

Effect of soil micronutrients (Zinc and Boron) on yield and uptake of wheat in an acid soil of West Bengal, India

Abstract: ~~Wheat~~ The production of wheat (*Triticumaestivum*), an important staple food in the world, ~~production~~ is often restricted due to micronutrients status in soil. Micronutrient deficiency in soil including boron (B) and zinc (Zn) is quite widespread in Asian countries including India due to prevalent soil and environmental conditions. A field experiment was conducted following randomized complete block design over a two-year period in an acid soil of *Terai* region of West Bengal to study the effect of zinc and boron on the yield and uptake of nutrients by wheat. The highest grain yield (4.38 t/ha) was obtained after the combined application of Zn and B over that of other treatment combinations (variable rates of B and Zn application with nutrients) or control (no nutrients, B and Zn). Application of one micronutrient might have accelerated the uptake of other micro- and macro-nutrients resulting in higher yield. A positive correlation was observed between the grain yield and the uptake of different nutrients with the weakest with Zn. A build-up of the nutrients in soils was also observed at the harvest. High response clearly demonstrated the necessity of micronutrients for improving production in the studied regions with acid soils.

Key words: micronutrients, synergistic effect, *Terai* region, deficiency, growth stages

Introduction

Wheat (*Triticumaestivum* L.) is the most important staple food for humans and is grown on more land than any other commercial crops in the world. It was grown on 216.6 million ha land in 2012 [1] producing 674.9 million tonnes of wheat globally, the third most produced cereal (perhaps any food crop) after maize (875.1 million tonnes) and rice (718.3 million tonnes) [1]. Wheat has higher protein content than either maize (corn) or rice or any other cereals and is the leading source of vegetable protein in human food globally reference?. In 2012, India produced 94.9 million tonnes of wheat from a cultivated area of 29.9 million ha, the largest area devoted to wheat production by any country in the world [2].

With the demand of ever-increasing population, the present day agriculture became more and more intensive and mined available nutrients from soil over years. However, one of the major

Comment [MA1]: Maybe wheat has more protein than other cereals, but I think there should be a reference. It seems as if the author says that wheat is a leading source of vegetable protein, but surely legumes will fit this role better?

31 triggering factors behind the dramatic improvement in the production and yield of wheat was the
32 supply of artificial nutrient source for plant growth and development especially the use of
33 synthetic nitrogen fertilizer. Potash and phosphorus fertilizer in addition to the nitrogen fertilizer
34 supplied the major nutrients for the growth, development and production of wheat. In addition to
35 these major (macro) nutrients, there are some nutrients, which are essential for wheat growth but
36 needed only in very small (micro) quantities. Among these, Boron (B), Zinc (Zn), Iron, (Fe),
37 Copper (Cu), Manganese (Mn), and Chlorine (Cl) are known to have effect on the grain- as well
38 as straw-yield of wheat. These micronutrients play a pivotal role in the yield improvement of
39 wheat crop [3]. They are needed in trace amount while the adequate supply improves nutrient
40 availability and positively affects the cell physiology that is reflected in yield as well [4, 5]. A
41 number of micronutrients are part of the photosynthesis and respiration processes, chlorophyll
42 formation, nucleic acid and protein synthesis, nitrogen-fixation and other biochemical pathways
43 [6-8]. However, the deficiency of micronutrients are wide spread in many Asian countries
44 including India due to calcareous nature of soils, high pH, low organic matter, salt stress,
45 prolonged draught, high bicarbonate content in irrigation water and imbalanced application of
46 NPK fertilizers [9, 10]. The deficiency of micronutrients can induce the stress in plants including
47 low crop yield and quality, imperfect plant morphological structure (such as fewer xylem vessels
48 of small size), widespread infestation of various diseases and pests and low fertilizer use
49 efficiency.

50 Zinc is one of the important micronutrients, which is important in the production of various
51 crops including wheat [11, 12]. It improves the number of grains per spike [13]. In addition to
52 the improvement of yield and yield components of wheat [14, 15], adequate supply of Zn can
53 improve the water use efficiency of wheat plants [16]. Zinc is also known to provide thermo-
54 tolerance to the photosynthetic apparatus of wheat [17]. The presence of Zn is important in plant
55 metabolism and thus the growth and production [18]. The Zn deficiency is the third most
56 common deficient nutrient after nitrogen and phosphorus [19, 20]. Zinc deficiency in plants not
57 only reduces the grain yield, but also the nutritional quality [21].

58 Boron is another important micronutrient that is essential for plant growth and improves the
59 production efficiency of wheat. However, the deficiency of B is the most frequently encountered
60 one in field [22]. Boron is essential for cell division and elongation of meristematic tissues, floral
61 organs and the flower male fertility, pollen tube germination and its elongation and the seed

and fruit formation. Lack of B can cause the 'wheat sterility' resulting in increased number of open spikelets and decreased number of grains per spike [23]. The B deficiency in soil can affect seedling emergence and cause an abnormal cellular development in young wheat plant [24]. Deficiency of B also inhibits root elongation by limiting cell division in the growing zone of root tips [25]. Deficiency of B is known to inhibit the leaf expansion and reduction in photosynthesis though the exact role of boron in photosynthesis is still least understood of all the mineral nutrients. In the field, sexual reproduction is often affected by low B reducing the grain yield significantly but without any visual symptoms expressed during vegetative growth.

The *Terai* region is located at the south of the outer foothill of the Himalaya and Siwalik hills and the north of the Indo-Gangetic plain. It spreads over a number of states in India including Himachal Pradesh, Haryana, Uttaranchal, Uttar Pradesh, West Bengal, Sikkim and Assam. It also covers a major part in Nepal, Bhutan and Bangladesh. The *Terai* region is the habitat of millions of people. It is a very productive region and agriculture is the base of the economy of the habitants. Rice and wheat are important crops of this region. The rice-wheat system is the most important cropping pattern in this region and considered to be the major determinant factor of the agriculture-based economy. However, the intensive cultivation practices overexploited the natural soil resource base, which was further enhanced by the imbalanced use of inputs [22, 26].

The deficiency of B and Zn in soils of different agro-climatic zones is not rare and *Terai* region is not an exception ~~of that~~ in this regard. Deficiency of different micronutrients has been reported from this region. Among the most prevalent ones, the deficiency of Zn is estimated to be the highest [27, 28]. Incidence of B deficiency from the areas of West Bengal and Bihar has also been reported [26]. Comprehensive study on the effect as well as the interaction of these nutrients on the production of wheat at this part of the world would help understanding constraints of cultivation and decreasing the yield gap to secure food for the future.

Based on the above perspectives the present study was undertaken in the *Terai* region of West Bengal on the following objectives:

- ❖ To assess the effect of Zn and B on the yield of wheat.
- ❖ To study the interaction effect of Zn and B on the yield of wheat.
- ❖ To evaluate the residual status of Zn and B in soil at different stages of wheat crop.

