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LONG-TERM EFFECTS OF STRAW AND MANURE ON CROP MICRONUTRIENT NUTRITION UNDER A WHEAT-MAIZE CROPPING SYSTEM

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□ *Management practices have significant effects on crop micronutrient contents. This study examined effects of applying chemical fertilizers of nitrogen (N), phosphorus (P), and potassium (K) (NPK), alone or supplemented with straw or manure, under a wheat-maize cropping system in a 18-year experiment, on the crops' iron (Fe), manganese (Mn), copper (Cu), and zinc (Zn) contents throughout the crops' development. The micronutrient contents of both wheat and maize were above critical values during vegetative development, but Zn contents of maize ear leaves were sub-sufficient under all treatments. The wheat grain Mn, Cu, and Zn contents were lower under fertilized treatments than in unfertilized controls. Nutrient balance calculations showed that NPK application alone or with straw resulted in deficits of the four micronutrients, but not application of NPK supplemented with manure. Hence, application of micronutrients, such as Zn, through organic or inorganic fertilizers is recommended for this cropping system.*

Keywords: iron, manganese, copper, zinc, nutrient balance

INTRODUCTION

Sufficient levels of micronutrients in crops are essential for high yields, product quality, pathogen protection, and seed vigor (Kirchmann et al., 2009). Moreover, cereals (and other crops) are important sources of mineral micronutrients for animals and humans. However, their concentrations in

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crops are dependent on soil type, nutrient availability, crop species, and to a lesser extent, season and cultivars (Ascher et al., 1994). Hence, since fertilization can substantially improve soils' micronutrient status (Prasad and Sinha, 1982; Wei, et al., 2006; Li et al., 2007), and thus crop micronutrient nutrition and accumulation (Ascher et al., 1994; Li et al., 2006), it is an important practice in crop production.

Winter wheat-summer maize rotation is the principal agricultural production system in the northern part of China; around 16 million ha are cultivated in this fashion, accounting for about a quarter of the total national food production. According to Zou et al. (2008), soils covering up to 48.6 and 20.3 Mha are deficient in zinc (Zn) and manganese (Mn), respectively, mainly calcareous soils in the northern part of the country. Fe deficiency is also widespread in these calcareous soils [diethylenetriaminepentaacetic acid (DTPA)-extractable Fe < 4.5 mg kg^{-1}] (Zou et al., 2008). Furthermore, Tong et al. (2004) and Niu and Zhang (2010) found, in surveys, that most farmers in the region only applied nitrogen (N) and phosphorus (P) chemical fertilizers in their winter wheat-summer maize rotations, without organic manure, although some farmers have recently started to incorporate crop residues into soil after harvests (Tong, 2010; Kong and Zhao, 2011).

Long-term experiments are particularly valuable when examining management sustainability, since they can highlight problems associated with management practices that may threaten future productivity (Berzsenyi et al., 2000). Hence, much attention has focused on long-term effects of using chemical fertilizers and cropping on soil properties (Zhang et al., 2006; Blair et al., 2006; Hati et al., 2007; Yang et al., 2010; Yang et al., 2011b; Yang et al., 2012). However, limited attention has been paid to the associated changes in crop micronutrient status. Previously, we have shown that fertilization can improve the DTPA-extractable iron (Fe) content of soils under maize-wheat rotations in northern China, but chemical fertilizers alone or combined with crop residue or organic manure only maintain adequate levels of DTPA-extractable Mn and Zn in their upper 10 cm layers, not in subsoil layers. Therefore, the purpose of this study was to examine the effects of long-term applications of chemical fertilizers (alone or supplemented with either crop residue incorporation or organic manure) on the micronutrient balances of the soil and the crops' micronutrient status throughout their development (including final levels in grain).

