Effect of Zn and B on the Growth and Nutrient Uptake in Groundnut

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Authors’ contributions
This work was carried out in collaboration among all authors. Authors RN, NC and AGB designed the study and collected all the data. Authors RN and HR performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Author GCH corrected the manuscript. All authors read and approved the final manuscript.

ABSTRACT

Aims: To investigate the effect of combination between foliar zinc and boron on groundnut growth, yield, nutrient uptake and its accumulation in pods.

Study Design: Completely random design (CRD).

Place and Duration of Study: Bidhan Chandra Krishi Viswavidyalaya, Nadia, West Bengal, India during 2016.

Methodology: The pot experiment was comprised of three levels of Zn (0, 0.5 and 0.75% Zn), three levels of B (0, 0.3 and 0.45% B) and their combinations. The treatments were replicated thrice. Zn and B were applied through foliar spray twice at vegetative and flower initiation stage. Chlorophyll content, leaf area, root–shoot dry biomass, plant height, nutrient uptake and nutrient concentrations in pods were studied.

Results: Foliar spray of Zn and B jointly increased the leaf area to the tune of 55% and 29% at flowering and pod formation stages, respectively. Despite sole application of B and Zn increased the
leaf chlorophyll content in groundnut; the combined applications were much more prominent. Moreover, lower level of Zn combined with higher level of B significantly (p<0.05) had higher uptake of N (18.8%), P (11.5%) and K (5.9%) over higher level of sole Zn application. The improved biomass accumulation of groundnut amplified the efficient utilization of primary nutrients and resulted in higher nutrient uptake as well as their concentration in pods. Groundnut when sprayed with elevated doses of Zn and B produced the maximum yield (30.8 g/plant).

**Conclusion:** Spraying of Zn and B increased plant biomass, leaf area, chlorophyll content noticeably and with the increase in concentration of Zn and B in spray, the increment became quite intense. The combined spray of Zn and B at critical growth stages promoted better growth and productivity of groundnut.

**Keywords:** Foliar spray; chlorophyll; leaf area; nutrient uptake; biomass, groundnut.

### 1. INTRODUCTION

Groundnut (*Arachis hypogaea* L.) is a crop of global importance. It is classified as both a grain legume and an oilseed crop, because of its high oil content. It is unpredictable, heavy feeder legume and is cultivated worldwide on almost all types of soil. It contains about 50% oil, 25-30% protein, 20% carbohydrate and 5% fiber and ash which make groundnut a rich source of nutrition [1]. It is a valuable cash crop cultivated by millions of small farmers throughout the world, because of its economic and nutritional value. In spite of recommended application of fertilizer (NPK – nitrogen, phosphorus, potassium) the yield does not reach the potential level [2]. One of the major constraints for low yield of groundnut relates to deficiency of micronutrients. Intensification of agriculture, usage of straight fertilizers, rising crop requirements due to increasing productivity levels have heightened the micronutrients demand in soil fertility management and are increasingly becoming major constraints to achieve agricultural production.

Nowadays zinc deficiency is virtually an all India problem and in West Bengal 9-68% of soils are Zn deficient [3]. In 1980 the extent of B deficiency was about 2% [4] and it has increased to 18.3% in just 32 years [5] in India. It is well established that zinc is one of the most important nutrient required for plant growth as it plays as an activator of several enzymes in plants and is directly involved in the biosynthesis of growth substances such as auxin which produces more plant cells and more dry matter. On the other hand, boron is the next important micronutrient after Zn required by the plant for their growth and development. Boron has the ability to increase photosynthetic and enzymatic activity in plant; moreover, it causes pollen grain germination, pollen tube growth and viability of pollen grains [6]. It was evident that application of zinc and B enhanced the seed and oil yields/ha and protein percentage in groundnut [7,8,9]. Additionally, foliar spray enables plants to absorb the applied nutrients from the solution through their leaf surface and thus, may result in the economic use of fertilizer [10]. So, the proper micronutrient fertilizer management of groundnut crop with reference to amount, method and time of application has significant effect on yield and quality [11]. However, information of different concentration of Zn and B and their combinations on groundnut is meager. Keeping these in view, a study was undertaken to investigate the effect of combination between foliar zinc and boron on growth, yield and nutrient constituents of groundnut.

