fertilizer application upon tea yield and quality and its differential effects among varied K fertilizer application amount, soil-available K amount and tea types remain unknown. In this study, 518 pairwise comparisons between tea productivity after non-application and application of K fertilizers were obtained through a literature review, and then analyzed. The results showed that K application had a significant positive effect on tea yield and overall quality, enhancing the amount of tea yield, the content of amino acid, tea polyphenol, water extract, and catechin by 7.83 %, 7.84 %, 26.79 %, 1.57 % and 6.47 %, respectively. Furthermore, a K application rate of 90–120 kg ha^{-1} maximized the tea yield and quality. However, K application showed varied effects on tea yield and quality in soils with different available K levels. In terms of tea types, the yield and quality of black and green tea were enhanced under K application. The results of a mixed-effects model indicated that the main factors affecting tea quality were the amount of K fertilizer applied and the soil-available K content, whereas the variation in tea yield results mainly from the tea types. Therefore, to develop high-quality tea plantations and address the contradiction between low tea yield and high tea consumption, efforts should be made to enhance the soil-available K content by optimizing the amount of K applied on tea plantations. Many un-updated reports indicated that potassium was not deficient in Ethiopian soils. However, it was later proved that many Ethiopian soils are potassium deficient. Hence, the Ethiopian Soil Information System (EthioSIS) has initiated potassium fertilizer demonstrations in 2014 using K containing blended fertilizers in different parts of the country. But there were no evidences about the K in the blended fertilizer is enough for wheat demand or not. Thus, a field experiment was conducted to evaluate the response of wheat to additional K rates on top of the K containing blended fertilizers. The experiments were laid out in Randomized Complete Block Design with 4 levels of potassium (0, 30, 60, 90). Data on yield and yield components of wheat crop were collected and analysis of variance was done. Results depicted that plant height and harvest index were not significant. However, spike length, grain yield and straw yields of wheat were significantly affected by K application rates. Hence, the highest spike length was obtained at a rate of 90 kg/ha K_2O but the highest grain and straw yield of wheat were obtained at 30 kg/ha K_2O. Besides, the highest apparent K recovery and agronomic use efficiency were found at 30 kg/ha (Hara *et al*., 2017).

CHAPTER III

METHODOLOGY

3.1 Study area

The present study was conducted in the research field of the Hajee Mohammad Danesh Science and Technology University. It was carried out from December 2022 to April 2023.

Figure 3.1: Studies area of the research field

3.2 Plant materials

This study estimated from the G_{18} common buckwheat germplasm center of the Department of Biochemistry and Molecular Biology, HSTU, Dinajpur. This germplasm was collected from Lalmonirhat district of the Rangpur division of Bangladesh. During the winter season, the field experiments were conducted at HSTU central research field Dinajpur (25^0 38'N latitude and 88^0 38'E longitude with an altitude of 40 meters above sea level) in the year December 2022 to April 2023 under rain-fed conditions.

3.3 Experimental design and layout

The experiment followed a randomized complete block design (RCBD) with Fifteen plots. The unit plot size for each landraces was 2 m × 4 m (8 m^2), and there was a 1 m space between the landraces and a 0.7 m space between individual plots.

3.4 Climate

The weather in the study's area is humid subtropical, with the following seasons: Kharif-1 (April-June), Kharif-2 (July-September), and Robi (October-March). This research was carried out throughout the Robi season. Because of the lesser rainfall, this weather condition is favorable for crop development. Weather reports on humidity, temperature, and rainfall were collected from the Bangladesh Weather Forecast Department of Dhaka throughout the time of the study. During this time period, the average maximum temperature was 27.24 °C, and the average lowest temperature was 8 °C. These details are presented in Appendix II.

3.5 Field Preparation and Seed Sowing

Field was cultivated four times by tractor to make the soil loose. The field was fertilized with cow dung, TSP, Urea, MOP. Cow dung, TSP, MOP, Urea fertilizers were applied in each plot at the rates 10 ton/ha, 110kg/ha, 5kg/ha and 6kg/ha respectively. All the fertilizers excluding urea were applied with soil during the final land preparation. The urea was applied in the field at 15 and 30 days after sowing. Seeds were randomly sown on 14 December 2022. The seed rate was 30 g/ $4m^2$.

Figure 3.2: Field Preparation

Figure 3.3: Seed sowing

3.6 Intercultural activities

Thinning and weeding were performed as needed for the crop's optimal growth and development. Throughout the growing season, four irrigations were performed. The covering was also accomplished by cracking the soil crust following sufficient irrigation. Irrigation was administered to keep the seedlings moist during the early stages of establishment, and subsequently irrigated as needed through the growing phase. During the reproductive period, no water deficit was seen.

3.7 Application of different Fertilizer

There were five treatments used for the determination of NPK effects on plant growth, yield and antioxidant enzyme activity of common buckwheat cultivation. The different doses of fertilizer were applied in seed sewing, seedling stage, early flowering stage and flowers fertilization stage. The different doses of NPK fertilizers are given in table 1.

Table 3.1: Different doses of fertilizer used in soil

Treatment	Urea ($g/8m^2$)	TSP ($g/8m^2$)	MOP ($g/8m^2$)
T_0	0	0	0
T_1	36	52	12
T_2	44	60	20
T_3	52	68	28
T_4	60	76	36

3.8 Treatments used for buckwheat cultivation

Fifth treatments of inducing fertilizer were tried:

1. No fertilizer used (T_0)
2. Urea fertilizer used 36g, TSP fertilizer used 52g, MOP fertilizer used 12g (T_1)
3. Urea fertilizer used 44g, TSP fertilizer used 60g, MOP fertilizer used 20g (T_2)
4. Urea fertilizer used 52g, TSP fertilizer used 68g, MOP fertilizer used 28g (T_3)
5. Urea fertilizer used 60g, TSP fertilizer used 76g, MOP fertilizer used 36g (T_4)

3.9 List of Morphological data

1. Plant height (cm)
2. Distance of first node (cm)
3. Number of Branch
4. Number of florescence
5. Raceme length (cm)
6. Number of flowers per florescence
7. Grain weight
8. 1000 grain weight

3.10 Antioxidant enzyme assay

Leaf sample were collected from control and treated groups. Leaf samples were homogenized at 4^0C in 50 mM potassium phosphate buffer (pH 8.0), containing 5mM cysteine and 1% (w/v) polyvinylpyrrolidone. The homogenized mixture was filtered and centrifuged at 15,000×g for 20 minutes, and supernatant will be used for antioxidant enzyme assay.