MATERIALS AND METHODS

Study Site and Experimental Design

A long-term experiment was established in October 1990 at the Chinese National Soil Fertility and Fertilizer Efficiency Monitoring Base of Loess Soil (N 34°17′51″, E 108°00′48″, altitude 524.7 m a.s.l.), at a site located on leveled land of the third terrace of the Weihe River, a tributary of the Yellow River, Yangling, Shaanxi, China. According to the United States Department of Agriculture (USDA) texture classification system, the soil at the site is a silt clay loam (clay 32%, silt 52% and sand 16%; Eumorthic Anthrosol, FAO) derived from loess materials. On average, at the time of establishment, the soil at the site contained 7.44 g kg^{-1} organic carbon (C), 0.93 g kg^{-1} total N, 9.57 mg kg^{-1} Olsen P, 191 mg kg^{-1} exchangeable potassium (K), 92.5 g kg^{-1} calcium carbonate ($CaCO_3$) and had a mean pH of 8.62 across all plots, with low variability (C.V. $\leq$ 6%, except for Olsen-P, 15%, Yang et al., 2012). The experimental site has a mean annual temperature of 13.0°C, and mean annual precipitation of ca. 550 mm, which mainly falls from June to September.

All treatments were applied to plots in which winter wheat (*Triticum aestivum* L.) and summer maize (*Zea mays* L.) were grown under a double crop rotation every cropping year. The experiment included nine nutrient management regimes in total; details of the experimental design have been previously described by Yang et al. (2012). Four nutrient management regimes were evaluated in this study: (1) no added fertilizer or manure (the control treatment, henceforth CK), (2) combined applications of N, P and K inorganic fertilizers (NPK), (3) NPK plus wheat or maize straw (SNPK), and (4) NPK plus dairy manure (MNPK).

For winter wheat, the inorganic N, P, and K application rates were 165, 57.6, and 68.5 kg ha^{-1} a^{-1}, respectively, in the NPK and SNPK treatments. In the MNPK treatment, in which 70% N was supplied by dairy manure and the remaining 30% N by inorganic fertilizer, application rates of total N, inorganic P, and K were as 1.5 times greater than those in the NPK treatment. In addition, the dairy manure supplied 159 and 209 kg ha^{-1} a^{-1} P and K, respectively. For summer maize, the same amounts of inorganic N, P, and K were applied in each of the fertilizer (NPK, SNPK, and MNPK) treatments: 187.5, 24.6, and 77.8 kg ha^{-1} a^{-1}, respectively. The SNPK plots also received 4500 kg (air-dried) wheat straw ha^{-1} annually between 1990 and 1998, and then from 1999 until the end of the experiment they received the above-ground parts of the maize straw harvested in the preceding season (mean weight 4392 kg ha^{-1}, ranging from 2630 to 5990 kg ha^{-1}). The added straw was manually chopped into small pieces ca. 3 cm long, and incorporated into the soil in autumn, before the winter wheat was sown. Similarly, dairy manure with a mean dry weight of 20.0 t ha^{-1} was added once per year, immediately before the sowing of the wheat. All inorganic fertilizers and organic materials applied were incorporated into the soil to plowing depth (ca. 20 cm) before the winter wheat was sown and in the inter-row areas about a month after maize was planted. The N-containing inorganic fertilizer used in the experiment was urea, P was added as single super-phosphate, and K as potassium sulfate. The Fe, Mn, copper (Cu), and Zn contents of the P fertilizer were 6078, 913, 20, and 157 mg kg^{-1}, respectively. The micronutrient contents of the K fertilizer, manure, wheat straw and maize straw were: 84, 0, 0, and 0 mg kg^{-1} (Fe), 6247, 586, 37, and 170 mg kg^{-1} (Mn), 286, 31, 1.2, and 5.9 mg kg^{-1} (Cu), and 474, 44, 7.8, and 18.8 mg kg^{-1} (Zn), respectively. Winter wheat was sown in October and harvested in the following June; summer maize was planted and harvested about three months later, at the end of September or in early October. The plots were irrigated with ground water once or twice during the winter wheat season and 0 to 3 times during the summer maize season, as required, applying approximately 90 mm of water on each occasion. All above-ground crop residues were removed after harvesting, unless otherwise specified. The fields were conventionally tilled with a rototiller.