Materials and Methods

Experimental site:

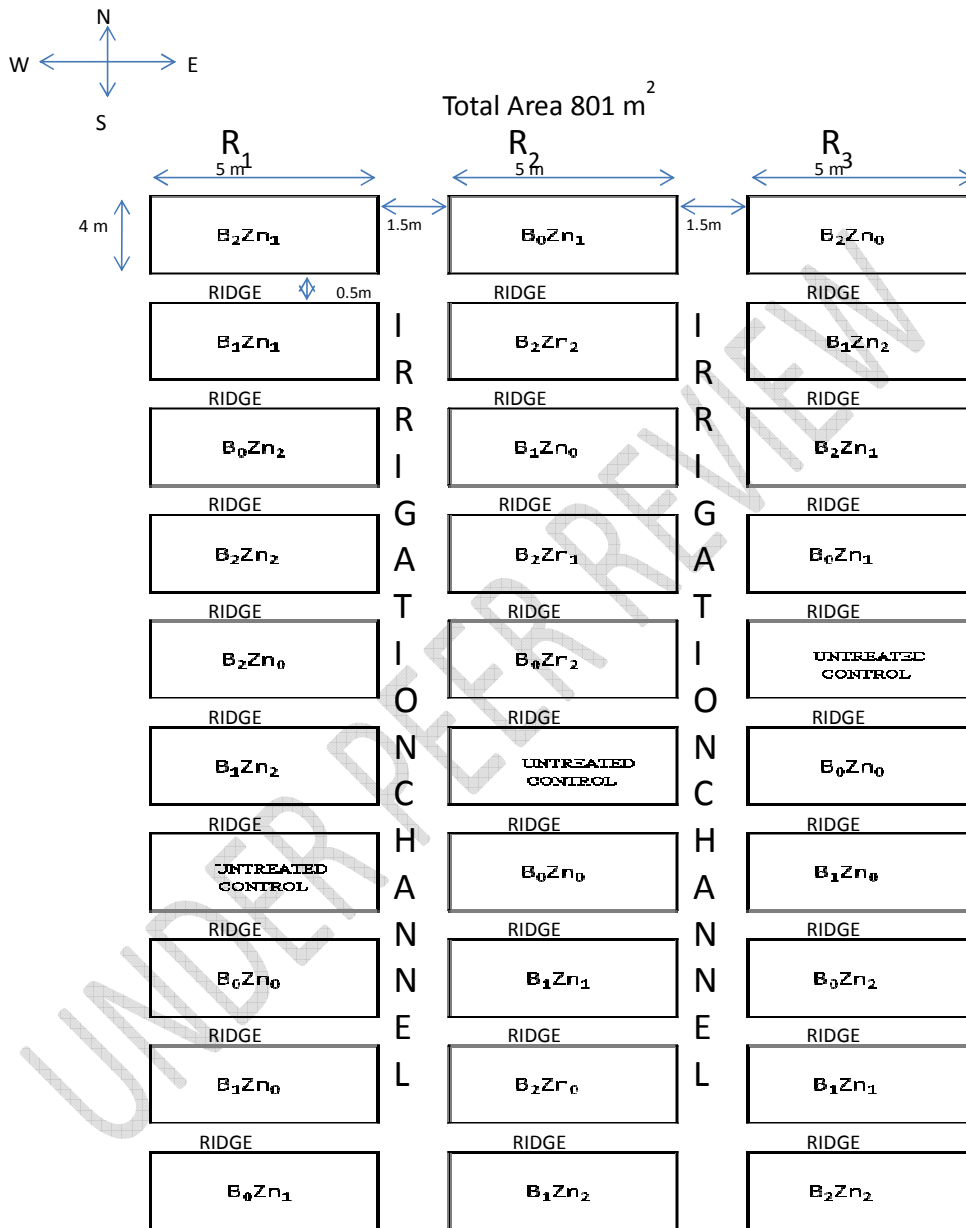
A field experiment was carried out at the agricultural farm of Uttar Banga Krishi Viswavidyalaya, Pundibari, Cooch Behar, West Bengal, India. The farm is located within the *Terai* Agro-climatic zone and its geographic location is 26°19'86" N latitude and 89°23'53" E longitude. The elevation of the farm is 43 meters above the mean sea level. The field experiment was carried out in the same field during the winter season of 2010-11 and 2011-12.

Experimental plots:

The local topography of the study area is almost flat with good drainage facilities. The soil of the experimental site is sandy loam in texture. Before laying out the experimental plots, a set of surface soil samples were collected over the whole experimental area, composite together and tested in the laboratory following the methods described in the following sub-section. The measured physical, chemical and physico-chemical properties were used as the baseline measurement for the experimental plots (Table 1).

Table 1: Initial characteristics of experimental soil for two years

Characteristics	Measurements	
	2011	2012
pH	5	5
EC (dS/m)	0.05	0.05
Organic Carbon (%)	1.04	1.02
Nitrogen (kg/ha)	206.98	188.16
Phosphorus (kg/ha)	0.76	0.89
Potassium (kg/ha)	89.60	88.48
Boron (kg/ha)	0.68	0.62
Zinc (kg/ha)	0.73	0.84



108
 109 Fig. 1: Layout of experimental plots for the field experiment. Same layout was used for
 110 both years.

A set of 30 experimental plots (5 m × 4 m) were laid out following randomized complete block design (RCBD) for this experiment (Fig. 1). Ten treatment combinations (Table 2) were developed following three doses of B (0 kg/ha, 5 kg/ha and 10 kg/ha), three doses of Zn (0 kg/ha, 12.5 kg Zn sulphate/ ha and 25 kg Zn sulphate/ ha) and a treatment without application of any nutrients (T₁₀). Though the treatment T₁ (B₀Zn₀) received recommended doses of N, P, and K, treatment T₁₀ (control) did not receive any nutrient or micronutrients (B and Zn). The treatments were replicated three times in this field experiment (Fig. 1).

Table 2: Details on the experimental plots and treatment combinations

Experimental details	
Crop	: Wheat (<i>Triticum aestivum</i> L.)
Variety	: NW 1014
Experimental design	: Randomized Complete Block Design
Total Area	: 801m ²
Plot size	: 5 m × 4 m
Number of replication	: 3
Spacing	: 23 cm (Row to Row)
Treatments	: T ₁ -B ₀ Zn ₀ , T ₂ -B ₀ Zn ₁ , T ₃ -B ₀ Zn ₂ , T ₄ -B ₁ Zn ₀ , T ₅ -B ₁ Zn ₁ , T ₆ -B ₁ Zn ₂ , T ₇ -B ₂ Zn ₀ , T ₈ -B ₂ Zn ₁ , T ₉ -B ₂ Zn ₂ , T ₁₀ - Control. B ₀ = without Boron Zn ₀ = without Zinc Sulphate B ₁ = 5 kg/ha of Boron Zn ₁ = 12.5 kg/ha of Zinc Sulphate B ₂ = 10 kg/ha of Boron Zn ₂ = 25 kg/ha of Zinc Sulphate

Field operations:

The land preparation for this experiment was started with a deep ploughing (21 and 22, December 2010 and 12 and 13 December 2012) using a tractor. A laddering (similar to levelling of soil surface) was performed after a day of soil drying following two secondary tillage using a power tiller in order to prepare a good soil tilth. The weeds and stubbles were removed by hand picking and the final laddering was performed to prepare the seed bed. Bunds and channels were prepared manually to prepare the experimental plots following the specifications mentioned in Table 1. Nitrogen (N, 100 kg/ha), Phosphorus (P, 60 kg/ha) and Potassium (K, 30 kg/ha), in the form of Urea, single super phosphate, muriate of potash; B as Borax (10 kg/ha, sodium borate), and Zn as Zn Sulphate (25 kg/ha) were applied to the soil as per the treatments. Full dose of P and K and half of the recommended dose of N and full dose of B, Zn were surface applied as

Comment [MA2]: Is it necessary to write the elements' names in capital letters? If you decide to, then Borate and Potash should surely also be in capital letters?

131 basal dose and incorporated in the soil. The remaining half of the recommended dose of N was
132 applied as top dressing at 21 days after sowing (DAS), after completion of the first weeding.

133 The wheat variety of NW-104 was used for this experiment @100 kg ha⁻¹. Sowing was
134 completed in rows (spacing 23 cm) in North-South using a duck-foot tyne at a depth of 2.5 to 3
135 cm. Two weeding operations were performed manually on 21 DAS and 45 DAS. Two irrigations
136 were applied on 21 DAS (after weeding and fertilizer application) and 65 DAS. The excess water
137 was drained out using drainage channels.