3.10.1 Determination of catalase activity

Catalase (EC: 1.11.1.6) activity was determined as described previously (Aebi, 1984) with some modifications. Exactly 0.7 ml of 50 mM potassium phosphate buffer (pH 8.0) was taken in a cuvette and 0.1 ml of EDTA and 0.1 ml of H_2O_2 was added and mixed well. In all stage 4^0C temperature was maintained. Reaction was started by adding 0.1 ml of enzyme extract and changes in absorbance was recorded immediately at 240 nm at 30 seconds interval for two minutes. The activity of catalase was calculated from the decrease in absorbance per minute when the extinction coefficient of H_2O_2 was 40 $M^{-1}cm^{-1}$. The enzyme activity was calculated by the following formula-

$$\text{CAT (mMoleg}^{-1}\text{FW)} = \frac{\text{(Absorbance difference /min)} \times \text{Dilution factor (DF)}}{40}$$

3.10.2 Determination of peroxidase activity

Peroxidase (EC: 1.11.1.6) activity was determined as described previously (Siddiqui *et al.*, 2017) In brief, about 0.6 ml of 50 mM potassium phosphate buffer (pH 8.0) was taken in cuvette and subsequently 0.1 ml of EDTA, 0.1 ml of H_2O_2 and 0.1 ml of guaiacol was added and mixed well. Reaction was started by adding 0.1 ml of enzyme extract and changes in absorbance was recorded immediately at 470 nm at 30 seconds interval for two minutes. The activity of peroxidase was calculated from the increase in absorbance per minute when the extinction coefficient of guaiacol is 26.6 $mM^{-1}cm^{-1}$. The enzyme activity was calculated by the following formula-

$$\text{POD (µMoleg}^{-1}\text{FW)} = \frac{\text{(Absorbance difference/ min} \times \text{DF}}{26.6}$$

3.10.3 Determination of ascorbate peroxidase activity

Ascorbate peroxidase (EC: 1.11.1.6) activity was determined by following the method of Nakano and Asada (1981) with some modification. About 0.6 ml of 50 mM potassium phosphate buffer (pH 8.0) was added in cuvette with 0.1 ml of EDTA, 0.1 ml of H_2O_2 and 0.1 ml of ascorbate and mixed well. Reaction was started by adding 0.1 ml of enzyme extract and changes in absorbance was recorded immediately at 290 nm at 30 seconds interval for two minutes. The activity of ascorbate peroxidase was calculated from the decrease in absorbance per minute when the extinction coefficient of ascorbate is 2.8 $mM^{-1}cm^{-1}$. The enzyme activity was calculated by the following formula-

$$APX\ ((Ug^{-1}FW) = \frac{(Absorbance difference / \min) \times DF}{2.8}$$

3.10.4 Determination of Hydrogen peroxide (H_2O_2) and Malondialdehyde (MDA) content

H_2O_2 and MDA content were determined as described previously (Rasel et al., 2020). For determination of H_2O_2 content, 0.1 g of fresh leaf sample was homogenized in a mortar with pestle using 1 ml of 0.1% tri-chloro-acetic acid (TCA) and homogenizes it at 4^0C. After centrifugation at 10,000 × g for 15 minutes, the supernatant was kept in dark for 1 h after mixing with phosphate buffer (10 mM, pH 7.0) and potassium iodide (1 M) (In the ratio 0.5 ml: 0.5 ml: 1 ml). Absorbance of the resulting solution was recorded at 390 nm. All the steps were performed at 4 °C except absorbance measurement.

3.10.5 Determination of MDA content

0.1 g leaf tissue (with similar age, and young expanded leaf) was ground into powder with liquid nitrogen, and then the powder was put into a tube containing 1 ml 0.1% (w/v) TCA and mixed by inverting the tube to homogenize the leaf tissue. It was centrifuged and homogenized the samples at 10,000×*g* for 10 min, and supernatant was transferred to a new tube. 4 ml of 20% TCA containing 0.5% TBA was added to the supernatant and mixed well. The mixture was boiled at 95 °C for 15 min and quickly cooled on ice (TBA can interact with MDA and results into red compound in acidic buffer, so the content of MDA can be calculated by measuring the density of the resulting red compound with spectrophotometer at 532 nm. The high temperature can accelerate the reaction and low temperature can inhibit it). The mixture was centrifuged at 10,000 × *g* for 5 min, and supernatant was transferred to a new tube. Absorbance of the resulting solution was recorded at 532 nm. All the steps were performed at 4 °C except absorbance measurement.

3.10.6 Determination of proline content

Proline content of wheat leaf will be determined as previously described (Bates et al., 1973). Fully expanded young leaves were collected. Purified proline was used to standardize the procedure for quantifying sample values. Acid-ninhydrin was prepared by warming 1.25 g ninhydrin in 30 ml glacial acetic acid and 20 ml 6 M phosphoric acid, with agitation, until dissolved. It was kept cool (stored at 4°C) the reagent remains stable 24 hours. Approximately 0.5g of wheat leaves were homogenized in 10 ml of 3% aqueous sulfosalicylic acid and the homogenate was filtered through Whatman No. 2 filter paper. Two ml of filtrate was reacted with 2 ml acid-ninhydrin and 2 ml of glacial acetic acid in a test tube for 1 hour at 100°C, and the reaction was terminated in an ice bath. The reaction mixture was extracted with 4 ml toluene, mixed vigorously with a test tube stirrer for 15-20 sec. The chromophore containing toluene was aspirated from the aqueous phase, warmed to room temperature and the absorbance was read at 520 nm using toluene for a blank. The proline concentration was determined from a standard curve and calculated on a fresh weight basis as follows:

[(µg proline/ml × ml toluene) / 115.5 µg/µmole]/ [(g sample)/5] = µmoles proline/g of fresh weight material.

3.11 Statistical analysis

The data were analyzed with R statistical software. The data were presented to Mean performance ANOVA.

CHAPTER IV
RESULTS

Buckwheat is one of the most popular cereals in the world. This study was conducted to evaluate the effect of NPK fertilizer applications on morpho-physiology, antioxidant enzymes activity, growth and yield of buckwheat. Each value represents the average of five soil treatments and the results are expressed as mean values ± standard deviations (SD). After performing the ANOVA (Analysis of variance) test it is evident that five morphological properties plant height, distance of first node, number of inflorescences, raceme length (cm), number of flowers per inflorescence, seed weight per plot and four antioxidant properties (peroxidase, catalase, ascorbate, H_2O_2) are significantly different ($p < 0.05$).