### 2. MATERIALS AND METHODS

#### 2.1 Seed Material

A popular variety of ground nut Tag 24 was grown for the experiment. The seeds were chosen with uniform size, colour and weight. Seeds were grown in earthen pots of 25 cm diameter and 25 cm height arranged in completely randomized design at Bidhan Chandra Krishi Viswavidyalaya, Nadia, West Bengal, India during 2016.

#### 2.2 Soil Preparation

Bulk soil samples were collected at a depth of 0-20 cm for conducting pot experiment. Approximately 5 Kg air dried soil was filled in each earthen pot. The soil was moderately fine texture soil (clay loam) and represented the taxonomical class of Aeric Haplaquept. The pH of the soil was ranged 6.7-7.3. The chemical and physical properties of soil are presented in Table 1. The organic matter content of the soil ranged from 1.14-1.25%. The available N, P and K status of soil were 125.6, 50.5 and 252 kg/ha,
respectively. Urea, single super phosphate and muriate of potash were used in equivalent amount in pot to supply 17 Kg N, 34 Kg P2O5 and 50 Kg K2O /ha, respectively. Entire dose of P and K were applied as basal and N was applied in two split doses viz. two third of N at the time of sowing and rest one third as top dressing at peak vegetative stage.

2.3 Treatments

The pot experiment was comprised of three levels of Zn (0, 0.5 and 0.75 % Zn as ZnSO4), three levels of B (0, 0.3 and 0.45 % B as H3BO3) and their combinations. The treatments were replicated thrice. The treatments are as follows -

\[\begin{align*}
Z0B0 & - \text{Control} \\
Z0B1 & - \text{B spray by 0.30\%} \\
Z0B2 & - \text{B spray by 0.45\%} \\
Z1B0 & - \text{Zn spray by 0.50\%} \\
Z1B1 & - \text{Zn spray by 0.50\% + B spray by 0.30\%} \\
Z1B2 & - \text{Zn spray by 0.50\% + B spray by 0.45\%} \\
Z2B0 & - \text{Zn spray by 0.75\%} \\
Z2B1 & - \text{Zn spray by 0.75\% + B spray by 0.30\%} \\
Z2B2 & - \text{Zn spray by 0.75\% + B spray by 0.45\%}
\end{align*}\]

Zn and B were applied through foliar spray twice at vegetative and flower initiation stage. Soil moisture content in all pots was kept at field capacity (25-30\% v/v basis) during the experiment. Mean maximum and minimum temperature during the study were 35\ºC and 18\ºC, respectively.

2.4 Soil and Plant Analysis

2.4.1 Soil pH

The pH of the soil was determined by using soil and water ratio 1:2.5 (w/v) [12] and glass electrode pH meter (Systronix, Hyderabad, India).

2.4.2 Soil organic matter

Oxidisable organic carbon (Cox) of the soil was determined following the method of Walkley and Black [13]. 1.0 g of soil was wet oxidized by 10 ml of 1(N) K2Cr2O7 and 20 ml concentrated H2SO4. The digested material was kept in dark place for 30 min followed by titration with 0.5 N ferrous ammonium sulphate after the addition of 200 ml of water and 10 ml of orthophosphoric acid. It is expressed as g C/kg soil. A factor of 1.724 was multiplied to get soil organic matter (SOM).

2.4.3 Available nitrogen

The available N was determined by Kjeldahl flask using alkaline permanganate method [14] following titration with H2SO4.

2.4.4 Available phosphorus

Available phosphorus content was determined following Olsen’s method using 0.5 (M) NaHCO3 (pH 8.5) extractant as the reaction of the soil samples were in the range of neutral to slightly alkaline (pH 6.7-8.1)[15].

2.4.5 Available potassium

Available K2O was estimated by neutral normal ammonium acetate using flame photometer.

2.4.6 Leaf area

The leaf area of each plant was taken during vegetative, flowering and harvesting stages. First, the green leaf portions were separated and the area of the leaves was measured in graphical method. Mean value of three leaf samples collected from top middle and lower portion of plant was calculated. Then this mean value per plant was multiplied with leaf number to get total leaf area of the plant.