138 The soil and plant samples were collected for laboratory analysis on 21 (CRI- crown root
139 initiation stage), 55 (tillering stage), 70 (booting stage) and 110 DAS (maturity). Leaving the
140 border rows, half of the area in each plot was marked for recording biometrical observation
141 including destructive plant sampling and other half for recording yield components and yield of
142 wheat. The height (from ground level) of five randomly selected plants were recorded and
143 averaged from each plot. The measured plants were tagged after first measurement for
144 subsequent measurements. Dry weight of both roots and shoots were also recorded. The number
145 of tillers per m² was recorded from 10 randomly selected plants. The crop was harvested from
146 net plot area discarding the border row. The number of panicles per plant was recorded from 10
147 randomly selected plants and converted to number of panicles per m². Length of panicles was
148 measured prior to harvest and average length was calculated. Number of grains per panicle as
149 well as 1000 grain dry weight ~~was~~ were also recorded for each treatment. The final yield of
150 wheat and straw was recorded after sun drying and thrashing. The yields were recorded and
151 calculated as tonne per ha following,

152 **Grain yield (t ha⁻¹)= (Plot yield (kg) × 10000 / Plot size (m²) × 1000)**

153 **Experimental methods:**

154 Collected soil and plant samples were tested in laboratory following the methods described
155 below.

156 **a) Soil pH:** pH of soil samples (soil:water 1:2.5) was determined in suspensions using a
157 Systronics glass electrode-pH meter [29].

158 **b) Soil Electrical Conductivity (EC):** Electrical Conductivity was measured in soil-water
159 suspensions (soil:water 1:2.5)[29] using a digital conductivity meter of Systronics
160 (Model No. 304).

Comment [MA3]: kg ha⁻¹ or kg/ha? You use both. Stick to one. See page 6 for example.

- c) **Soil Organic Carbon (OC):** Organic carbon content of samples was estimated by Walkley-Walkley and Black's titration method [30].
- d) **Mechanical Analysis of soils:** Clay-content of soils was determined by the hydrometer method [31]. The texture of the soils was also ascertained from the particle-size distribution of sand, silt and clay.
- e) **Available nitrogen (N):** Available nitrogen in soil and plant was determined by alkaline KMnO_4 method developed by Subbiah and Asija [32].
- f) **Available phosphorus (P):** Available P in soil and plant content was determined by extracting the soil with a mixture of 0.03 M NH_4F and 0.025 M HCl [33] followed by colorimetric measurement using spectrophotometer (Systronics Model No. 167) [34].
- g) **Available potassium (K):** Available K concentration in soil and plant was measured using a flame photometer (Systronics Model No. 128) [34]. The extraction was carried out with neutral normal ammonium acetate.
- h) **Available Zinc (Zn):** DTPA-(Diethylenetriaminepentaacetic acid) extractable Zn^{+2} of soil and plant samples were determined by extraction with the extractant containing 0.005M DTPA, 0.01M CaCl_2 and 0.1M Triethanol amine buffered at pH 7.3 [35] followed by the measurement using Atomic Absorption Spectrophotometer.
- i) **Available Boron (B):** Available Boron in soil and plant was extracted by boiling a known amount of soil with double distilled water (in 1:2.5 ratio) prepared by quartz glass distillation apparatus, for five minutes under a reflux condenser, followed by cooling and filtration [36]. The concentration was measured using Atomic Absorption Spectrophotometer.

Statistical analysis

Statistical analysis for the collected data was performed in SigmaPlot (Systat Software Inc.). The significant difference between the treatments was tested using ANOVA and LSD. The interaction between the effect of B and Zn was tested using two-way ANOVA. The correlation between the yield components and nutrient uptakes were also calculated. The figures were prepared using the SigmaPlot.

Results and Discussion

The yield components and grain yield of wheat are shown in Table 3. A significant difference was observed among the treatment combinations on yield components and grain yield of wheat.

Comment [MA4]: Surely there was more than one analysis? Then analyses.

192 The maximum mean grain yield (4.38 t/ha) was observed in the treatment T₆ (B₁Zn₂), while
 193 minimum was observed in the control (1.68 t/ha) (Table 3). Relatively higher yield was obtained
 194 from the treatments T₁ (B₀Zn₀) to T₉ (B₂Zn₂) over that of the control (T₁₀). The lowest harvest
 195 index was observed in T₄ (B₁Zn₀) and the highest in controls (T₁₀) (Table 3). The application of
 196 B and Z in combination significantly ($p < 0.05$) increased the grain yield of wheat. The grain
 197 yield increase with B and Zn addition was reported by Chaudry et al. [37]. Boron concentration
 198 has been reported to increase grain yield of durum wheat by 16% [38]. This may be due to the
 199 requirement of B in wheat during the vegetative stage leading to high response to the grain yield
 200 [39, 40]. Therefore, even a small amount of Zn and B directly affected the grain yield. Mandal
 201 [41] reported a direct relationship between the number of grains and tillers and the wheat yield
 202 under B deficient soils of Terai region of West Bengal.

Comment [MA5]: Format: t/ha or t ha⁻¹

203 The effect of B on the grain and straw yield found was found to be significant at $\alpha = 0.05$
 204 (95% significant level). However, the scenario was little different for Zn. For example, for no B
 205 application, Zn had not any effect on crop yield. While with regular dose of B (5 kg B/ha), the
 206 yield increased linearly. This indicated that with a regular application of B, the efficiency of Zn
 207 increased with the application rate considered in this study. However, with excess (more than
 208 regular) application of B, the effect of Zn decreases indicating antagonistic effect between the
 209 micronutrient at high dose, specifically the high dose of B. The two-way ANOVA following a
 210 general linear model with $\alpha = 0.05$ found showed a significant interaction between the effect
 211 of B and Zn on the grain and straw yield of wheat. This means that the difference in the mean
 212 values among the different levels of B and Zn is great enough to exclude the possibility that the
 213 difference is just not due to random sampling variability after allowing for the effects of
 214 differences in Zn and B, respectively. Therefore, care should be taken in deciding the amount of
 215 micronutrient application, which may have different effect. From this study, it could be
 216 suggested to choose a regular dose of B for better efficiency of Zn. Sometime a high dose of Zn
 217 could be even beneficial with a controlled application of B.

Comment [MA6]: Not sure: was found to be significant or was found to be not significant?

Comment [MA7]: format

218 In spite of the highest dry biomass production until the booting stage in T₆ (B₁Zn₂), the T₈
 219 (B₂Zn₁) produced the highest dry straw at maturity (Fig. 2). Combination of B and Zn might have
 220 boosted the vegetative growth during the early stage, while the high amount of Zn along with a
 221 regular dose of B improved the yield and yield components of wheat at maturity [14, 15]. A
 222 combination of regular dose of Zn and B (T₅) could not produce high amount of straw compared

to other treatments with single or double dose of either Zn or B or in combination (Fig. 2). For example high straw yield with very little difference was observed among treatments T₄ (B₁Zn₀), T₆ (B₁Zn₂), T₇ (B₂Zn₀) and T₈ (B₂Zn₁). The lowest biomass production was recorded in control (T₁₀) at all stages of crop growth.