Plant Sampling and Analyses

Samples (three replicates $treatment^{-1}$) of both wheat and maize plants were collected on three occasions during their growth courses in 2008. On the first occasion, before shooting, 0.2 m^2 of wheat seedlings or five maize plants were randomly selected and cut close to the ground, and pooled per replicate. On the second occasion, at the grain-filling stage, the flag leaf was collected from 30 randomly selected wheat stems, or the ear leaf of 20 maize plants per replicate. On the third occasion, at crop maturity, 8 and 20 m^2 of wheat and maize crops, respectively, were harvested manually with sickles cutting close to the ground per replicate to estimate their yields. Subsamples of wheat and maize grain and straw were then separated. In each case, the sampled material was washed with distilled water, dried at 70–80°C for 72 h, ground to pass through a 0.25 mm nylon sieve then digested with concentrated nitric acid/perchloric acid (HNO_3/ $HClO_4$), and their Fe, Mn, Cu, and Zn contents were analyzed using a Varian (Varian Medical Systems, Palo Alto, CA, USA) inductively coupled plasma atomic emission spectrometer, following Emteryd (2002).

Micronutrient Input and Output Calculations

The amounts of micronutrients added to the soil in the fertilizer treatments were calculated by adding the amounts in the fertilizers applied per unit area in 2007–2008 and, for the SNPK and MNPK treatments, in the maize straw returned to the field or supplementary manure. Micronutrient inputs from atmospheric deposition were also measured at the site, but the inputs from irrigation water at the site were very minor (data not shown), and thus ignored. Output amounts of micronutrients were calculated by multiplying the biomass of the aboveground parts (grain and straw of both wheat and maize) per unit area by their micronutrient contents, and the elemental field balances were calculated as the differences between element inputs and outputs.

Statistical Analyses

One-way analysis of variance was used to analyze differences in tested parameters among the treatments, and the least significant difference method (LSD) was used to test the significance, at the 5% probability level, of differences between treatment means (SPSS Inc., Chicago, IL, USA).

RESULTS AND DISCUSSION

Micronutrient Concentrations of Crops in the Vegetative Phase

The Fe concentrations in the wheat samples taken in the vegetative phase ranged from 556 to 860 mg kg^{-1} (Table 1), and no significant difference was observed among treatments, but their Mn concentrations were significantly higher under the control (CK) treatment than under the fertilizer-added treatments. There was no significant difference in the Cu concentrations of the wheat plants between treatments, while their Zn concentrations ranged from 16.4 to 31.6 mg kg^{-1}, and were significantly lower under the NPK than under the CK and MNPK treatments.

The Fe concentrations in the maize samples in this phase ranged from 172 to 218 mg kg^{-1} (Table 2), and did not significantly differ among treatments, but their Mn concentrations were significantly lower under MNPK than under the other treatments. There was no significant difference in the Cu concentrations among treatments, but the Zn concentration in the maize plants was significantly higher under CK than under the fertilizer-added treatments.

TABLE 1 Micronutrient contents in wheat plants in 2008

Growth stage	Treatment	Fe	Mn	Cu	Zn
Before shooting (whole plant)	CK	713 a	137 a	7.19 a	30.0 a
	NPK	556 a	81.0 b	6.04 a	16.4 b
	SNPK	781 a	87.5 b	6.75 a	21.9 ab
	MNPK	618 a	77.0 b	6.80 a	31.6 a
Grain-filling (flag leaf)	CK	325 a	48.2 c	4.74 a	18.7 a
	NPK	349 a	110 a	5.45 a	18.9 a
	SNPK	299 a	112 a	5.32 a	18.6 a
	MNPK	301 a	87.2 b	4.00 a	19.6 a
Maturity (grain)	CK	52.9 a	53.5 a	7.01 a	55.5 a
	NPK	46.9 a	46.0 b	6.85 a	24.4 b
	SNPK	58.3 a	47.8 b	6.31 ab	24.4 b
	MNPK	75.5 a	45.2 b	5.07 b	23.9 b
Maturity (straw)	CK	335 a	48.6 a	3.28 a	13.4 a
	NPK	306 a	43.2 a	2.69 a	7.55 b
	SNPK	372 a	50.4 a	2.41 a	7.33 b
	MNPK	361 a	46.4 a	3.46 a	10.4 ab

Different letters within columns indicate significant differences between treatments at $P < 0.05$.

