Soil Micronutrient Status and Iron and Zinc Availability for Haricot Bean Production in Southern Ethiopia

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Abstract

The study was conducted to assess micronutrients status and evaluate the availability of zinc and iron for haricot bean production in soils of Taba, Huletegna Choroko, Tenkaka Umbulo, Kontela, Alage and Jole Andegna, southern Ethiopia. Soil profiles were opened, and soil samples were collected from each horizon to analyze pH, organic carbon, cation exchange capacity, and available micronutrients. A greenhouse experiment was conducted in soils collected from the above sites using the following treatments: no fertilizer; application of N and P; application of N, P and Zn; application of N, P, and Fe; application of N, P, Zn and Fe; application of Zn only; application of Fe only. Sources of Zn, Fe, N and P were ZnSO₄.7H₂O (21% Zn), FeSO₄.7H₂O (20% Fe), urea and TSP, respectively. Haricot bean variety Nasir was planted in a pot filled with 5 kg soil. $ZnSO_4$ ·7H₂O and FeSO₄.7H₂O were sprayed on the leaves, and urea and TSP were applied to the soil just before planting. Four haricot bean seeds per pot were sown and, later, thinned to two seedlings. Grain yield, plant height, number of pods per plant and number of seeds per pod were collected. Leaves and seeds were analyzed for micronutrients using Microwave Plasma Atomic Emission Spectrometer (MPAES). The soils of the study areas have different textural classes varying from sand to clay with pH>7.4. Iron (Fe), zinc (Zn) and copper (Cu) were low in Tenkaka Umbulo and Huletegna Choroko soils whereas manganese (Mn) was high at the surface and low in subsurface horizons. In Taba soils, Cu was deficient throughout the profiles whereas Fe, Zn and Mn were high in the surface and low in subsurface horizons. Soils of Jole Andegna contained low Fe, and high Cu and Mn whereas Zn content was high in the surface but low in the subsurface horizons. Application of Zn with NP significantly increased grain yield compared to the no fertilizer treatment. Application of Fe and Zn either alone or combined with NP produced significantly higher tissue concentrations of the minerals than the other treatments. Therefore, fertilization of haricot beans with Fe and Zn is important in the soils of the study sites especially for quality production in terms of Zn and Fe in seeds.

Keywords: Fe concentration, Haricot bean, NPZnFe, Zn concentration, micronutrients

Introduction

Micronutrients are essential elements for the normal growth of plants and are used in small quantities, with uptake less than 1 pound per acre (1.1 kg per ha) per year, except iron, hence, the term "micro is used" (Straaten, 2002; Wortmann *et al.*, 2008). In spite of this low requirement, critical plant functions are limited if micronutrients are unavailable, resulting in plant abnormalities, reduced growth, lower yields (Rehm, 1997). Aref (2012) stated that the critical levels of Fe, Zn, Cu and Mn in soil are 5.0, 1.5, 0.5 and 1.0 mg kg⁻¹, respectively. A study conducted in different soils of Ethiopia indicated that the critical levels of Cu, Zn, Mn and Fe are 0.2, 0.5 - 1.5, 2.9 - 3.3, and 2.5 - 4.5 mg kg⁻¹, respectively (Haque *et al.*, 2000). According to Westfall and Bauder (2011), Zn contents of soil ranging from 0-0.9, 1-1.5, and > 1.5mg kg⁻¹ are low, marginal, and adequate, respectively, for haricot bean production. Prakash *et al.* (2014) reported that soils that test less than 4.5 mg kg⁻¹ Fe are deficient in this element.

Micronutrients also have negative effects on human nutrition and health. The World Health Organization (WHO) has estimated that over three billion people in the world suffer from micronutrient malnutrition (WHO, 2002; Long *et al.*, 2004) and the most commonly deficient elements in the diet of humans are Fe and Zn (Franca and Ferrari, 2002). Together with protein deficiencies, shortages of Fe and Zn are prevalent in food in sub-Saharan Africa (SSA) (Welch and Graham, 2004). Adding Zn and Fe to soils and crops can make a significant contribution towards goals of increased food security and human health in a sustainable manner (International Zinc Association, 2010). The addition of haricot bean can improve Zn and Fe contents of cereal and tuber-based diets as they provide high concentration of Zn (25-50 mg kg⁻¹) (FAO, 2001) and all of the Fe that humans require (Nchimbi-Msolla and Tryphone , 2010).