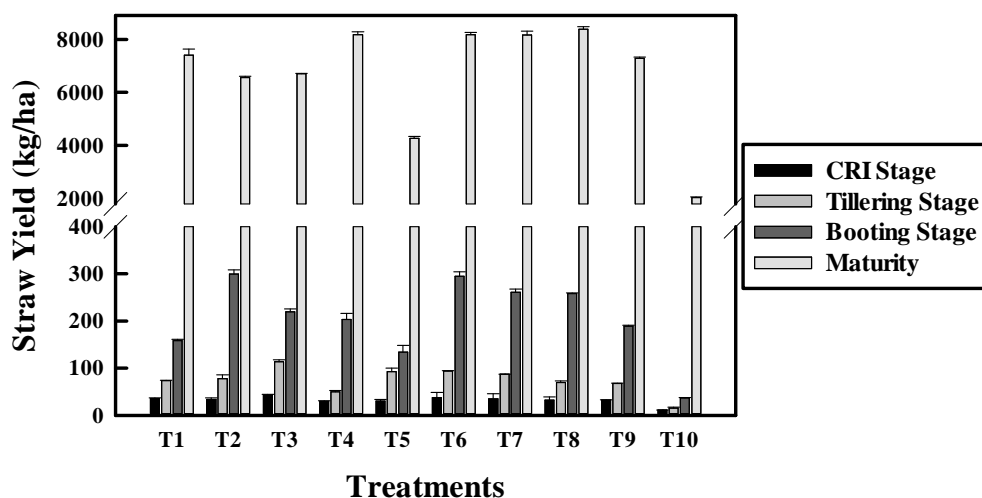


Fig. 2:Effect of treatments on average straw yield (kg/ha) over two years at different stages of wheat growth. The standard deviation of measurement is shown as error bars. The CRI stage indicates crown root initiation.

A significant difference in the nutrient uptake was recorded in different treatments and at different growth stages. The highest uptake (kg/ha) of N over the entire growth period was recorded in treatment T₆ (B₁Zn₂) and minimum in T₁₀ (control) (Fig. 3). The maximum amount of N uptake at different growth stages was not consistent. For example, T₂ (B₀Zn₁) was recorded with the highest amount of N uptake during booting stage (Fig. 3). While the highest amount of P uptake was recorded in T₇ (B₂Zn₀), the highest amount of K uptake was recorded in T₈ (B₂Zn₁) (Fig. 3). Similar to N uptake, a variable amount of P and K uptake was also recorded at different growth stages in different treatments. The highest amount of B and Zn uptake was

Comment [MA8]: format

Comment [MA9]: format

242 recorded in treatment T₉ (B₂Zn₂) (Fig. 3). High amount of B and Zn application might show
243 some synergistic

Comment [MA10]: Layout problem?

UNDER PEER REVIEW

244 **Table 3:** Effect of treatments on the yield components and grain yield (t/ha) of wheat. The data from 2011 and 2012 are shown along
 245 with the average over two years. S.D. stands for standard deviation.

Treatments	Tiller/Sq. m.			Grains/Spike			1000 Grain weight			t/ha			Harvest Index (%)		
	2011	2012	Mean (S.D.)	2011	2012	Mean (S.D.)	2011	2012	Mean (S.D.)	2011	2012	Mean (S.D.)	2011	2012	Mean (S.D.)
T1	171	168	169.5 (2.12)	37	36	36.5 (0.70)	45.72	45.75	45.74 (0.02)	2.89	2.77	2.83 (0.08)	27.7	27.6	27.6 (0.07)
T2	156	156	156.0 (0)	41	42	41.5 (0.70)	42.74	42.80	42.77 (0.04)	2.73	2.80	2.77 (0.05)	29.5	29.8	29.7 (0.21)
T3	165	162	163.5 (2.12)	39	39	39.0 (0)	45.92	45.72	45.82 (0.14)	2.95	2.89	2.92 (0.04)	30.6	30.1	30.4 (0.35)
T4	143	145	143.8 (1.41)	43	43	43.0 (0)	44.54	44.00	44.27 (0.38)	2.73	2.74	2.74 (0.01)	25.2	24.9	25.1 (0.21)
T5	158	150	153.8 (5.66)	42	42	42.0 (0)	51.50	51.51	51.51 (0.01)	3.41	3.25	3.33 (0.11)	44.1	43.5	43.8 (0.42)
T6	188	185	186.3 (2.12)	48	51	49.5 (2.12)	47.64	47.34	47.49 (0.21)	4.29	4.47	4.38 (0.13)	34.6	35.2	34.9 (0.42)
T7	176	174	174.8 (1.41)	50	53	51.5 (2.12)	43.80	43.83	43.82 (0.02)	3.84	4.04	3.94 (0.14)	32.3	32.8	32.6 (0.35)
T8	134	137	135.3 (2.12)	45	46	45.5 (0.70)	44.74	44.20	44.47 (0.38)	2.69	2.79	2.74 (0.07)	24.4	24.8	24.6 (0.28)
T9	143	142	142.3 (0.71)	52	50	51.0 (1.41)	41.38	39.38	40.38 (1.41)	3.07	2.80	2.93 (0.19)	29.7	27.6	28.7 (1.48)
T10	122	124	122.8 (1.41)	36	39	37.5 (2.12)	37.00	36.00	36.50 (0.71)	1.62	1.74	1.68 (0.08)	44.1	46.4	45.2 (1.63)

252 **Table 4:**Effect of treatments on the uptake of nutrients (kg/ha) by seed.The data from 2011 and 2012 are shown along with the
253 average over two years. S.D. stands for standard deviation.

Treatments	Nitrogen			Phosphorus			Potassium			Boron			Zinc		
	2011	2012	Mean (S.D.)	2011	2012	Mean (S.D.)	2011	2012	Mean (S.D.)	2011	2012	Mean (S.D.)	2011	2012	Mean (S.D.)
T1	70.47	59.66	65.06 (7.64)	1.40	1.74	1.57 (0.24)	23.14	24.90	24.02 (1.24)	0.280	0.255	0.267 (0.02)	0.297	0.228	0.262 (0.05)
T2	58.17	58.89	58.53 (0.51)	1.58	1.50	1.54 (0.06)	19.14	22.43	20.78 (2.33)	0.249	0.253	0.251 (0.01)	0.314	0.168	0.241 (0.10)
T3	64.54	59.85	62.19 (3.32)	1.40	1.24	1.32 (0.11)	19.21	21.66	20.44 (1.73)	0.292	0.271	0.282 (0.02)	0.406	0.361	0.384 (0.03)
T4	69.54	66.06	67.80 (2.46)	2.29	1.90	2.09 (0.28)	25.93	27.43	26.68 (1.06)	0.211	0.239	0.225 (0.02)	0.300	0.645	0.472 (0.24)
T5	83.94	72.69	78.32 (7.95)	2.25	1.88	2.07 (0.26)	18.74	21.09	19.92 (1.66)	0.237	0.289	0.263 (0.04)	0.554	0.292	0.423 (0.18)
T6	121.25	116.31	118.78 (3.49)	2.61	2.46	2.54 (0.08)	38.59	44.67	41.63 (4.30)	0.231	0.385	0.308 (0.11)	0.482	0.402	0.442 (0.06)
T7	104.39	101.86	103.12 (1.79)	2.53	2.41	2.47(0.11)	28.83	36.38	32.60 (5.34)	0.215	0.246	0.230 (0.02)	0.413	0.333	0.373 (0.06)
T8	63.97	70.97	67.47 (4.50)	1.20	1.11	1.16 (0.06)	18.81	30.64	24.73 (8/36)	0.190	0.232	0.211 (0.03)	0.255	0.195	0.225 (0.04)
T9	77.27	72.02	74.65 (3.71)	1.77	1.47	1.62 (0.21)	18.40	26.56	22.48 (5.77)	0.171	0.197	0.184 (0.02)	0.284	0.175	0.229 (0.07)
T10	13.14	9.75	11.45 (2.40)	0.76	0.79	0.77 (0.02)	7.28	9.58	8.43 (1.62)	0.061	0.060	0.060 (0.00)	0.020	0.065	0.043 (0.03)

254

effect to provide higher amount of uptake. The treatment T₁₀ (control) always recorded with the least amount of nutrient uptake.