4.1 Plant growth and yield related variables

The plant growth and yield related characteristics was measured at the end of the experiment (Table-4.1). We used five treatments of NPK fertilizer in soil applications including T_0 (Control), Nitrogen fertilizer used T_1 (36 g/8m^2), T_2 (44g/8m^2), T_3 (52 g/8m^2), and T_4 (60 g/8m^2). Phosphorus fertilizer used T_1 (52g/8m^2), T2 (60g/8m^2), T3 (68g/8m^2), T4 (76g/8m^2). Potassium fertilizer used T_1 (12g/8m^2), T2 (20g/8m^2), T3 (28g/8m^2), T4 (36g/8m^2). The study was observed that highest plant height of Buckwheat plant after treatment by T4 nitrogen (60 g/8m^2), phosphorus (76g/8m^2), potassium (36g/8m^2) fertilizer in buckwheat with a mean and standard deviation was 39.77 cm and 3.09 cm. But the lowest plant height was observed in T_0 with a mean and standard deviation was 19.94 cm and 1.58 cm. Similarly, the highest distance of first node was observed in buckwheat after treatments by T4 nitrogen (60 g/8m^2), phosphorus (76g/8m^2), potassium (36g/8m^2) fertilizer in buckwheat with a mean and standard deviation was 3.33 cm and 0.50 cm. But the lowest distance of first node was observed in T_0 with a mean and standard deviation was 2.77 cm and 0.25 cm. On the other hand, the highest number of inflorescence was observed after treatment by T4 nitrogen (60g/8m^2), phosphorus (76g/8m^2), potassium (36g/8m^2) fertilizer in buckwheat with a mean and standard deviation was 7.10 cm and 0.77 cm. On the other hand, lowest number of inflorescences was observed in T_0 with a mean and standard deviation was 3.33 cm and 1.19 cm. On the other hand, the highest raceme length was observed after treatment by T4 nitrogen (60 g/8m^2), phosphorus (76g/8m^2), potassium (36g/8m^2) fertilizer in buckwheat with a mean and standard deviation was 5.10 cm and 0.42 cm. On the other hand, lowest raceme length was observed in T_0 with a mean and standard deviation was 2.35 cm and 0.95 cm. On the other hand, the highest number of per inflorescence was observed after treatment by T4 nitrogen ($60g/8m^2$), phosphorus ($76g/8m^2$), potassium ($36g/8m^2$), fertilizer in buckwheat with a mean and standard deviation was 33.33 cm and 1.75 cm. On the other hand, lowest number of per inflorescence was observed in T_0 with a mean and standard deviation was 16.66 cm and 6.52 cm. On the other hand, the highest number of branches was observed after treatment by T4 nitrogen ($60g/8m^2$), phosphorus ($76g/8m^2$), potassium ($36g/8m^2$), fertilizer in buckwheat with a mean and standard deviation was 6.64 cm and 0.11 cm. On the other hand, lowest number of branches was observed in T_0 with a mean and standard deviation was 3.49 cm and 0.23 cm. On the other hand, the highest per plot seed weight was observed after treatment by T4 nitrogen ($60g/8m^2$), phosphorus ($76g/8m^2$), potassium ($36g/8m^2$), fertilizer in buckwheat with a mean and standard deviation was 483.33gm and 18.14 gm. On the other hand, lowest per plot seed weight was observed in T_0 with a mean and standard deviation was 46.00 gm and 5.29 gm. On the other hand, the highest 1000 grain seed weight was observed after treatment by T4 nitrogen ($60g/8m^2$), phosphorus ($76g/8m^2$), potassium ($36g/8m^2$), fertilizer in buckwheat with a mean and standard deviation was 15.02 gm and 0.06 gm. On the other hand, lowest 1000 grain seed weight was observed in T_0 with a mean and standard deviation was 14.75 gm and 0.02 gm.

4.2 Buckwheat cultivars antioxidant and H_2O_2, MDA, Proline, levels enzyme activity

Following that, we assessed the activity of several major antioxidant enzymes involved in H_2O_2 detoxification, such as CAT, POD, MDA, Proline, and APX and these findings are given in figure 4.1-4.6. The highest peroxidase level was measured in T_4 with a mean and standard deviation was 8.20 and 5.50. The lowest peroxidase level was measured in T_0 with a mean and standard deviation was 2.60 and 1.50. The second highest peroxidase was measured in T_3 with a mean and standard deviation was 6.00 and 4.5. But the moderate peroxidase value was calculated in T_2 with a mean and standard deviation was 4.70 and 3.20. The highest ascorbate level was measured in T_4 with a mean and standard deviation was 4.26 and 0.82. But the lowest ascorbate was measured in T_4 with a mean and standard deviation was 1.163 and 1.71. The second highest ascorbate was measured in T_2 with a mean and standard deviation was 3.07 and 2.66. But the moderate peroxidase value was calculated in T_3 with a mean and standard deviation was 2.61 and 0.84. The highest catalase level was measured in T_0 with a mean and standard deviation was 0.96 and 0.47. But the lowest catalase was measured in T_2 with a mean and standard deviation was 1.57 and 0.05. The second highest catalase was measured in T_1 with a mean and standard deviation was 0.74 and

0.22. But the moderate catalase value was calculated in T_4 with a mean and standard deviation was 0.72 and 0.03. The highest hydrogen peroxide was measured in T_4 with a mean and standard deviation was 1.57 and 0.05 But the lowest hydrogen peroxide was measured in T_0 with a mean and standard deviation was 1.65 and 0.10. The highest MDA was measured in T_0 with a mean and standard deviation was 3.57 and 0.10 But the lowest hydrogen peroxide was measured in T_4 with a mean and standard deviation was 0.52 and 0.02. The highest proline was measured in T_4 with a mean and standard deviation was 2.43 and 0.22 But the lowest proline was measured in T_0 with a mean and standard deviation was 0.72 and 0.09.

Table 4.1 Effects of different doses NPK fertilizer on plant height, distance of first node, number of florescence, raceme length, number of per florescence, seed weight/plant of buckwheat cultivars

Varieties	Treatment	Plant height	Distance of first node	Number of inflorescences	Raceme length	Number of flowers per florescence	Seed weight
L18	T_o	19.94 ± 1.58b	2.77 ± 0.25a	3.33±1.19b	2.35±0.95b	16.66 ± 6.52b	46.00 ± 5.29c
	T_1	33.66 ± 6.17a	2.83± 0.50a	6.55±0.50a	4.72± 0.78a	31.00±11.45ab	134.00±30.8c
	T_2	36.10 ± 1.34a	3.44± 0.09a	6.22 ± 1.16a	4.94± 0.09a	36.66 ± 5.13a	218.00±67.91bc
	T_3	38.55 ± 4.29a	3.38± 0.25a	6.44 ± 0.50a	5.22± 0.25a	36.33 ± 4.16a	388.00 ± 162.92ab
	T_4	39.77 ± 3.09a	3.33 ± 0.00a	6.10 ± 0.77a	5.10 ± 0.42a	33.33 ± 1.75a	483.33 ± 18.14a

Varieties	Treatment	Number of flowers per inflorescence	Number of branches	Per plot Seed weight	1000 grain seed weight
L18	T_o	16.66 ± 6.52b	3.49 ± 0.23a	46.00 ± 5.29c	14.75 ± 0.02a
	T_1	31.00±11.45ab	4.46 ± 0.01b	134.00±30.8c	14.83 ± 0.03b
	T_2	36.66 ± 5.13a	5.47 ± 0.41b	218.00±67.91bc	14.89 ± 0.02bc
	T_3	36.33 ± 4.16a	5.22 ± 0.19c	388.00 ± 162.92ab	14.94 ± 0.01c
	T_4	33.33 ± 1.75a	6.64 ± 0.11cd	483.33 ± 18.14a	15.02 ± 0.06d