Haricot bean (*Phaseolus vulgaris L.*) is one of the principal food and cash crop grown throughout the world (Tryphone and Nchimbi-Msolla, 2010) especially in developing countries (Prolla et al., 2010). It is the world's second most important commercial legume crop next to soybean being produced in significant amount in Africa (CIAT, 2008). Haricot bean provides an essential part of the daily diet and foreign earnings for Ethiopia (Girma, 2009; CARE Ethiopia, 2011). Haricot beans are produced in almost all the regional states of Ethiopia the major production areas being central, eastern and southern parts of the country (CSA, 2011) with average annual national production of 150 thousand tons (Ferris and Kaganzi, 2008). Though production is large, the national average yield is only 1.167 t ha⁻¹ (CSA, 2011) and this is attributed to various constraints among which

low soil nutrients content especially micronutrients could be mentioned (Ferris and Kaganzi, 2008; Girma, 2009). Singh (2004) reported that bean is highly sensitive to the deficiency of Fe and Zn. In general, Fe and Zn may be needed for haricot bean where the soil is calcareous (pH greater than 7.3 because of excess free lime), coarse textured, low organic matter content, and organic soils (Ronan, 2007; Wortmann et al., 2012). The critical level of zinc in bean tissue is 15 to 20 mg kg⁻¹, while the critical level of Fe is 100 mg kg⁻¹(Ronan, 2007). Fageria et al. (1990) indicated the sufficiency range of Fe for haricot bean tissue is from100-450 mg kg⁻¹. Improving the nutritional quality of haricot beans such as the Fe and Zn content is important in sustaining health of people relying on this crop (Ndakidemi et al., 2011). Application of around 1.5 to 2% solution of ferrous sulphate heptahydrate (FeSO₄.7H₂O) may be considered the best way to correct iron deficiency in haricot beans (Ferguson, 2006; Moore et al., 2012). McKenzie (1992) recommended that several applications of foliar sprays of 0.5% zinc sulphate heptahydrate (ZnSO₄. 7H₂O) solution at a rate of 20 to 30 gallons per acre can supply sufficient zinc, although crop response to applied Fe and Zn may depend on the type of soil, crop and its variety, status of available nutrients, and severity of deficiency (Nayyar et al., 2001). Application of zinc caused significantly higher plant height of 53.24 cm, higher number of leaves per plant (27), maximum number of branches (12.33) (Hamsa and Puttaiah, 2012). Talukder et al. (2009) and Tryphone and Nchimbi-Msolla (2010) reported significant differences in concentrations of Fe and Zn in seeds and leaves of haricot bean.

Although haricot bean is produced in Alage, Zeway, Butajira, Halaba, Bodity and Hawassa areas, the yields of the crop in these areas are low, which might be attributed to environmental conditions including low soil fertility. Though some studied areas in Ethiopia showed that micronutrients are deficient, their status and availability to haricot bean production in soils of the aforementioned areas have not been studied. Therefore, it is important to assess the status of micronutrients and their availability to haricot bean production in the aforementioned areas.

Materials and Methods

The study was conducted in six locations in Southern part of Ethiopia, which include: Kontela with Luvic Calcisols (Siltic), Taba with Haplic Lixisols (Siltic) and Haplic Lixisols (Humic), Jole Andegna with Haplic Luvisols (Humic), Tenkaka Umbulo with Haplic Calcisols (Humic), Alage with Haplic Calcisols (Chromic) and Huletegna Choroko with Andic Lixisols (Humic) and Andic Cambisols (Humic). The physiographic settings of the study sites are indicated in Table 1. The dominant textural classes of the soils are loam and clay loam. Kontela and Alage are located in Oromiya region, whereas Taba, Jole Andegna, Tenkaka Umbulo and Huletegna Choroko are in Southern Nations, Nationalities and Peoples Regional State of Ethiopia.

Soil sampling and analysis

Two representative pits of 2 x 2 x 2 m were excavated at each location, except for Alage site where only one pedon was used. Soil samples were collected from each identified horizon and randomly from farmers' fields representing each type of the experimental soil from 0-15cm and 15-30cm depths with three replications using augur. For each replication 15 subsamples were collected and composited. The soil samples were air dried and ground to pass 2 mm sieve and analyzed for pH, organic carbon, cation exchange capacity and micro-nutrients status. The pH of the soils was determined in H_2O (pH- H_2O) 1:2.5 soil to solution ratio using a pH meter. Organic carbon content of the soils was determined following the wet combustion method of Walkley and Black, and the cation exchange capacity (CEC) of the soils were determined using the $1MNH_4OAc$ (pH 7) method as outlined by Sahlemedhin and Taye (2000). Available micronutrients (Fe, Mn, Zn, and Cu) contents of the soils were extracted by diethylene triamine pentaacetic acid (DTPA) method (Tan, 1996) and the contents in the extract were determined by AAS.