A significant difference was observed in the uptake of different nutrients by seed (Table 4). The highest uptake of almost all nutrients (N, P, K, and B) ~~were~~ was recorded in treatment T₆ (B₁Zn₂) except for Zn, the highest uptake of which was recorded in treatment T₄ (B₁Zn₀) (Table 4). The highest production as well as the interaction between the micro-nutrients (B and Zn) in treatment T₆ facilitated higher amount of nutrients uptake in seed [42]. The lowest uptake of all nutrients was recorded in treatment T₁₀ (control). A similar trend was observed for the uptake of nutrients by straw (Table 5).

Table 5: Effect of treatments on the uptake of nutrients (kg/ha) by straw along with average (in brackets) grain yield and straw yield over two years.

Comment [MA11]: If the brackets refer to the average? Unclear.

Treatments	Grain yield (S.D.)	Straw yield (S.D.)	Nitrogen (S.D.)	Phosphorus (S.D.)	Potassium (S.D.)	Boron (S.D.)	Zinc (S.D.)
T1	2829.83 (89.10)	7410.68 (220.6)	60.26 (7.66)	0.34(0.01)	109.61 (2.83)	0.118 (0.02)	1.130 (0.01)
T2	2768.95 (49.49)	6558.93 (45.6)	19.26 (6.36)	0.38(0.00)	107.67 (2.34)	0.252 (0.03)	2.134 (1.60)
T3	2921.77 (46.67)	6702.67 (13.2)	51.61 (1.43)	0.21(0.03)	111.44 (12.46)	0.172 (0.02)	0.821 (0.21)
T4	2736.29 (9.89)	8177.83 (107.7)	27.29 (28.78)	0.36(0.05)	131.95 (14.75)	0.296 (0.26)	1.723 (0.71)
T5	3325.93 (114.55)	4268.08 (68.7)	25.67 (2.12)	0.21(0.00)	73.80 (1.44)	0.230 (0.01)	0.581 (0.06)
T6	4377.06 (126.57)	8171.53 (84.7)	84.69 (7.35)	0.23(0.01)	141.31 (3.87)	0.477 (0.03)	1.756 (2.11)
T7	3942.73 (140.71)	8160.33 (146.6)	20.36 (22.25)	0.40(0.07)	149.46 (13.66)	0.413 (0.00)	1.947 (2.34)
T8	2736.62 (68.59)	8376.87 (96.9)	34.00 (1.26)	0.33(0.00)	155.96 (8.56)	0.227 (0.03)	5.001 (6.15)
T9	2931.12 (190.92)	7288.23 (44.6)	25.52 (4.48)	0.17(0.00)	120.57 (5.46)	0.567 (0.01)	3.448 (4.61)
T10	1679.67 (86.97)	2033.42 (28.2)	2.28 (0.03)	0.06(0.04)	30.75 (0.89)	0.049 (0.00)	0.246 (0.31)

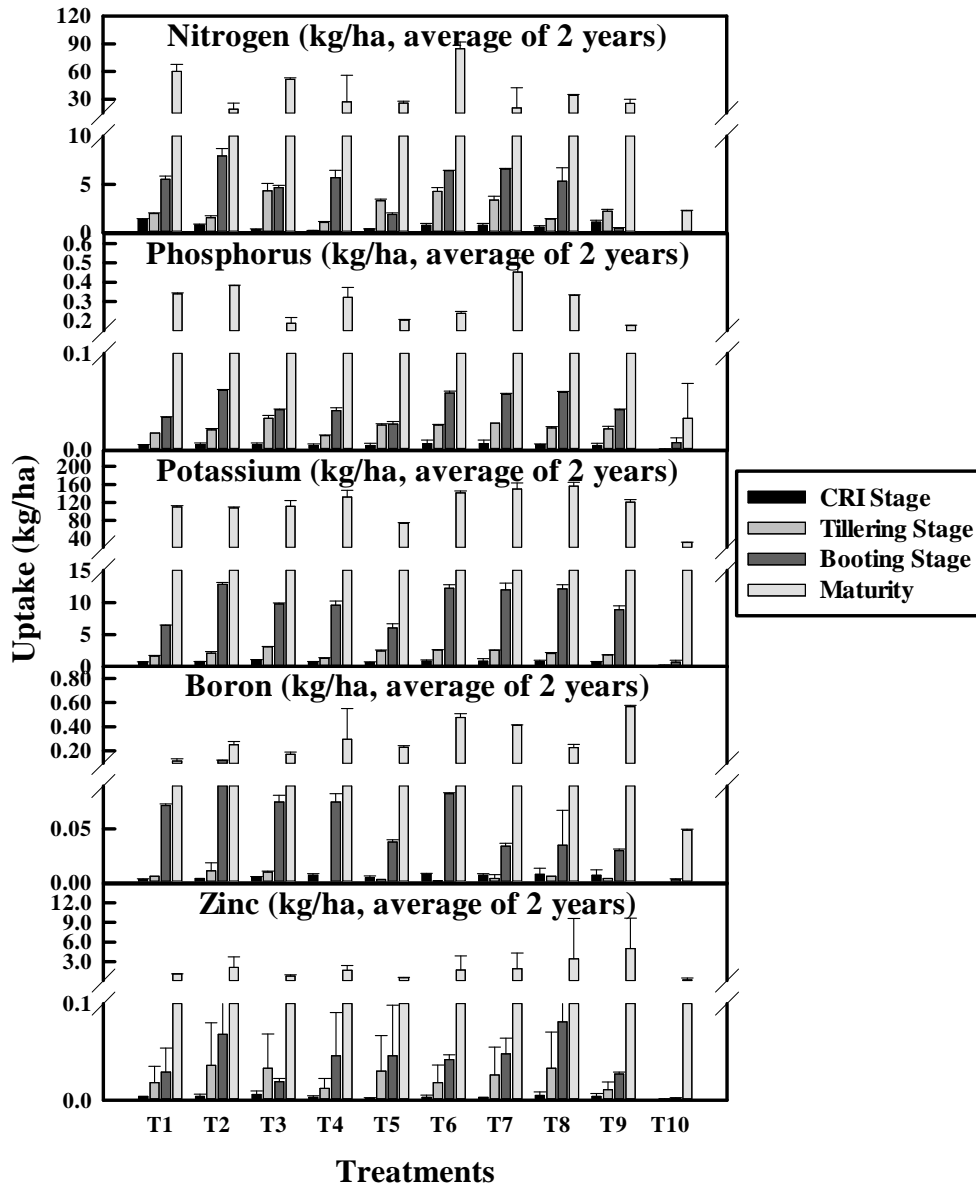


Fig. 3: Effect of treatments on the uptake of nutrients (N, P, K, B, and Zn) at different growth stages (CRI, Tillering, Booting and Maturity) of wheat. The standard deviation of measurement is shown as error bars. The CRI stage indicates crown root initiation.