TABLE 2 Micronutrient contents in the maize plants in 2008

Growth stage	Treatment	Fe	Mn	Cu	Zn
Before shooting (whole plant)	CK	218 a	63.3 a	8.01 a	34.1 a
	NPK	192 a	66.1 a	8.31 a	21.1 b
	SNPK	188 a	62.5 a	8.39 a	18.5 b
	MNPK	172 a	52.5 b	8.30 a	15.5 b
Grain-filling (ear leaf)	CK	152 c	38.5 b	5.34 c	14.9 b
	NPK	182 b	53.6 a	9.78 a	9.4 c
	SNPK	191 ab	58.4 a	8.52 b	17.4 ab
	MNPK	208 a	52.6 a	9.58 ab	18.6 a
Maturity (grain)	CK	29.3 a	3.73 a	1.71 a	19.0 a
	NPK	27.0 a	4.90 a	1.92 a	17.4 a
	SNPK	24.0 a	3.90 a	1.83 a	19.7 a
	MNPK	31.8 a	4.13 a	1.50 a	16.2 a
Maturity (straw)	CK	635 a	46.0 a	7.38 b	34.6 a
	NPK	475 b	59.4 a	9.83 a	15.7 b
	SNPK	372 b	45.0 a	7.04 b	13.8 b
	MNPK	413 b	53.7 a	6.88 b	13.0 b

Different letters within columns indicate significant differences between treatments at $P < 0.05$.

According to Campbell (2009), the critical concentrations of Fe, Mn, Cu, and Zn in wheat plants at this stage are 25, 15, 3, and 15 mg kg^{-1}, respectively, and sufficiency ranges of these micronutrients in maize plants are 40–250, 25–160, 6–20, and 20–60 mg kg^{-1}, respectively. The contents of the wheat plants were higher than these critical values under all treatments, indicating that no deficiency occurred during the vegetative stage, although the soil analysis indicated that available micronutrient contents were in their respective marginal ranges (Yu et al., 1984) in the soil's surface or subsurface layers. However, under SNPK and MNPK the Zn concentrations of the maize plants were below the sufficiency range, possibly due to its high biomass (Table 3) and low level of available Zn level in the soil profile.

TABLE 3 Wheat and maize yields in 2008 (kg ha^{-1})

	Wheat		Maize	
Treatment	Grain	Straw	Grain	Straw
CK	895 c	1956 c	1907 c	1651 c
NPK	6125 b	8232 b	5941 b	5620 b
SNPK	6768 ab	9859 a	6575 b	5990 b
MNPK	7010 a	10527 a	7810 a	6919 a

Different letters within columns indicate significant ($P < 0.05$) differences among treatments.

Micronutrient Concentrations of Crops at the Grain-Filling Stage

The Fe concentrations in the sampled wheat flag leaves ranged from 299 to 349 mg kg^{-1} (Table 1), and did not significantly differ among treatments. Concentrations of Cu and Zn were also similar among treatments. However, the Mn concentrations in wheat plant significantly differed among treatments, decreasing in the order CK < MNPK<NPK, SNPK.

The Fe concentrations in maize ear leaves ranged from 152 to 208 mg kg^{-1} (Table 2), and were significantly lower under CK than under the fertilizer-added treatments. The Mn and Cu concentrations in maize ear leaves were also significantly higher under the fertilizer-added treatments relative to CK. In contrast, the Zn concentrations in the maize ear leaves, which ranged from 9.4 to 18.6 mg kg^{-1}, were lowest under NPK and highest under MNPK.

The sufficiency concentrations of Fe, Mn, Cu, and Zn are 30–200, 20–150, 4.5–15, and 18–70 mg kg^{-1}, respectively, in wheat flag leaves, and 30–250, 15–150, 5–25, and 20–70 mg kg^{-1} in maize ear leaves, according to Campbell (2009). In comparison, the Cu content of the wheat flag leaves was insufficient under the MNPK treatment (Table 1), although soil available Cu contents in the examined 60 cm soil profile were higher than the marginal range, possibly due to the large biomass yield under MNPK. In addition, Zn contents of the maize ear leaves were insufficient under all treatments, although their Zn contents were just above the critical value of 15 mg kg^{-1} (Campbell, 2009) under the SNPK and MNPK treatments, again possibly due to the high biomass and lower levels of available Zn level in the soil under these treatments.