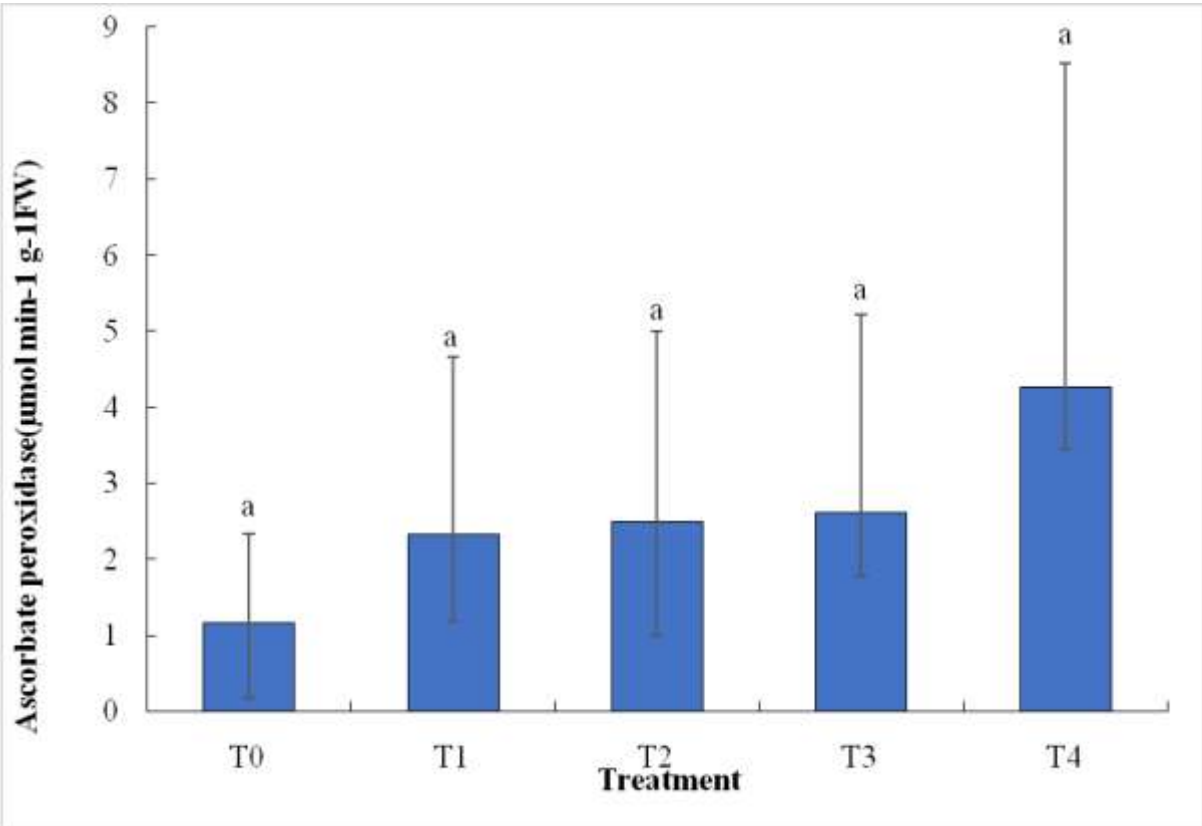

Figure 4.1 Effect of NPK on ascorbate peroxidase in different buckwheat cultivars