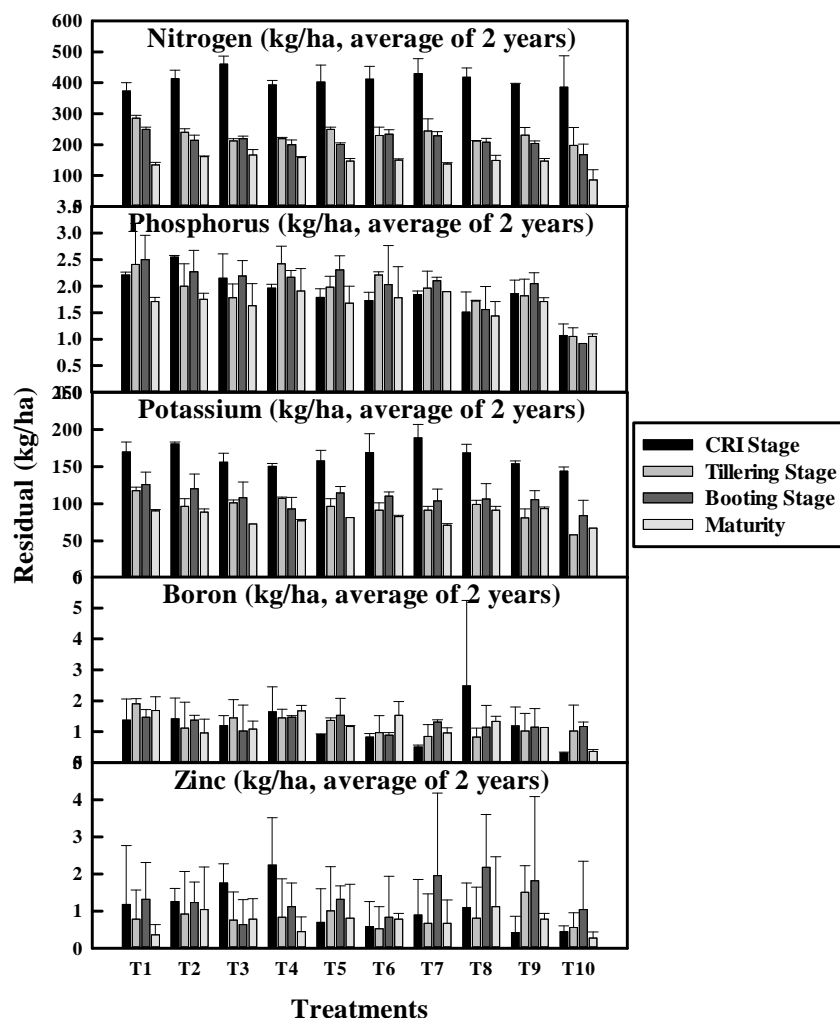


Fig. 4: Effect of treatments on the residual nutrient (N, P, K, B, and Zn) status at different growth stages (CRI, Tillering, Booting and Maturity) of wheat. The standard deviation of measurement is shown as error bars. The CRI stage indicates crown root initiation.

The B and Zn concentration in seeds (Table 4) and straw (Table 5) were calculated after dividing the total uptake of nutrients by the total grain and straw production. It clearly showed that with the increasing production, the concentration of nutrients, both B and Zn in seed and straw decreased.

280 There was significant difference in the residual N status of soil at different treatment plots
 281 and at different growth stages. Initial application of N resulted a high amount of residual N at the
 282 CRI stage and gradually decreased towards maturity, which had the least amount of residual N
 283 (Fig. 4). Minimum demand of the applied N at the beginning of the growth stages resulted in a
 284 high amount of residual N at the CRI stage, while the high demand towards maturity left the least
 285 amount of residual N. High demand during the peak growth stages such as tillering and booting
 286 resulted in a very similar amount of residual N, which was lower than that at CRI stage. The
 287 highest amount of residual N was recorded in treatment T₃ (B₀Zn₂) and the ~~least~~ lowest amount
 288 was recorded in treatment T₁₀ (control) (Fig. 4). The residual K status in soil at different growth
 289 stages of wheat showed a very similar trend as that of N (Fig. 4). The CRI stage was recorded
 290 with the highest amount of residual K, which in general decreased towards maturity. There was a
 291 significant difference between the treatments at different growth stages. Treatment T₇ (B₂Zn₀)
 292 was recorded with the highest of amount of residual K at the CRI stage, while Treatment T₁
 293 (B₀Zn₀) was recorded with the highest amount of K at other growth stages (Fig. 4). The absence
 294 of micronutrients in treatment T₁ might have inhibited the uptake resulting in a high amount of
 295 residual K.

296 The residual P in soil showed a little different trend than N and K (Fig. 4). There was no
 297 specific trend of residual P at different growth stages. In general, a higher amount of residual P
 298 was recorded at the CRI stage compared to tillering and booting stage. This might be due to the
 299 presence of unavailable form of P at the beginning of the growth stage. While the difference
 300 between the growth stages of wheat was not significant, the difference between the treatments
 301 was significant. The highest amount of P was recorded in treatment T₂ (B₀Zn₁) for the CRI stage
 302 while treatment T₄ (B₁Zn₀) at the maturity. The ~~least~~ lowest amount of residual P was recorded
 303 in treatment T₁₀ (control) (Fig. 4).

304 There was a significant difference in the residual B status in soil at different nutrient
 305 treatment combinations. However, the difference was not significant at different growth stages.
 306 There was no specific trend on the residual amount of B among the growth stages (Fig. 4). For
 307 example, while the treatment T₈ (B₂Zn₁) was recorded with the highest amount of residual B at
 308 the CRI stage, treatment T₁ (B₀Zn₀) was recorded with the highest amount of B at the tillering
 309 stage (Fig. 4). A similar trend in the residual Zn content was observed at different growth stages
 310 and at different treatments. For example, the highest amount of residual Zn was observed in

Comment [MA12]: Least for something that cannot be measured/counted. Least for something measurable.

311 treatment T₄ (B₁Zn₀) at the CRI stage, while the highest amount of residual Zn was observed in
 312 treatment T₈ (B₂Zn₁) at the booting stage (Fig. 4). A growth stage dependent Zn demand and the
 313 residual Zn were also reported by Ozturk et al. [43]. The variation in the residual Zn might also
 314 be due to the combined effect of pH, EC, organic carbon and P, which ultimately controls the Zn
 315 availability [44]. ~~A least~~ The lowest amount of residual B and Zn was observed in treatment T₁₀
 316 (control). In general a lower amount of Zn was recorded at maturity, which indicates a demand
 317 of Zn in the production of crop.

318 **Table 6:** Correlation (*r*) between soil available nutrient status at harvest and the nutrient content
 319 in straw averaged over two years

	Plant N	Plant P	Plant K	Plant B	Plant Zn
Soil N	0.41**	0.59**	0.66**	0.39**	0.33*
Soil P	0.36**	0.63**	0.67**	0.60**	0.11
Soil K	0.28*	0.39**	0.38**	0.31*	0.64**
Soil B	0.68**	0.57**	0.63**	0.26*	0.25*
Soil Zn	0.10	0.50**	0.48**	0.35	0.64**

320 *- p < 0.05, ** p < 0.01

321
 322 **Table 7:** Correlation (*r*) between soil available nutrient status at harvest and the nutrient content
 323 in seed averaged over two years

	Soil N	Soil P	Soil K	Soil B	Soil Zn
Seed N	0.54**	0.76**	0.23*	0.56**	0.37**
Seed P	0.40**	0.83**	-0.03	0.47**	0.07
Seed K	0.48**	0.71**	0.19	0.63**	0.25*
Seed B	0.80**	0.74**	0.32*	0.69**	0.41**
Seed Zn	0.70**	0.80**	-0.06	0.64**	0.15

324 *- p < 0.05, ** p < 0.01

325 A positive correlation was observed between the uptake of different nutrients and the grain
 326 and straw yield irrespective of different treatments (Fig. 5). The correlation coefficient (*r*) was as
 327 high as 0.97 between K uptake (kg/ha) and the straw yield (t/ha). There was a very weak
 328 correlation between the Zn uptake and the grain yield (Fig. 5). The uptake of nutrients was
 329 governed by the soil, environmental and management practices. For example, the availability of
 330 B was determined by the availability of Zn in soil [28]. Santra et al. [45] also reported an
 331 increased amount of DTPA extractable Zn with the application of B. The relationship between B
 332 and Zn was found to be synergistic making high amount of Zn available in soil. A high
 333 correlation was also observed between the residual nutrient status in soil and the nutrient status in
 334 straw (Table 6) or between residual status in soil and the nutrient status in seed (Table 7).