2.4.7 Chlorophyll content

Plant leaf samples were also used for the determination of chlorophyll concentration. Total chlorophyll concentration was determined from 10 to 15 fully expanded groundnut leaves from plants of each pot by 95\% (v/v) ethanol extraction method [16] during the vegetative, flowering and pod-formation stages. Known weight of leaves was mildly crushed and homogenised using 3 ml chilled ethanol (4\ºC), kept overnight. The extract was decanted and the volume was made up to 5 ml. The absorbance (E) of the extract was measured at 645 and 663 nm in a Vis-spectrophotometer (Systronics, Hyderabad, India. Model-104) against a blank of ethanol. The total chlorophyll content was calculated by using the following formula and expressed in g fresh weight per litre:

\[
\text{Chlorophyll a} = 0.0127E_{663} - 0.00268E_{645} \\
\text{Chlorophyll b} = 0.0229E_{645} - 0.00468E_{663} \\
\text{Total Chlorophyll} = \text{Chlorophyll a} + \text{Chlorophyll b}
\]

Where, \(E_{645}\) – absorbance of the extract measured at 645 nm,
Table 1. Hydro-physical and chemical properties of the soil (0-20 cm)

| Texture | Bulk density (g/cm³) | Water holding capacity (%) | pH | SOM† (%) | Available N (kg/ha) | Available P₂O₅ (kg/ha) | Available K₂O (kg/ha) |
|---------|---------------------|----------------------------|----|----------|---------------------|------------------------|------------------------|
| Sand (%) -36 | 1.37                | 44                         | 6.7-7.3 | 1.14-1.25 | 125.6               | 50.5                   | 252.0                  |
| Silt (%) -28  | Clay loam            |                            |      |          |                     |                        |                        |
| Clay (%) -36  |                      |                            |      |          |                     |                        |                        |

† Soil organic matter

E₆₆₃ - absorbance of the extract measured at 663 nm,

0.0127, 0.00269, 0.0229 and 0.00468 formed by the extinction coefficients of chlorophyll a and chlorophyll b.

2.4.8 Total plant biomass

Plant samples from each pot in each replication were selected for biomass measurement and the mean of three replications was computed. Plant samples were collected by uprooting whole plant by soil excavation at harvesting periods. After excavation, the samples were slaked by dipping it into water and washed by gently flowing water. For dry biomass analysis the collected plant samples were oven dried at 60°C for more than 48 h till constant weight of the samples was observed. Calculated total dry matter was expressed as g/plant.

2.4.9 Plant sample analysis (Nutrients concentration and uptake)

The sample plants were oven-dried for dry matter content at 60°C for 48 h and were ground and analysed for total nutrient concentrations. P and K concentrations in the plant dry matter were determined after wet digestion with HNO₃ and HClO₄, and measured by the vanadomolybdate method for P and by flame photometry for K. For N plant sample was digested with sulphuric – salicylic acid mixture and measured N by the micro-Kjeldahl procedure. Nutrient uptake was calculated as the product of nutrient concentration and biomass of the crop on a dry matter basis.

2.5 Yield

The crops were harvested manually simply by uprooting at maturity stage. The pods were seperated and allowed to dry in the sun. After complete sun drying, when the moisture content of pod nearly 10-12%, the crop was cleaned and pod weight was recorded.

2.6 Statistical Analysis

Pot experiment data were analyzed statistically using analysis of variance (ANOVA) by CRD. All data were analyzed using the SPSS (Statistical Package for the Social Sciences). Data presented here are mean values and standard deviation (±SD). One-way ANOVA was carried out using Post hoc multiple comparison from the Duncan’s test at a significance level of p < 0.05.