Micronutrient Concentrations in Grain and Straw

The Fe concentrations in the wheat grain and straw ranged from 52.9 to 75.5 mg kg^{-1} and 306 to 372 mg kg^{-1}, respectively (Table 1), and no significant difference in them was observed among treatments. The Mn concentration in wheat straw was similar among treatments, but in wheat grain it was significantly lower under the fertilizer-added treatments relative to CK. There was no significant difference in the Cu concentrations of wheat straw between treatments, or in those of wheat grain between SNPK and the other treatments, but they were significantly lower under MNPK than under CK and NPK. The Zn concentration in wheat straw ranged from 7.33 to 13.4 mg kg^{-1}, and was highest under CK. Similarly, the Zn concentration in wheat grain was significantly higher under CK than under the fertilized treatments.

Shi et al. (2009) found that the Fe, Mn, Cu and Zn concentrations in grain of 262 Chinese wheat genotypes grown under same soil and climatic condition were 34.2–61.2, 20.9–56.7, 3.4–9.8, and 26.3–76.0 mg kg^{-1}, respectively. Generally, the wheat grain Fe, Mn, and Cu concentrations found in the present study were within the above ranges, but the Zn concentrations under the fertilized treatments were slightly lower (Table 1), possibly due to differences in varieties and soil conditions (Zhao et al., 2009). In the "UN-Biofortification Program", a concentration of 60 mg Zn kg^{-1} grain is targeted to counteract zinc deficiency among the population in many parts of the world. Hence, management practices such as application of Zn-containing fertilizer is recommended to improve the grain Zn concentration under the current cropping system (Yang et al., 2011a). The similarity of wheat grain Fe concentrations among treatments could be explained by their sufficiency (and similarity) during the plants' vegetative growth and reproductive growth phases under all treatments (Table 1). The higher wheat grain Mn, Cu, and Zn concentrations under CK than the fertilizer-added treatments might be related to associated differences in the root to shoot ratio. This is because these elements require mobilization by roots; hence the size of the root system relative to the above ground biomass can affect their concentrations in crops (Kirchmann et al., 2009). Unfertilized crops generally have higher root to shoot biomass ratios than fertilized crops, and can thus take up more nutrients through their roots.

The concentrations of the studied micronutrients in maize grain were all similar among treatments (Table 2). In addition, the Mn concentration did not significantly differ among treatments in maize straw. However, in maize straw the Fe concentrations were significantly higher under CK than under the fertilizer-added treatments, there were significant differences in its Cu concentrations between NPK and the other treatments, and its Zn concentrations were significantly higher under CK relative to the fertilizer-added treatments.

As well as being similar among treatments, the maize grain concentrations of Fe, Mn, Cu, and Zn were similar to those found by Berenguer et al. (2008), Li et al. (2007), Yu et al. (2011) and surveys of maize grown in the Hebei province of China (Jia et al., 2009). However, the Cu concentrations were clearly too low to meet the physiological requirements of ruminant animals (9–11 mg kg^{-1}; McDowell, 2003), which is a source of concern since maize grain is commonly used for feeding animals in the study region. The general lack of apparent effects of the tested fertilization practices on maize grain micronutrient concentrations can be ascribed to the similarity of their contents from the seedling through grain-filling stages (Table 2). The exception to this pattern (significantly lower maize grain Zn concentrations under fertilized treatments than CK) can be explained by lower root to shoot ratios under the former treatments than under CK, as described above.

Crop Yields in 2007–2008

Wheat grain and straw yields ranged from 895 to 7010 kg and 1956 to 10527 kg ha^{-1}, respectively, while maize grain and straw yields ranged from

1907 to 7810 kg and 1651 to 6919 kg ha^{-1}, respectively (Table 3). All of the fertilization treatments significantly increased wheat and maize yields compared to the unfertilized controls. In addition, the MNPK treatment resulted in significantly higher wheat and maize yields than NPK, and significantly higher maize yields than SNPK. This was probably due to a pronounced improvement of soil physical and biochemical properties due to the supplementary manure in the MNPK treatment (Zhang et al., 2006; Yang et al., 2010; Li et al., 2011), since N, P and K application rates in the NPK treatment in the present study were similar to the local recommended amounts for this soil (Fu et al., 2009, 2010). Although Zn concentrations were subsufficient under all treatments at the grain-filling stage for maize, they were significantly lower under NPK than under MNPK (Table 2), which might have been partially responsible for the lower maize yields under NPK than under MNPK. Both wheat and maize yields were similar under the SNPK and NPK treatments, except that the wheat straw yield was significantly higher under SNPK than under NPK. The generally minor effect of SNPK on crop yields over NPK might be related to the low rates of straw incorporation (less than 4.5 t ha^{-1}), and consequently weaker effects on soil properties than manure addition (Yang et al., 2010; Li et al., 2011). Similar results have also been reported under a rice-wheat cropping system by Yadvinder-Singh et al. (2004).