Comment [MA13]: Don't understand -p?

Formatted: Highlight

Comment [MA14]: format

Comment [MA15]: format

Comment [MA16]: Is this what was meant? Or it can be that B determined the availability of Zn?

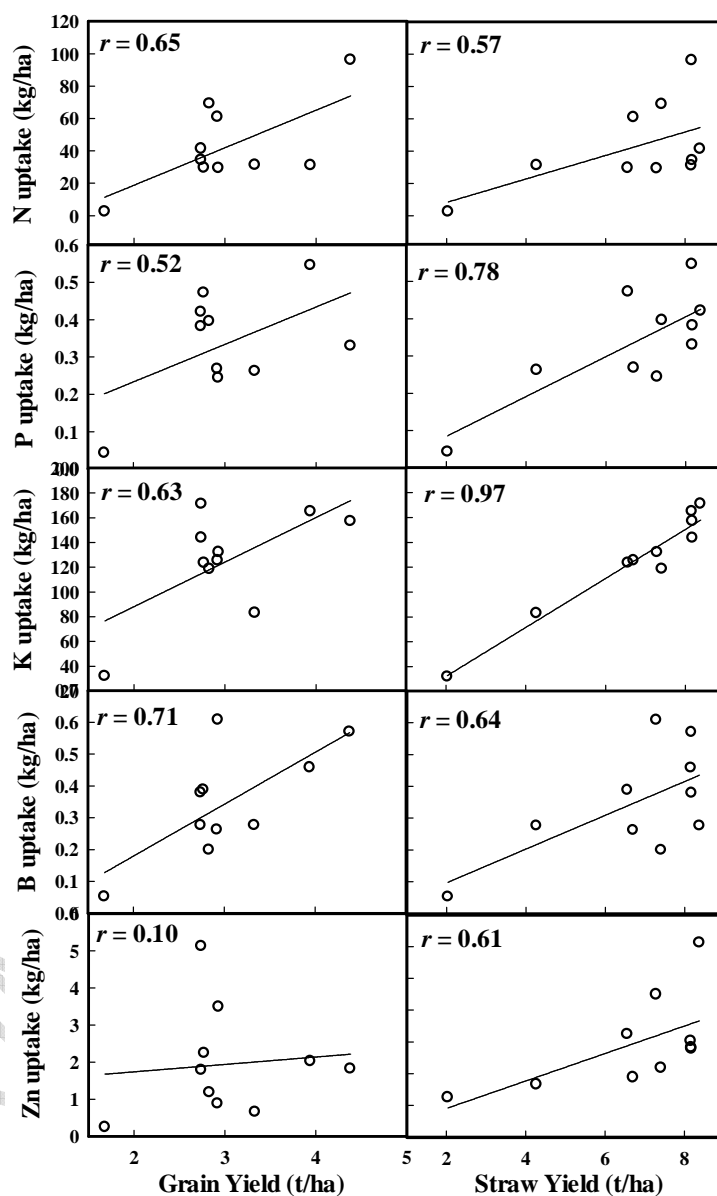


Fig. 5: Correlation coefficient (r) between the nutrient (N, P, K, B, and Zn) uptake (kg/ha) by plants and the grain and straw yield (t/ha) of wheat.

Conclusions

This study examined the effect of Boron and Zinc on the yield and uptake of different nutrients by wheat in the acid soil region of West Bengal, India. The yield components and grain yield of wheat showed a significant difference among the treatment combinations. The maximum average grain yield (4.38 t/ha) over two years was observed in the treatment T₆ with higher amount of Zn application along with recommended dose of Boron application. The minimum grain yield was observed in treatment T₁₀ (the control) (1.68 t/ha). A relatively higher yield was obtained from the treatments with any nutrient combination over that of the control (T₁₀). Along with the difference in grain yield, a significant difference in straw yield was also observed among the treatments. The application of Boron and Zinc might show some synergistic effects leading to high grain and straw yield in the acid soil region. The presence of micro-nutrients and their combination also affected the uptake of nutrients in different growth stages of wheat. The interaction effect was also visible in the uptake nutrients by seeds. A positive correlation was observed between the uptake of nutrients and the yield of grain and straw in this study region with acid soils. The residual nutrient status showed a build-up of nutrients in soils.

References

1. FAO (Food and Agriculture Organization of the United Nations) (2012) Food and Agriculture Organization of the United Nations Crop Production and Trade Statistics. (available from: <http://faostat.fao.org/site/339/default.aspx>)
2. Index Mundi (2011) Afghanistan Wheat Area Harvested by Year, last viewed 10 Aug. 2011. <http://www.indexmundi.com/agriculture/?country=af&commodity=wheat&graph=area-harvested>.
3. Rehm, G., and Sims, A. (2006) Micronutrients and production of hard red spring wheat. Minnesota Crop News, University of Minnesota.
4. Taiwo, L.B., Adediran, J.A., Akande, M.O., Banjoko, V.A., and Oluwatosin, G.A. (2001) Influence of legume fallow on soil properties and yield of maize in south-western Nigeria. *Journal of Agriculture in Tropics and Subtropics* **102**(2): 109-117.
5. Adediran, J.A., Taiwo, L.B., Akande, M.O., Idowu, O.J., and Sobulo, R.A. (2004) Application of organic and inorganic fertilizer for sustainable yield of maize and cowpea in Nigeria. *Journal of Plant Nutrition* **27**(7): 1163-1181.
6. Goldbach, H.E., Blaser-Grill, J., Lindemann, N., Porzelt, M., Horrmann, C., Lupp, B., and Gessner, B. (1991) Influence of boron on net proton release and its relation to other metabolic processes. *Current Topics in Plant Biochemistry and Physiology* **42**: 925-929.
7. Reddy, S.R. (2004) Principles of Crop Production – Growth Regulators and Growth Analysis, 2nd Ed. Kalyani Publishers, Ludhiana, India.