3. RESULTS AND DISCUSSION

3.1 Plant Height

The effect of Zn and B on plant height of groundnut is depicted in Fig. 1. The plant height of groundnut increased with the age of the crop and attained maximum at harvest. At vegetative stage of the crop the treatments did not show any significant differences on plant height. Although, there was significant differences among the treatment in plant height at flowering and harvesting stages. The height of the plant varied from 28.3 – 35.6 cm and 31.3 - 41.5 cm at flowering and harvesting period respectively among the treatments. Combined application of Zn and B significantly enhanced the height of the plant being the maximum under Z2B2 at both flowering and harvesting followed by Z2B1. Z2B2 recorded maximum 39% increase in plant height at flowering stage followed by Z2B1 (32%). Das (1992) also observed the increased vegetative growth of groundnut with foliar applications of Zn and B. Though, the sole application of B did not have any effect on plant height during the entire growth phase [17]. The lowest value of plant height was noted under control (Z0B0) throughout the growth stages.
3.2 Leaf Area Per Plant

Leaf area of plant denotes the activity of photosynthesis by regulating the interception of sunlight. There was continuous increase of leaf area over time from vegetative to maturity. Throughout the growing phase leaf area was ranged 0.032 – 0.034, 0.036 – 0.053 and 0.046 – 0.068 m²/plant during vegetative, flowering and harvesting period, respectively. Though at vegetative stage, application of Zn and B either singly or in combination did not affect the leaf area significantly. Leaf area of Z2B2 significantly increased over other treatments during flowering (55%) and pod formation period (29%). The Z2B1 and Z1B2 were increased over Z1B1 and other sole application of Zn or B. Z1B1 showed 8.5% more leaf area than Z2B0 during flowering period; though during harvesting period their value became statistically at par. This result clearly indicated the effect of less concentration of simultaneous Zn and B application was better than application of higher concentration of Zn or B separately. This might be due to the synergistic interaction between Zn and B which was corroborated with findings of Sinha et al. in mustard [18]. During flowering to harvesting period the leaf area of groundnut under Z2B1 increased the maximum about 65% followed by Z1B2. The Z0B2 and Z0B1 had no significant effect on leaf area development throughout the growing period.

![Bar chart showing plant height under different Zn and B treatments during vegetative, flowering and harvesting stage of groundnut](Fig. 1)

Fig. 1. Plant height under different Zn and B treatments during vegetative, flowering and harvesting stage of groundnut

(*lower-case letters indicate comparisons of treatment during flowering stage and **upper-case letters indicate comparisons of treatment during harvesting stage)

![Line graph showing leaf area under different concentration of Zn and B foliar spray treatments during vegetative, flowering and harvesting stage of groundnut](Fig. 2)

Fig. 2. Leaf area under different concentration of Zn and B foliar spray treatments during vegetative, flowering and harvesting stage of groundnut
3.3 Chlorophyll Content

Chlorophyll content is the main indicator of plant photosynthetic products. In general, combine application of Zn and B increased chlorophyll content from vegetative to flowering period but thereafter no such significant increase in chlorophyll content was noticed (Fig. 2). Whereas the sole application of Zn or B increased chlorophyll content significantly during flowering to pod formation period. Duyingqiong et al. [19] similarly found that B significantly improved chlorophyll content and photosynthetic activity in leaves. During vegetative period there was no such significant difference of chlorophyll content of leaf among the treatments. It ranged from 1.47 – 1.53 g/l during vegetative period. However, the effect of Zn and B application on chlorophyll content was prominent during flowering stage of groundnut. Z2B2 increased the highest about 49% during flowering period from vegetative period; followed by Z2B1 and Z1B2 with an increase 44 and 28%, respectively. This is supported by the result of Saini et al. [20] that application of Zn increased chlorophyll content in leaves. However, sole application of B experienced no significant effect on chlorophyll content; whereas Z2B0 chlorophyll content (2.33 g/l) was significantly higher than that of Z1B0 (2.19 g/l) during pod formation period. From flowering to pod formation period, the maximum increase in chlorophyll content was found under Z2B0 followed by Z1B0.

3.4 Biomass

The dry biomass (shoot and root) of groundnut varied significantly among the treatments (Table 2). The nutrient treatment with Zn and B affected shoot biomass production than root. The maximum shoot (38.89 g/plant) and root biomass (1.57 g/plant) accumulation were recorded under Z2B2 followed by Z2B1 with shoot and root biomass values of 37.48 and 1.51 g/plant, respectively. The increase in biomass is mainly ascribed to optimum utilization of solar radiation, higher assimilates production and its conversion to starch [21]. The lesser dose of combined Zn and B application (Z1B1) had significantly increased biomass development over Z2B0 and Z0B2. But the increase in dry root biomass in Z1B1 over Z2B0 was less evident than development in dry shoot biomass. Although, sole application of higher doses of Zn and B did not have much influence on dry matter accumulation in both shoot and root.