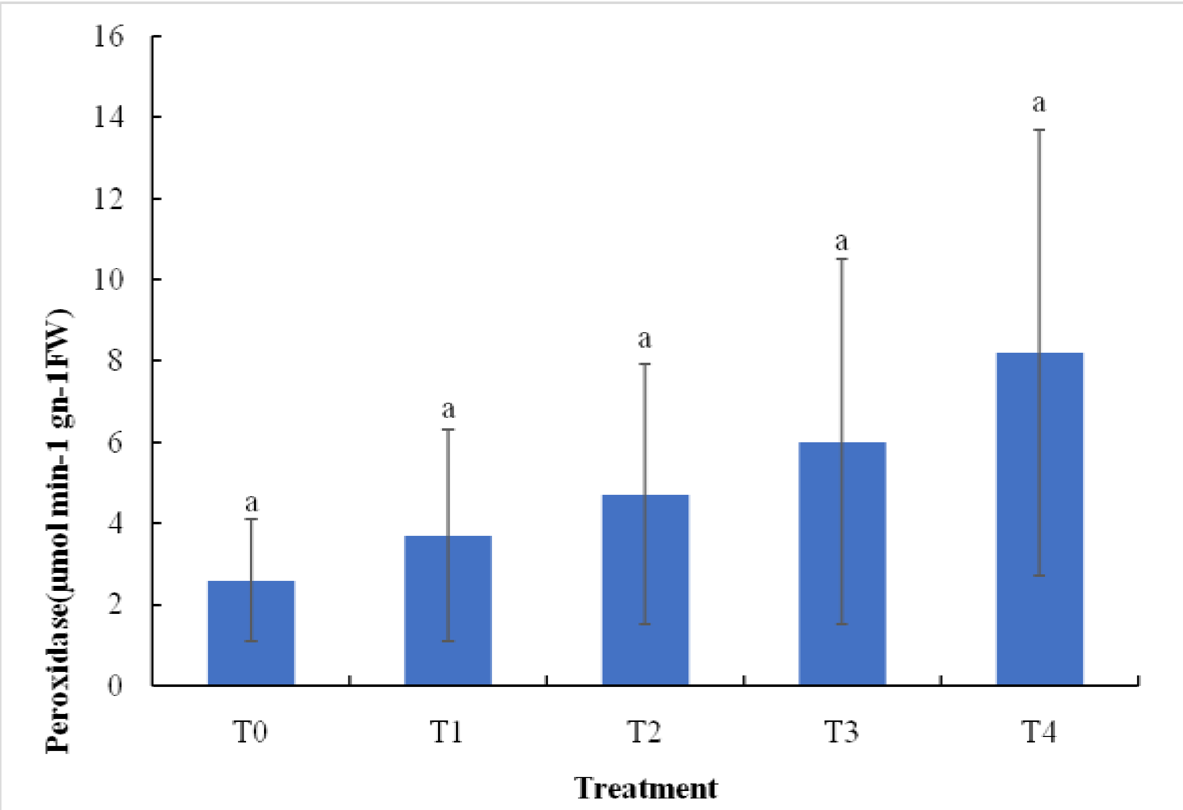

Figure 4.2 Effect of NPK on peroxidase in different buckwheat cultivars

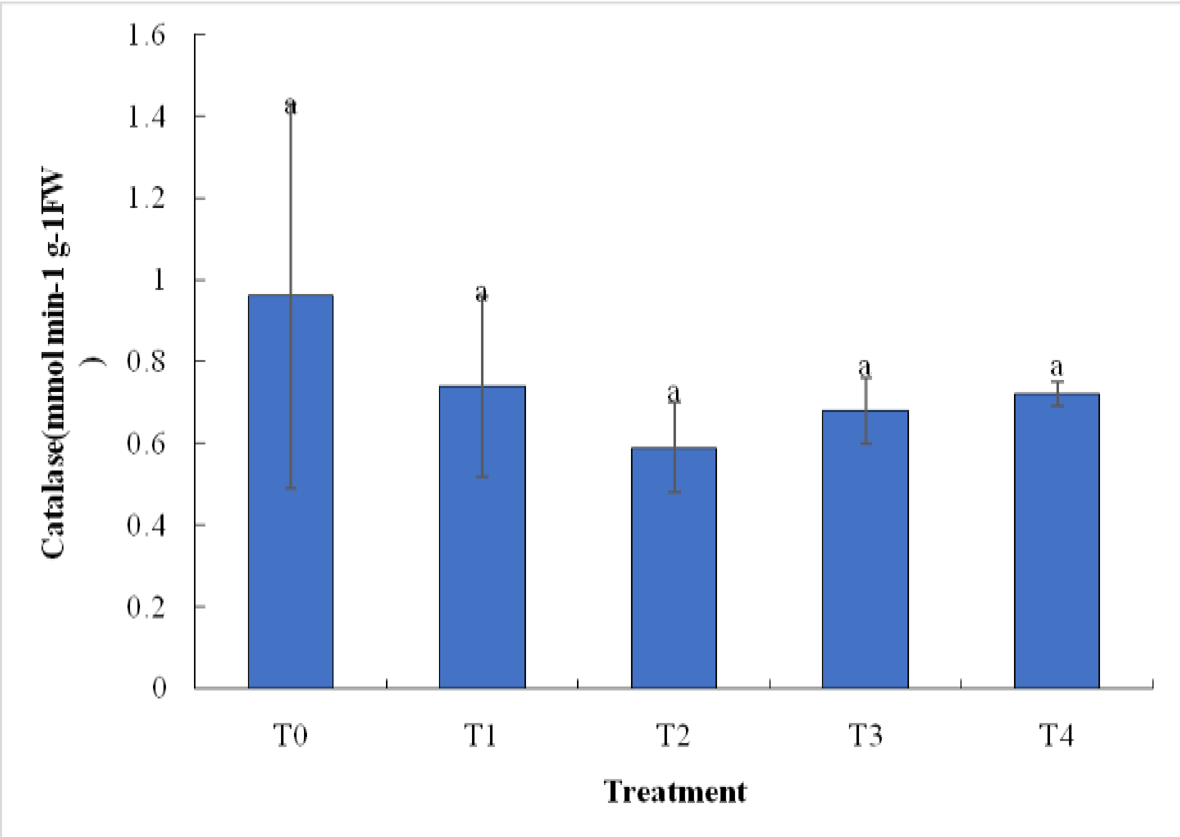

Figure 4.3 Effect of NPK on catalase in different buckwheat cultivars

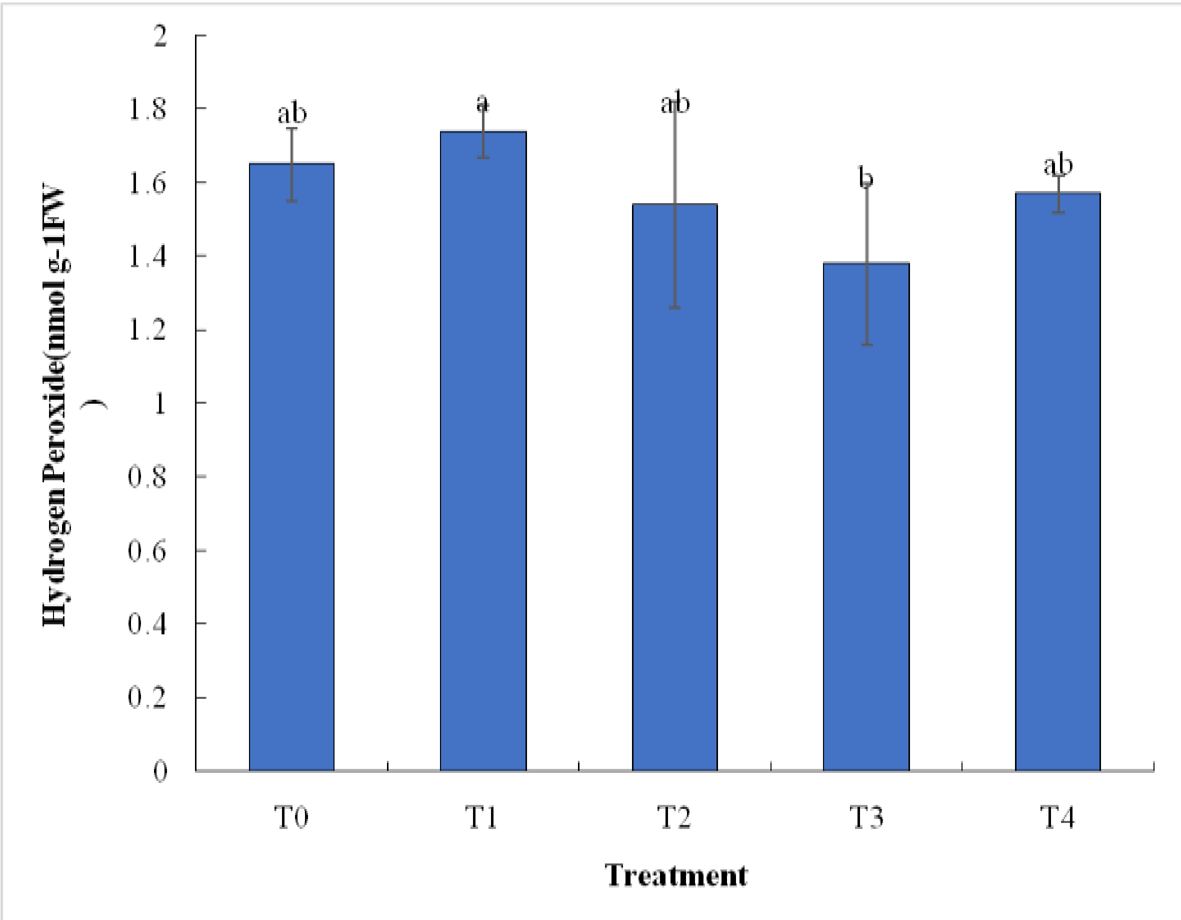

Figure 4.4 Effect of NPK on hydrogen peroxide in different buckwheat cultivars

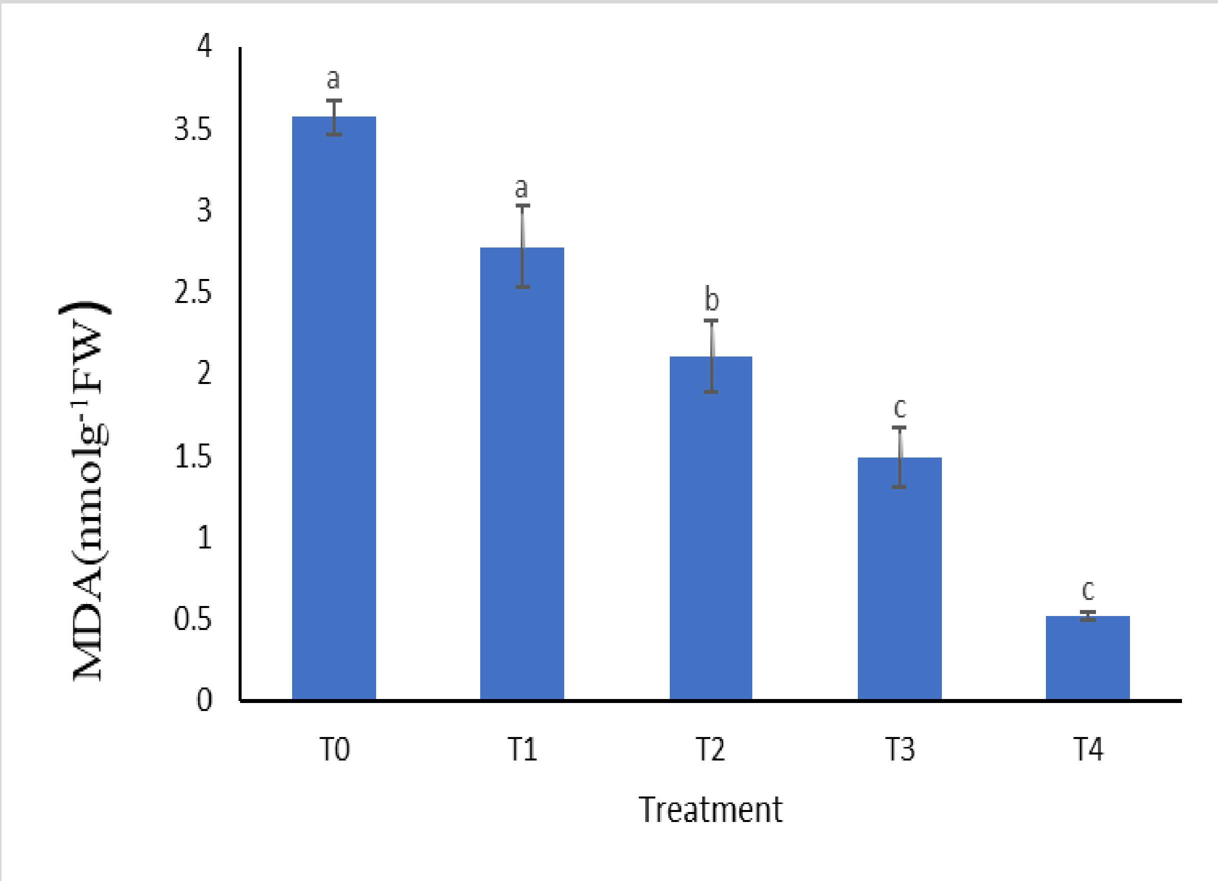

Figure 4.5 Effect of NPK on MDA in different buckwheat cultivars

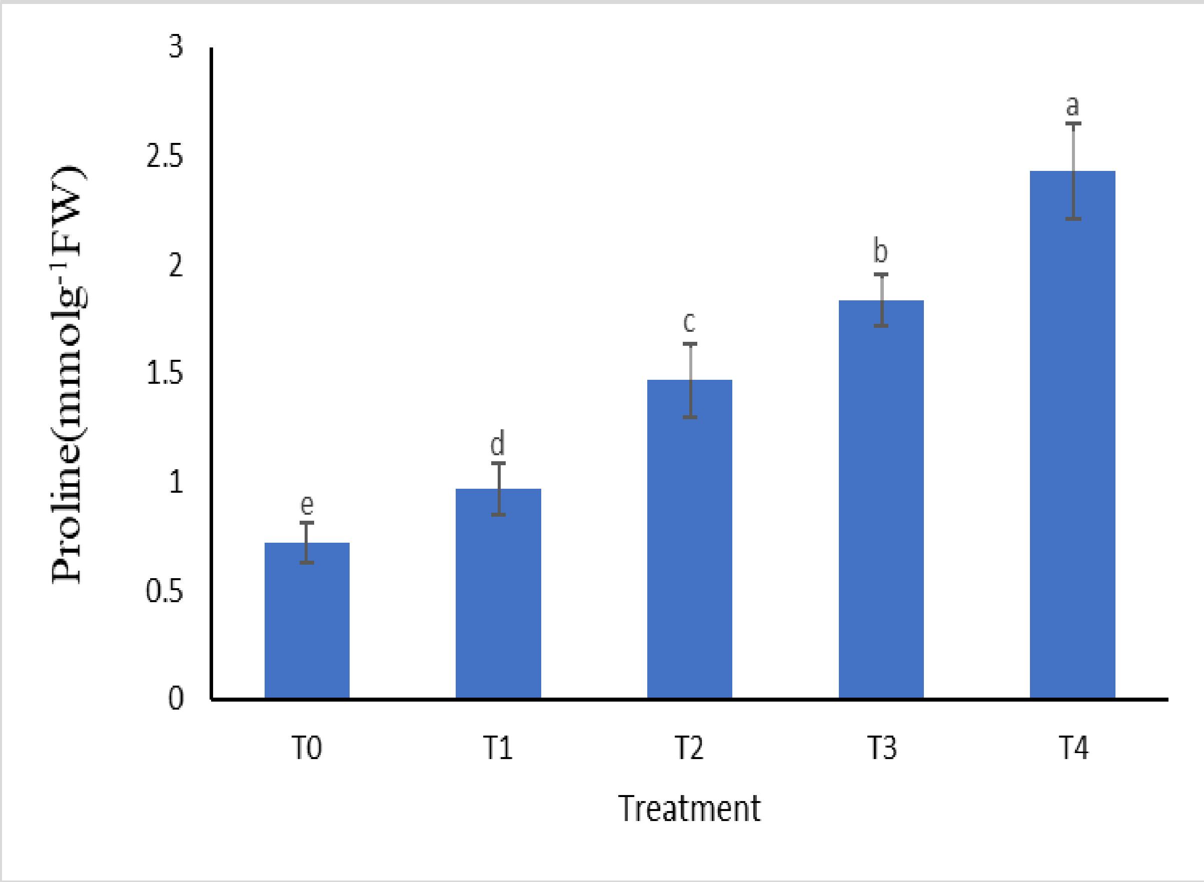

Figure 4.6 Effect of NPK on Proline in different buckwheat cultivars

CHAPTER V
DISCUSSION

Common buckwheat is a nutrient-rich pseudo cereal that is considered as suitable crop to diversify our cropping system. One of the characteristics of this smuggled crop is that it can adapt to any environment. The reason why it is called a smuggled crop is that it has many flowers but few fruits because it is a genetically modified crop. Although this crop was cultivated earlier, it was once on the verge of extinction due to a lack of knowledge about its nutritional value. But over the last 20-30 years, growing research on its nutritional value, health benefits, and other important uses (medicinal uses) has led to the revival of this crop. One of the genetic variations in the common buckwheat gene pool today is the traditional landraces that allow for the growth and development of nutrient-rich varieties (Chrungoo, 2021).

The experiment aimed to assess the impact of different NPK fertilizer treatments on the growth and yield-related characteristics of Buckwheat plants. Five treatments were applied, each varying in the amount of nitrogen (N), phosphorus (P), and potassium (K) fertilizers. The control group (T_0) received no fertilizer, while the other treatments (T_1 to T_4) were subjected to increasing amounts of N, P and K fertilizers. The measured variables included plant height, distance of the first node, number of branches, number of inflorescences, raceme length, number of flowers per florescence, per plant grain, and grain weight. The results revealed significant differences in these variables across the various fertilizer treatments. The highest plant height was observed in T_4, where nitrogen, phosphorus, and potassium were applied at rates of 75Kg/h, 95Kg/h, and 45Kg/h, respectively. This treatment resulted in a mean plant height of 39.77 cm, significantly taller than the control group (T_0) with a mean height of 19.94 cm.

Similarly, the distance of the first node, an indicator of plant development, was maximized in T_4, demonstrating the positive effect of higher fertilizer concentrations. T_4 also exhibited the highest number of florescence, raceme length, number of flowers per florescence, and seed weight, emphasizing the importance of optimal NPK levels for overall plant development and yield. These effects are similar to Rodríguez *et al*. (2020).