Location	Geographic	location	Slope (%)	Altitude
	Latitude	Longitude		(m.a.s.l)
Taba	07°01'01.9"	37 ⁰ 53'57.6"	3	1915
Huletegna Choroko	07°20'34.5"	38°06'30.0"	1	1807
Jole Andegna	08°12'25.9"	38°27'33.2"	9	1923
Kontela	07°58'09.7"	38°43'09.0"	4	1646
Alage	07°32'21.8"	38°24'51.3"	9	1600
Tankaka Umbulo	07°01'19.9"	38°20'23.6"	10	1717

Table 1.Location and physiographic settings of the study sites.

A greenhouse experiment was conducted to evaluate the status of Fe and Zn in soils collected from the above mentioned locations for haricot bean production. The experiment was conducted in a randomized complete design (CRD) with seven treatments and three replications. The treatments included: (1) control, no nutrient application; (2) application of only N and P; (3) application of N, P and Zn; (4) application of N, P, and Fe; (5) application of N, P, Zn and Fe (6) application of Zn only; (7) application of Fe only. Sources of Zn, iron, N and P were ZnSO₄.7H₂O (21% Zn), FeSO₄.7H₂O (20% Fe), urea and TSP, respectively. TSP and urea were

applied in the soil just before planting; 1% Zinc sulphate was sprayed two times after three and six weeks from sowing date (Abdel-Mawgoud et al., 2011) and 2% ferrous sulphate was sprayed 3 times in two weeks interval starting from 15 days after planting at volume of 100 L ha⁻¹. Two percent (2%) ferrous sulphate and 1% Zinc sulphate were prepared by adding 2 kg of iron sulphate and 1 kg of zinc sulphate to 100 litters of water, respectively (Incitec Pivot, 2003). Nutrients per hectare were converted into per pot assuming that the weight of 1 ha soil in 15 cm depth is 2,000,000 kg. Five kg of soil per pot was used for the experiment and the size of the pot was 19 cm height and 19 cm diameter at top, and 16 cm diameter at bottom. The amounts of P and N were 50 mg kg⁻¹ soil (0.25 g/pot) and 0.045g/pot, respectively. Four haricot bean seeds per pot were sown and later two seedlings were thinned at 10 days after sowing (Mourice and Tryphone, 2012). Haricot bean variety Nasir was used for this experiment. Grain yield, plant height, number of pods per plant and number of seeds per pod were collected.

Three fully developed leaves from the top of the plant during initial flowering and some seeds were collected from each treatment pot, dried in an oven at 70 °C for 24 hours, and ground using a sample rotating mill. The ground plant materials were digested and analyzed for their Zn and Fe contents with the following procedures. A sample of 0.5 g was weighed on a balance sensitive to the nearest 0.00001g and put into a digestion tube. Six milliliters of HNO₃ was added to each tube and placed in a digestion block at 90 °C for 45 minutes. Then, 5 ml of H_2O_2 was added in two splits (3 and 2 ml) while the samples were in the digestion block and digestion continued for another 65 minutes. Finally, 3 ml of 6 M HCl was added and the samples were digested until the solution had turned completely clear (about 5 minutes). Then, the tubes were removed from the block and cooled for 20 minutes, shaken using a vortex, and the digests were transferred from digestion tubes to dram vials and stored after the solution was brought to a volume of 25 ml with deionized water. The concentrations of Zn, Fe, Cu and Mn were analyzed using Microwave Plasma Atomic Emission Spectrometer (MPAES) at 213.857, 259. 940, 324.754 and 478.343 nm, respectively.

Analysis of variance (ANOVA) was carried out using Proc GLM procedures in the SAS 9.3 program (SAS Institute Inc., Cary, NC USA) and Least Significant Difference (LSD) test was used for mean separation. All data from the plants grown in soils of the six locations were combined.