3.5 Nutrient Uptake

The nutrient uptake by groundnut at maturity was affected due to various levels of Zn and B application to plants. The uptake of N, P and K increased with the increase in concentration of Zn and B either combined or sole application (Table 3). However, the combined application of Zn and B enhanced the uptake of nutrients showing more efficient utilization of nutrients per unit dry matter production. The uptake of N (1188.8 mg/plant), P (129.5 mg/plant) and K (577.9 mg/plant) was the maximum under Z2B2 followed by Z2B1. The reason behind that may be due to the increase in growth that ascribed to better root formation which in turn activated higher absorption of nitrogen from soil and improved metabolic activity inside the plant [22]. Lower level of Zn combined with higher level of B (Z1B2) significantly had higher uptake of N (18.8%), P (11.5%) and K (5.9%) over higher level of sole Zn application (Z2B0). The effect of Zn and B were more in N uptake than P and K. The uptake of N, P and K were 994, 115 and 539 mg/plant for Z1B2 and for Z1B1 these were 910, 110 and 528 mg/plant, respectively. The uptake of N increased significantly due to increased level of B application. Jiang et al. (1994) also suggested that increasing the level of B application significantly increased the N uptake by groundnut [23]. Whereas, the elevated amount of B application had no such significant effect on P and K uptake by groundnut.

3.6 Nutrient Content in Pod

The results showed that the combined application of Zn and B had significant effect on pod nutrient concentration (Table 3). Nutrient content of pod increased with increasing the level of Zn and B. In the experiment, Z2B2 recorded the highest N content in groundnut pod (37.1 mg/g) followed by Z2B1 (35.8 mg/g). Similarly, Ramamoorthy and Sudarshan (1992) found that protein content in seed was increased due to Zn and B fertilization [24]. The N content of pods obtained in Z1B2, Z1B1 and Z2B0 were 32, 30.7 and 26.1 mg/g, respectively. Boron helped in proper seed setting, seed quality and the absorption of nitrogen to a certain extent [25]. The results exhibited that the concentration of P and K in groundnut pod under Z1B2 and Z1B1 were statistically at par. Similarly, the B application showed no such difference in P and K content in pod. Z0B1 and Z0B2 had P content 4.33 and 4.34 mg/g of pod, respectively; and K...
content of 7.11 and 6.97 mg/g of pod, respectively. Although, Z0B2 recorded the highest content of P (4.3 mg/g) and K (7.1) in groundnut pod followed by Z0B1.

3.7 Yield

Groundnut when sprayed with elevated doses of Zn and B (Z2B2) produced the maximum yield (30.8 g/plant) which was 12.6% higher than Z2B1 (27.3 g/plant) (Table 2). A mean value of 25.5 and 23.8 g/plant groundnut pod yield was obtained for Z1B2 and Z1B1. The combination of Zn and B had an advantage in carbohydrate transport and pollen tube growth that influenced fruit setting and yield [26,7]. Although, the sole application of higher dose of Zn (Z2B0) on pod yield of groundnut was observed only 4% higher than Z1B0. Similarly, the sole application of B did not influenced much in pod yield of groundnut. The average yield of Z0B1 and Z0B2 were 22 and 22.8 g/plant. Whereas, the combined application of Zn and B in lesser concentration (Z1B1) significantly increased groundnut yield about 7% over sole application of higher dose of B (Z0B2). Habbasha (2014) found the highest yield of 32.6 g/plant by applying 0.75% foliar spray of ZnSO4 [27].