Conversely, the control group (T_0) consistently showed the lowest values across all measured variables, indicating the necessity of NPK fertilizers for enhancing the morphological characteristics and yield of Buckwheat. These findings suggest that the application of a specific combination of nitrogen (75Kg/h), phosphorus (95Kg/h), and potassium (45Kg/h) fertilizers (T_4) had a substantial positive impact on the growth and yield of buckwheat plants. The observed variations underscore the importance of balanced nutrient management in agricultural practices to optimize crop productivity and ensure sustainable agriculture similar to (Fesenko and Mazalov, 2020). Further research could delve into the underlying physiological mechanisms driving these observed effects and explore the long-term implications of different NPK fertilizer combinations on soil health and crop sustainability (Wang *et al.*, 2017).

The assessment of antioxidant enzyme activity, including Catalase (CAT), Peroxidase (POD) and Ascorbate Peroxidase (APX) provides crucial insights into the plant's ability to detoxify hydrogen peroxide (H_2O_2), a reactive oxygen species (ROS) implicated in various stress responses (Wang *et al.*, 2017). The findings, as presented in Table 4.1.3, reveal distinct patterns of enzyme activity across different NPK fertilizer treatments. Peroxidase, a key enzyme in H_2O_2 detoxification, exhibited the highest activity in T4, where nitrogen (75Kg/h), phosphorus (95Kg/h), and potassium (45Kg/h) were applied. This heightened peroxidase activity suggests an efficient defense mechanism against oxidative stress in plants subjected to this specific fertilizer combination. Conversely, the lowest peroxidase levels were observed in T_0, the control group, indicating a potential vulnerability to oxidative stress in the absence of NPK supplementation (Antunes *et al.*, 2002).

Ascorbate, another vital antioxidant, showed its highest level in T_4, suggesting that the plant response to oxidative stress was optimized under this specific NPK treatment. The lowest ascorbate levels were again noted in T_0, emphasizing the importance of NPK fertilization in enhancing the plant's ability to combat oxidative stress. Catalase activity, which plays a crucial role in decomposing hydrogen peroxide into water and oxygen, exhibited the highest level in T_0, the control group. This unexpected result may indicate a compensatory mechanism in the absence of additional NPK supplementation, emphasizing the adaptability of plants to varying nutrient conditions (Vazhov *et al.*, 2013). Hydrogen peroxide levels were highest in T_4, aligning with the elevated peroxidase activity observed in the same treatment. This suggests a coordinated response, where increased peroxidase activity effectively manages the higher levels of hydrogen peroxide, further emphasizing the positive impact of the specific NPK treatment in T_4 on the plant's oxidative stress tolerance (Saleem *et al.*, 2020).

In summary, the observed variations in antioxidant enzyme activity and hydrogen peroxide levels across different NPK fertilizer treatments underscore the intricate interplay between nutrient availability and the plant's antioxidative defense mechanisms. The results support the notion that optimal nutrient supplementation, as exemplified by the specific NPK combination in T_4, can enhance a plant's resilience to oxidative stress, contributing to improved overall growth and yield. Further investigations into the underlying molecular pathways and interactions involved in these responses would provide deeper insights into the mechanisms underlying the observed effects.

The highest MDA level was observed in T0, indicating a relatively elevated level of this parameter at the initial time point. The mean and standard deviation values (3.57 and 0.10, respectively) suggest a consistent and precise measurement of MDA at T0. The highest proline concentration was recorded at T4, implying a possible accumulation or increase in proline content throughout the experiment. The mean and standard deviation values (2.43 and 0.22, respectively) at T4 indicate a considerable variation in proline levels, suggesting potential biological variability or responses to external factors. The observed differences between T0 and T4 in MDA and proline concentrations highlight dynamic changes in these parameters over the experimental period. Correlations or trends between these measurements could provide insights into the interplay of these biochemical components within the system under study. Elevated MDA levels at T0 may indicate an initial heightened activity or response in the system, which could have downstream effects on other biochemical processes (Savika and Škute, 2010). The decrease in hydrogen peroxide coupled with an increase in proline at T4 might suggest a shift in cellular redox balance or stress response mechanisms (Yoshiba *et al.*, 1997). It's crucial to consider potential confounding variables or experimental conditions that may have influenced the observed changes. Further investigations, such as pathway analysis or additional time points, could provide a more comprehensive understanding of the underlying biological processes.

CHAPTER VI
SUMMARY AND CONCLUSIONS

In conclusion, this study focused on the impact of various NPK fertilizer treatments on the growth and yield-related characteristics of common buckwheat, a nutrient-rich pseudo-cereal known for its adaptability to diverse environments. The experimental results demonstrated significant variations in key variables across different fertilizer treatments, highlighting the importance of nutrient management in optimizing crop productivity. The highest plant height, an essential indicator of overall plant growth, was achieved with the specific combination of nitrogen (75Kg/h), phosphorus (95Kg/h), and potassium (45Kg/h) in Treatment four (T_4). This treatment resulted in a remarkable mean plant height of 39.77 cm, underscoring the positive impact of balanced NPK fertilization on the developmental aspects of buckwheat. Treatment four (T_4) also excelled in promoting other crucial parameters, including the distance of the first node, number of branches, number of florescence, raceme length, number of flowers per florescence, and seed weight. These findings collectively emphasize the holistic influence of optimal NPK levels on various morphological characteristics and yield-related variables. Conversely, the control group (T_0), which received no fertilizer, consistently exhibited the lowest values across all measured variables. This highlights the essential role of NPK fertilizers in enhancing the overall morphological traits and yield of buckwheat, corroborating the crops historical struggle on the brink of extinction due to a lack of knowledge about its nutritional value. The study suggests that the common buckwheat's adaptability and resilience can be further harnessed through precise nutrient management practices, particularly through the application of a specific combination of nitrogen, phosphorus, and potassium. These findings contribute to the ongoing revival of common buckwheat, once on the verge of extinction, by shedding light on the nutritional and agronomic aspects that promote its growth and sustainability. In light of the observed variations, future research endeavors should delve into the underlying physiological mechanisms driving these effects. Understanding the intricate interactions between NPK fertilization, genetic variations, and environmental factors can provide deeper insights into sustainable agricultural practices and contribute to the continued success of common buckwheat cultivation. Overall, this study underscores the significance of balanced nutrient management for optimizing crop performance and ensuring the sustainability of agricultural systems. The assessment of antioxidant enzyme activity in common buckwheat plants exposed to different NPK fertilizer treatments provides valuable insights into the intricate relationship between nutrient availability and the plant's antioxidative defense mechanisms. The findings, as outlined in Table 4.1.3, highlight distinct patterns of enzyme activity across various treatments, emphasizing the significance of balanced nutrient supplementation in enhancing the plant's ability to cope with oxidative stress. Peroxidase, a key enzyme involved in H2O2 detoxification, exhibited its highest activity in Treatment for (T_4), characterized by the application of nitrogen (75Kg/h), phosphorus (95Kg/h), and potassium (45Kg/h). This heightened peroxidase activity in T_4 suggests an efficient defense mechanism against oxidative stress, showcasing the positive impact of the specific NPK combination on the plant's antioxidative response. Conversely, the lowest peroxidase levels observed in the control group (T0) highlight a potential vulnerability to oxidative stress in the absence of NPK supplementation. Ascorbate, another vital antioxidant, reached its peak level in T4, underscoring the optimized plant response to oxidative stress under this specific NPK treatment. The lowest ascorbate levels in T_0 further emphasize the crucial role of NPK fertilization in enhancing the plant's capacity to combat oxidative stress. Surprisingly, Catalase activity, responsible for decomposing hydrogen peroxide, exhibited the highest level in the control group (T_0). This unexpected result suggests a compensatory mechanism in the absence of additional NPK supplementation, highlighting the adaptability of plants to varying nutrient conditions. Hydrogen peroxide levels were highest in T4, aligning with the elevated peroxidase activity observed in the same treatment. This coordinated response indicates that increased peroxidase activity effectively manages higher levels of hydrogen peroxide, further emphasizing the positive impact of the specific NPK treatment in T_4 on the plant's oxidative stress tolerance. In summary, the observed variations underscore the importance of balanced nutrient management in influencing antioxidant enzyme activity and hydrogen peroxide levels. The results support the notion that optimal nutrient supplementation, as exemplified by the specific NPK combination in T_4, can enhance a plant's resilience to oxidative stress, ultimately contributing to improved overall growth and yield. Further investigations into the molecular pathways and interactions underlying these responses are warranted for a comprehensive understanding of the mechanisms at play. These insights hold promise for advancing sustainable agricultural practices and enhancing crop resilience in the face of environmental stressors. The observed patterns in MDA, proline, and hydrogen peroxide levels at different time points offer valuable insights into the dynamics of biochemical processes within the studied system. The highest MDA level at T0 indicates an initial heightened activity or response, potentially influencing subsequent biochemical pathways. This finding aligns with the notion that MDA serves as a marker for cellular activity and may have downstream effects on other biological processes. The recorded increase in proline concentration at T4 suggests a potential accumulation or upregulation of proline content throughout the experiment. The notable variation in proline levels (mean = 2.43, SD = 0.22) underscores the complexity and potential biological variability in the system. Correlations or trends between MDA and proline measurements highlight the intricate interplay of these biochemical components, providing a basis for further investigation into their functional relationships. The concurrent decrease in hydrogen peroxide at T4, coupled with the increase in proline, suggests a potential shift in cellular redox balance or stress response mechanisms. This dynamic interplay between oxidative stress markers and protective compounds like proline may reflect the adaptive nature of the system to environmental stimuli. To enhance the robustness of these findings, it is essential to consider potential confounding variables or experimental conditions that might have influenced the observed changes. Further investigations, including pathway analysis and additional time points, would contribute to a more comprehensive understanding of the underlying biological processes. Overall, the presented data provides a foundation for deeper exploration into the intricate biochemical dynamics of the system under study and opens avenues for future research.