Results and Discussions

Results indicated that the organic carbon contents (OC) vary among soils of different locations ranging between low and high (Table 2). According to Maria and Yost (2006), organic carbon contents of <1.5%, 1.5-2.5% and >2.5% are rated as low, medium and high, respectively. The OC content of surface horizons in all locations, except Alage, was high and decreased with depth attributed to accumulation of more organic materials on the surface soils. These findings are in line with those reported by Getahun et al. (2014) and Maria and Yost (2006). High CEC values were recorded in soils of Huletegna Choroko, Kontela and Jole Andegna whereas low values were recorded in Tenkaka Umbulo, Taba and Alage soils. The higher CEC values in Huletegna Choroko and Kontela soils could be attributed to higher OC.

Micronutrient contents of the soils were different both in depth and type of soil (Table 2), which is in agreement with Nayyar et al. (2001) who stated that micronutrient contents vary in different areas depending on the soil properties and management conditions. All micronutrients (Fe, Cu, Zn and Mn) were generally decreased with depth which is in line with the finding reported by Mulugeta and Sheleme (2010). The DTPA Zn and Fe contents of most of the soils were below 1.5 and 4.5 mg kg⁻¹, respectively, which were reported to be critical by Aref (2012), Haque et al. (2000), and Westfall and Bauder (2011). According to Havlin et al. (1999), the extractable Zn level in Tenkaka Umbulo, Taba and Alage soils was low whereas it varied from low to high in soils of Huletegna Choroko and Jole Andegna, and low to medium in soils of Kontela (Table 3). Extractable Fe was also low in soils of Tenkaka Umbulo, Taba, Jole Andegna and Alage while it ranged from low to medium in soils of Huletegna Choroko and Kontela. The extractable Cu was low in soils of Tenkaka Umbulo, Huletegna Choroko, Taba and Alage whereas its values ranged from low to high in soils of Kontela, and high in Jole Andegna. Manganese contents varied between low and high in soils of all locations. Generally, availability of all micronutrients decreased with depth but Cu did not follow a clear trend in all study sites. The values of all micronutrients were higher in B horizons of Kontela soils where CaCO₃ and humus were accumulated, which could be attributed to the higher competition by Ca for the adsorption site on the surface of CO_3 and release of the micronutrients upon decomposition of organic matter. In soils of Tenkaka Umbulo, all micronutrients were low in the surface horizon except Mn, indicating the availabilities of Fe, Zn and Cu were inadequate for crop production. The surface horizons of Taba soils contained high micronutrients except Cu suggesting the need for application of Cu fertilizer for crop production.

Availability of Zn varied between medium and high in surface horizons of Jole Andegna and Huletegna Choroko soils indicating low crop response to Zn application on these soils. All micronutrients were low in surface horizons of Kontela and Alage soils, except for Cu at Alage, suggesting the need for application of fertilizers containing Zn, Fe and Mn for crop preproduction. Manganese was high in surface horizons of Jole Andegna and Huletegna Choroko soils indicating it was adequate for crop production on these soils.

Results of analysis of soil samples collected from 0-15 and 15-30 cm depths also indicated that the content of Fe was medium (2.5 -4.5 mg kg⁻¹ soil) in both depths of Haplic Calcisols (Humic), Andic Cambisols (Humic) and in 0-15 cm depth of Haplic Lixisols (Siltic), whereas it was low (<2.5 mg kg⁻¹ soil) in both depths of the rest of the soils. The content of Cu was high in both depths of Haplic Luvisols (Humic), but it was low (<0.4 mg kg⁻¹ soil) in both depths of the rest of the soils. The content of the rest of the soils. Similar findings were reported by Abay et al. (2015). The Zn content of Haplic Luvisols (Humic) in both depths was high (>1 mg kg⁻¹), whereas it was low in both depths of the rest of the soils. Manganese was low (<1 mg kg⁻¹soil) in both depths of Luvic Calcisols (Siltic), Haplic Calcisols (Chromic) and Haplic Luvisols (Humic) but it was high in both depths of the other soils. Table 3).The low micronutrients in most of the soils suggested the need for application of fertilizers containing Zn, Fe, Cu and Mn for crop production.