Comment [MA17]: format

- 374 8. Ali, S., Shah, A., Arif, M., Miraj, G., Ali, I., Sajjad, M., Farhatullah, M., Khan, Y., and
375 Khan, N.M. (2009) Enhancement of wheat grain yield and yield components through foliar
376 application of Zinc and Boron. *Sarhad Journal of Agriculture* **25(1)**: 15-19.
- 377 9. Ahmadikhah, A., Narimani, H., Rahimi, M.M., and Vaezi, B. (2010) Study on the effects of
378 foliar spray of micronutrient on yield and yield components of durum wheat. *Archives of*
379 *Applied Science Research* **2(6)**: 168-176.
- 380 10. Nadim, M.A., Awan, I.U., Baloch, M.S., Khan, E.A., Naveed, K., and Khan, M.A. (2012)
381 Response of Wheat (*Triticumaestivum*L.) to different Micronutrients and their Application
382 Methods. *The Journal of Animal & Plant Sciences* **22(1)**: 113-119.
- 383 11. Kanwar, J.S., and Randhawa, N.S. (1974) Micronutrient research in soils and plants in India.
384 Indian Council of Agricultural Research, New Delhi. p. 18-32.
- 385 12. Takkar, P.N., Mann, M.S., and Randhawa, N.S. (1971) How zinc deficiency affects wheat
386 yields. *Indian Farming* **21(9)**: 31-32.
- 387 13. Asad, A., and Rafique, R. (2002) Identification of micronutrient deficiency of wheat in the
388 Peshawar valley. *Communication in Soil Science and Plant Analysis* **33**: 349-364.
- 389 14. Kaya, Y., Arisoy, R.Z., and Gocmen, A. (2002) Variation in grain yield and quality traits of
390 bread wheat genotypes by zinc fertilization. *Pakistan Journal of Agronomy* **1**: 142-144.
- 391 15. Singh, Y.P. (2004) Effect of nitrogen and zinc on wheat irrigated with alkali water. *Annals*
392 *of Agricultural Research* **25**: 233-236.
- 393 16. Bagci, S.A., Ekiz, H., Yilmaz, A., and Cakmak, I. (2007) Effect of zinc deficiency and
394 drought on grain yield of field-grown wheat cultivars in Central Anatolia. *Journal of*
395 *Agronomy and Crop Science* **193**: 198-206.
- 396 17. Graham, W.A., and McDonald, G.K. (2001) Effects of zinc on photosynthesis and yield of
397 wheat under heat stress. In: *Proceedings of the 10th Australian Agronomy Conference*,
398 Hobart.
- 399 18. Genc, Y., McDonald, G.K., and Graham, R.D. (2006) Contribution of different mechanisms
400 to zinc efficiency in bread wheat during early vegetative stage. *Plant and Soil* **281**: 353-
401 367.
- 402 19. NFDC (1998) Micronutrients in Agriculture: Pakistan Perspective. NFDC Publication No.
403 4/98, Islamabad.
- 404 20. Rafique, E., Rashid, A., Ryan, J., and Bhatti, A.U. (2006): Zinc deficiency in rain-fed wheat
405 in Pakistan: Magnitude, spatial variability, management, and plant analysis diagnostic
406 norms. *Communications in Soil Science and Plant Analysis* **37**: 181-197.
- 407 21. Erenoglu, B., Cakmak, I., Romheld, V., Derici, R., and Rengel, Z. (1999) Uptake of zinc by
408 rye, bread wheat and durum wheat cultivars differing in zinc efficiency. *Plant Soil* **209**: 245-
409 252.
- 410 22. Gupta, V.K., and Mehla, D.S. (1993) Depletion of micronutrients from soil and their uptake
411 in rice-wheat rotation. *Journal of Indian Society of Soil Science* **4**: 704-706.
- 412 23. Rerkasem, B., and Jamjod, S. (2004) Boron deficiency in wheat: a review. *Field Crops*
413 *Research* **89**: 173-186.

- 414 24. Snowball, K., and Robson, A.D. (1983) Symptoms of nutrient deficiencies:
415 Subterranean clover and wheat. Institute of Agriculture, University of Western Australia:
416 Nedlands, Western Australia.
- 417 25. Dell, B., and Haug, L.B. (1997) Physiological response of plants to low boron. *Plant and*
418 *Soil* **193**: 103-120.
- 419 26. Nayyar, V.K., Arora, C.L., and Kataki, P.K. (2001) Management of Soil Micronutrient
420 Deficiencies in the Rice-Wheat Cropping System. **P. 87-131**. In: Kataki, P.K. (ed.). *The Rice-*
421 *Wheat Cropping Systems of South Asia: Efficient Production Management*, Food Products
422 Press, New York, USA.
- 423 27. Takkar, P.N. (1982) Micronutrients: Forms, Contents Distribution in Profile, Indices of
424 Availability and Soil Test Methods. **p. 361-391**. In: *Reviews of Soil Research in India*.
425 Part 1. 12th International Congress of Soil Science, New Delhi, India.
- 426 28. Singh, M.V. (1999) Micronutrient Deficiency Delineation and Soil Fertility Mapping.
427 In: *National Symposium on Zinc Fertilizer Industry - Whither To* (Ramendra Singh
428 and Abhay Kumar, eds). Session II.
- 429 29. Jackson, M.L. (1967) *Soil Chemical Analysis*. Prentice-Hall of India, New Delhi.
- 430 30. Nelson, D.W., and Sommers, L.E. (1982) Total carbon, organic carbon and organic matter.
431 In: *Methods of Soil Analysis (Page et al. eds.)*, part 2, second edition, Agronomy Monograph
432 9, ASA and SSSA, Madison, Wisconsin. p. 539-594.
- 433 31. Dewis, J., and Freitas, F. (1984) Physical and Chemical Methods of Soil and
434 Water Analysis. Oxford and IBH Pub. Co., New Delhi. p. 51-106.
- 435 32. Subbiah, B.V., and Asija, G.L. (1956) A rapid procedure for the determination of available
436 nitrogen in soils. *Current Science* **25**: 259-260.
- 437 33. Bray, R.H., and Kurtz, L.P. (1945): Determination of total, organic and available forms of
438 phosphorus in soils. *Soil Science* **59**: 39-45.
- 439 34. Black, C.A. (1965) Methods of soil analysis. Part II, Agronomy Ser. No. 9. American
440 Society of Agronomy Inc. Madison, Wisconsin, U.S.A.
- 441 35. Lindsay, W.L., and Norvell, W.A. (1978) Development of DTPA soil test for zinc, iron,
442 manganese and copper. *Soil Science Society of America Journal* **42**: 421-428.
- 443 36. Saha, A.K. (2008) Methods of Physical and Chemical Analysis of Soil. Kalyani Publishers.
444 P. 1-469.
- 445 37. Chaudry, E.H., Timmer, V., Javed, A.S., and Siddique, M.T. (2007) Wheat response to
446 micronutrients in rainfed areas of Punjab. *Soil & Environment* **26**: 97-101.
- 447 38. Topal, A., Gezgin, S., Akgun, N., Dursun, N., and Babaoglu, M. (2002) Yield and yield
448 attributes of durum wheat (*Triticum durum* Desf.) as affected by boron application. In:
449 Goldbach, H.E., Rerkasem, B., Wimmer, M.A., Brown, P.H., Thellier, M., Bell, R.W.
450 (Eds.), *All Aspects of Plant and Animal Boron Nutrition*.
- 451 39. Marten, J.M., and Westermann, D.T. (1991) Fertilizer Applications for Correcting
452 Micronutrient Deficiencies. In *Micronutrients in Agriculture*, Mordtvedt; Mordtvedt, J.J.,

Comment [MA18]: P or p? Ed or ed?

Formatted: Highlight

Comment [MA19]: P or p?

Formatted: Highlight

Comment [MA20]: capital letter or not?

Comment [MA21]: capital letter or not?

Formatted: Highlight

- 453 | Giordano, P.M., Lindsay, W.L. (eds.) Soil Science Society of America Incorporation:
454 Madison, WI. 549–592.
- 455 40. Kutman, U. B., Yildiz, B., Ozturk, L., Cakmak, I. (2010) Bio-fortification of durum wheat
456 with zinc through soil and foliar applications of nitrogen. *Cereal Chemistry* **87**: 1–9.
- 457 41. Mandal, A.B. (1989) *Bangladesh Journal of Botany* **18**: 247 – 252.
- 458 42. Hosseini, S.M., Maftoun, M., Karimian, N., Rounaghi, A., Emam, Y. (2007) Effect of zinc
459 × boron interaction on plant growth and tissue nutrient concentration of corn. *Journal of*
460 *Plant Nutrition* **30**: 773–781.
- 461 43. Ozturk, L., Yazici, M.A., Yucel, C., Torun, A., Cekic, C., Bagci, A., Ozkan, H., Braun, H.J.,
462 Sayers, Z., and Cakmak, I. (2006) Concentration and localization of zinc during seed
463 development and germination in wheat. *Physiology of Plant* **128**: 144–152.
- 464 44. Sakal, R., Singh, A.P., and Singh, S.P. (1988) Distribution of available Zinc, Copper, Iron
465 and Manganese in Old Alluvial soils as related to certain soil characteristics. *Journal of*
466 *Indian Society of Soil Science* **36**: 59–63.
- 467 45. Santra, G.H., Mandal, B.K., and Das, D.K. (1989) *Environmental Ecology* **7**: 874 – 877.

Comment [MA22]: Capital letter or not?

Formatted: Highlight