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APPENDICES

Appendix I : Buckwheat cultivars antioxidant and H_2O_2, MDA,proline, levels enzyme activity

Buckwheat varieties	Treatment	Ascorbate	Peroxidase	Catalase	Hydrogen peroxidase	MDA	Proline
L18	T_0	1.163±1.17	2.60 ± 1.50	0.96±0.47	1.65 ± 0.10	3.57±0.10	0.72±0.09
	T_1	2.33 ± 1.14	3.70± 2.60	0.74±0.22	1.74± 0.07	2.78±0.25	0.97±0.12
	T_2	3.07± 2.66	4.70± 3.20	0.59±0.11	1.54± 0.28	2.11±0.22	1.47±0.17
	T_3	2.61± 0.84	6.00 ±4.5	0.68±0.08	1.38± 0.22	1.49±0.18	1.84 ±0.12
	T_4	4.26± 0.82	8.20 ±5.50	0.72±0.03	1.57± 0.05	0.52±0.02	2.43 ±0.22

Appendix II: Analysis of variance for plant height

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Treatment	4	766.31	191.578	11.8720	0.001911 **
Replication	2	12.02	6.008	0.3723	0.700486
Residuals	8	129.10	16.137		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Appendix III: Analysis of variance for distance of first node

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Treatment	4	1.24389	0.310973	4.2394	0.03925 *
Replication	2	0.19097	0.095487	1.3017	0.32402
Residuals	8	0.58683	0.073353		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Appendix IV: Analysis of variance for number of influorecence

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Treatment	4	1.24389	0.310973	4.2394	0.03925 *
Replication	2	0.19097	0.095487	1.3017	0.32402
Residuals	8	0.58683	0.073353		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Appendix V: Analysis of variance for receme length

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Treatment	4	17.2040	4.3010	24.2558	0.0001581 ***
Replication	2	2.1519	1.0759	6.0678	0.0249176 *
Residuals	8	1.4185	0.1773		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Appendix VI: Analysis of variance for number of per influorecence

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Treatment	4	17.2040	4.3010	24.2558	0.0001581 ***
Replication	2	2.1519	1.0759	6.0678	0.0249176 *
Residuals	8	1.4185	0.1773		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Appendix VII: Analysis of variance for number of brances

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Treatment	4	15.9977	3.9994	113.78	1.902e-06 ***
Replication	2	0.2489	0.1244	3.5404	0.08663*
Residuals	7	0.2460	0.0351		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1

Appendix VIII: Analysis of variance for per polot seed weight

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Treatment	4	388529	97132	16.4217	0.0006343 ***
Replication	2	17604	8802	1.4881	0.2821983
Residuals	8	47319	5915		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Appendix IX: Analysis of variance for 1000 grain seed weight

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Treatment	4	0.149893	0.037473	85.0057	5.163e-06 ***
Replication	2	0.009514	0.004757	10.7911	0.00727 **
Residuals	8	0.003086	0.000441		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1

Appendix X: Analysis of variance for Ascorbate

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Treatment	4	8.2052	2.0513	0.6484	0.6437***
Replication	2	0.1391	0.0695	0.0220	0.9783
Residuals	8	25.30	3.16		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Appendix XI Analysis of variance for Peroxidase

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Treatment	4	13.23	3.30	0.56	0.69***
Replication	2	6.95	3.47	2.953	2.95
Residuals	163	46.45	5.80		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Appendix XII Analysis of variance for catalase

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Treatment	4	0.23	0.05	0.94	0.48 ***
Genotype	9	5.307	0.590	2.3631	0.01553 *
Replication	2	0.10	0.05	0.81	0.47 ***
Residuals	8	0.49	0.06		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Appendix XIII: Analysis of variance for H_2O_2

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Treatment	4	0.21	0.05	4.60	0.03***
Genotype	9	1456.9	161.87	13.6647	3.223e-16 ***
Replication	2	0.20	0.10	8.56	0.01
Residuals	8	0.09	0.01		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Appendix XIV: Analysis of variance for MDA

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Treatment	4	17.3226	4.3306	424.2616	1.993e-08 ***
Replication	2	0.2457	0.1228	12.0335	0.00543 **
Residuals	7	0.0715	0.0102		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' '

Appendix XV: Analysis of variance for proline

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Treatment	4	5.9418	1.48544	737.149	2.904e-09 ***
Replication	2	0.2179	0.10897	54.077	5.538e-05 ***
Residuals	7	0.0141	0.00202		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' '