Micronutrient Uptake and Balance

Total amounts of Fe and Mn in wheat biomass (grain plus straw) were significantly higher under the fertilizer-added treatments than under CK. Furthermore, the MNPK and SNPK treatments resulted in higher grain and straw biomass yields, high concentrations of these elements, and (hence) markedly higher uptake of Fe than NPK (Table 4). The total amount of Cu in wheat biomass was also significantly higher under the fertilizer-added treatments than under CK. Total amounts of Zn in wheat biomass were similar between SNPK and NPK, and in both cases significantly higher than under CK, but significantly lower than under MNPK.

Total amounts of Fe, Cu and Zn in maize biomass were significantly higher under the fertilizer-added treatments than under CK, and there were no differences in these respects between the fertilizer-added treatments. Total amounts of Mn were similar between SNPK and NPK, MNPK and NPK, but Mn uptake was significantly higher under MNPK than under SNPK.

According to the calculated micronutrient inputs and crop uptake rates in 2007–2008, micronutrient inputs through MNPK were sufficient to maintain current yields of this cropping system (Table 4). However, combining retention of a single season's straw with NPK (SNPK) was not sufficient to balance the crop uptake of the four micronutrients, although deficits could be alleviated to some extent compared with NPK. However, some farmers

TABLE 4 Fe, Mn, Cu and Zn nutrient balances under the wheat-maize system in 2007–2008 (kg ha^{-1})

		Input			Output (crop removal)		
Element	Treatment	Chemical fertilizer	Organic material	Deposition	Wheat	Maize	Balance (input-output)
Fe	CK	0	0	0.004	0.602c	1.105b	−1.702
	NPK	4.188	0	0.004	2.806b	2.830a	−1.417
	SNPK	4.188	2.080	0.004	4.062a	2.386a	−0.176
	MNPK	6.282	124.3	0.004	4.329a	3.104a	123.2
Mn	CK	0	0	0.003	0.128c	0.083c	−0.208
	NPK	0.625	0	0.003	0.637b	0.363ab	−0.372
	SNPK	0.625	0.224	0.003	0.820a	0.295b	−0.264
	MNPK	0.938	11.632	0.003	0.805a	0.404a	11.381
Cu	CK	0	0	0.005	0.012b	0.015b	−0.022
	NPK	0.014	0	0.005	0.064a	0.067a	−0.112
	SNPK	0.014	0.034	0.005	0.066a	0.054a	−0.068
	MNPK	0.021	0.737	0.005	0.072a	0.059a	0.629
Zn	CK	0	0	0.003	0.072c	0.093b	−0.162
	NPK	0.107	0	0.003	0.212b	0.192a	−0.293
	SNPK	0.107	0.085	0.003	0.237b	0.212a	−0.254
	MNPK	0.161	3.377	0.003	0.277a	0.216a	3.047

Different letters within columns indicate significant differences between treatments at $P < 0.05$.

have reportedly incorporated two seasons' crop straw (Tong, 2010; Kong and Zhao, 2011), which could add 3.668, 0.497, 0.024, and 0.072 kg ha^{-1} more Fe, Mn, Cu and Zn to the system, and based on current yields under the SNPK treatment this could balance inputs and outputs of all the micronutrients except Zn. Application of chemical NPK fertilizer alone resulted in negative balances of all four micronutrients. Yu et al. (2011) found balances of the four elements to be positive under all treatments they considered in a single cropping system in northeast China. However, this was mainly due to high inputs through deposition (3.720, 1.112, 0.142, and 0.544 kg ha^{-1} a^{-1} of Fe, Mn, Cu, and Zn, respectively), while in the system we examined deposition was negligible (Table 4). Considering the soil available micronutrient levels and nutrient balances, additions of micronutrients, such as Zn, through applications of organic or inorganic fertilizers, is recommended under the wheat-maize cropping system to improve both grain quantity and its mineral nutrition quality.

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