![Fig. 3. The effect different Zn and B treatments on chlorophyll content during vegetative, flowering and pod formation stage of groundnut](image)

Table 2. The effect different Zn and B treatments on dry shoot biomass, dry root biomass and pod yield of groundnut

| Treatments | Shoot dry weight (g/plant) | Root dry weight (g/plant) | Yield (g/plant) |
|------------|---------------------------|--------------------------|-----------------|
| Z0B0       | 26.43 g                   | 1.23 e                   | 21.52 f         |
| Z0B1       | 27.62 f                   | 1.25 e                   | 22.01 ef        |
| Z0B2       | 28.15 f                   | 1.26 e                   | 22.83 e         |
| Z1B0       | 30.77 e                   | 1.33 d                   | 22.86 e         |
| Z1B1       | 35.19 c                   | 1.42 c                   | 24.33 d         |
| Z1B2       | 35.92 c                   | 1.45 c                   | 25.54 c         |
| Z2B0       | 32.81 d                   | 1.36 d                   | 23.79 e         |
| Z2B1       | 37.48 b                   | 1.51 b                   | 27.32 b         |
| Z2B2       | 38.89 a                   | 1.57 a                   | 30.77 a         |
| CD (p<0.05)| 0.687                     | 0.03                     | 0.479           |

Z0B0 – Control, B1 - B spray by 0.30 %, B2- B spray by 0.45%, Z1 - Zn spray by 0.50%, Z2- Zn spray by 0.75% and Z1B1, Z1B2 etc are combinations of Zn and B.

Different letters at the same column show significant differences at 0.05 level (DMRT-Duncan’s Multiple Range test)
Table 3. The effect different Zn and B treatments on uptake (N, P and K) of plant and pod nutrient (N, P and K) concentration of groundnut

| Treatments | Plant N (mg/plant) | Plant P (mg/plant) | Plant K (mg/plant) | Pod N (mg/g) | Pod P (mg/g) | Pod K (mg/g) |
|------------|--------------------|--------------------|--------------------|-------------|-------------|-------------|
| Z0B0       | 807.3 g            | 85.0 e             | 443.9 e            | 21.4 g      | 4.1 de      | 6.9 g       |
| Z0B1       | 820.4 g            | 91.9 de            | 451.8 e            | 22.7 g      | 4.3 de      | 7.0 g       |
| Z0B2       | 831.2 g            | 95.3 de            | 455.5 e            | 23.6 f      | 4.3 cd      | 7.1 ef      |
| Z1B0       | 836.8 f            | 97.9 de            | 485.3 d            | 24.6 d      | 4.5 cd      | 7.2 ef      |
| Z1B1       | 910.6 d            | 109.9 bcd          | 528.3 bc           | 30.7 cd     | 4.9 c       | 7.4 d       |
| Z1B2       | 994.4 c            | 115.3 abc          | 539.8 b            | 32.0 c      | 5.1 bc      | 7.4 bc      |
| Z2B0       | 859.1 e            | 103.4 cd           | 509.6 cd           | 26.1 e      | 4.7 c       | 7.3 e       |
| Z2B1       | 1014.8 b           | 120.8 ab           | 545.5 b            | 35.8 b      | 5.3 b       | 7.6 b       |
| Z2B2       | 1188.8 a           | 129.5 a            | 577.9 a            | 37.1 a      | 6.2 a       | 8.0 a       |
| CD (p<0.05)| 19.43              | 7.69               | 14.16              | 0.36        | 0.053       | 0.097       |

*Z0B0 – Control, B1- B spray by 0.30%, B2- B spray by 0.45%, Z1 - Zn spray by 0.50%, Z2- Zn spray by 0.75% and Z1B1, Z1B2 etc are combinations of Zn and B.

Different letters at the same column show significant differences at 0.05 level (DMRT-Duncan's Multiple Range test)
4. CONCLUSION

This study showed that the omitting of Zn and B retarded plant growth, reduced nutrient uptake and ultimately the yield. Spraying of Zn and B increased plant biomass, leaf area, chlorophyll content noticeably and with the increase in concentration of Zn and B in spray, the increment became quite intense. Relatively, sole application of Zn had influenced plant physiology than that of B application alone. The combined application of Zn and B increased the plant growth as well as nutrient content in pod. The leaf area, chlorophyll content influenced by higher dose of Zn and B resulted higher nutrient uptake and pod yield.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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