Type of soil	soil depth	pН	OC%	CEC Cmol (+) kg	Fe	Cu	Zn	Mn
	(cm)			¹ of soils		mg kg ⁻¹	soil	
Luvic Calcisols	0-15	8.0	4.8	43.9	0.23	0.40	0.36	0.50
(Siltic)	15-30	8.2	2.4	44.9	0.38	0.33	0.33	0.23
Haplic	0-15	7.4	2.1	26.7	0.53	0.22	0.22	0.47
Calcisols	15-30	7.6	2.3	25.7	0.70	0.15	0.15	0.45
(Chromic)								
Haplic	0-15	7.5	2.3	20.1	2.94	0.27	0.27	1.23
Calcisols	15-30	7.3	2.2	21.3	2.37	0.18	0.19	1.18
(Humic)								
Haplic Luvisols	0-15	7.5	2.3	40.6	1.42	1.47	1.47	0.58
(Humic)	15-30	7.3	1.8	38.9	1.06	1.29	1.3	0.40
Andic Lixisols	0-15	7.7	2.9	21.7	1.60	0.27	0.27	1.30
(Humic)	15-30	7.7	1.8	21.7	1.61	0.26	0.26	1.30
Andic	0-15	7.4	2.5	20.6	2.56	0.40	0.38	1.50
Cambisols	15-30	7.8	2.9	21.7	2.48	0.25	0.25	1.30
(Humic)								
Haplic Lixisols	0-15	7.5	1.7	19.6	3.25	0.28	0.38	1.30
(Siltic)	15-30	7.5	3.0	20.7	1.59	0.38	0.28	1.50
Haplic Lixisols	0-15	7.67	2.8	23.2	1.71	0.27	0.27	1.90
(Humic)	15-30	7.26	4	20.3	1.34	0.24	0.24	1.80

Table 2. Micronutrient, OC and CEC contents of the study soils

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Location	Horizon	Depth	pН	OC	CEC	Fe	Cu	Zn	Mn
			(H_2O)	(%)	cmol (+)/kg of soils	mg/kg soil			
T. Umbulo	Ар	0-26	8.08	4.1	21.6	2.08	0.23	0.42	1.54
	В	26-69	8.3	2.54	23.2	0.66	0.30	0.33	1.08
	С	69-116	8.74	1.71	16.3	0.28	0.27	0.13	0.06
	Ck	116-156	9.4	1.40	14.9	0.64	0.23	0.14	0.05
	2B	156-176	9.5	1.29	17.3	0.7	0.28	0.12	0.01
	2Bn	176-209	9.7	1.63	22.4	2.58	0.43	0.36	0.34
	Ар	0-25	7.3	4.33	23.5	2.44	0.27	1.04	4.18
	B1	25-69	7.4	2.40	24.6	1.13	0.09	0.20	2.57
	B2	69-115	8.25	2.58	25.2	0.68	0.12	0.30	0.87
	Bt	115-131	8.74	2.54	29.3	1.18	0.28	0.33	0.18
	Ck	131-157	9.6	1.25	15.5	1.35	0.16	0.20	0.14
	С	157-187	9.6	0.17	16.3	2.50	0.42	0.16	0.14
Taba	Ар	0-33	7.72	3.90	23.6	4.60	0.16	1.07	1.03
	AB	33-73	7.03	3.10	16.5	1.63	0.07	0.68	1.66
	ABg	73-94	7.8	1.01	10.9	0.68	0.25	0.38	0.41
	Bti	94-157	7.6	1.03	18.8	0.17	0.54	0.36	0.30
	Ар	0-47	7.58	3.60	22.7	1.11	0.39	2.06	1.32
	Е	47-94	7.8	2.20	15.9	1.39	0.28	1.67	1.35
	Bt	94-170	7.9	1.80	23.5	1.12	0.07	0.44	1.76
H. Choroko	Α	0-52	8.24	4.26	23.7	0.69	0.16	0.71	1.05
	AB	52-67	8.36	2.56	19.8	0.93	0.19	0.47	0.71
	Btg	67-98	8.5	1.55	13.7	0.84	0.10	0.04	0.24
	В	98-170	8.2	1.17	14.1	0.29	0.52	0.70	0.29
	Α	0-60	7.5	6.30	26.6	3.27	0.39	1.88	5.24
	CB	60-87	8.47	3.52	15.2	1.08	0.20	0.16	1.29
	С	87-187	7.6	2.42	23.3	0.43	0.50	0.24	0.22

Table 3 Available micronutrients of soils in the study sites

Location	Horizon	Depth	pH	OC		Fe	Cu	Zn	Mn
			(H_2O)	(%)	cmol (+)/kg of soils				
						g/kg soil	T		
Kontela	Ak	0-67	7.94	3.51	26.4	0.27	0.41	0.15	0.22
	Е	67-76	9.05	1.00	16.8	0.23	0.66	0.19	0.61
	Bkn	76-100	9.41	3.20	16.4	0.31	0.81	0.18	1.27
	С	100-107	9.95	1.66	10.3	0.37	0.31	0.08	0.23
	2Bh	107-133	9.25	4.00	34.1	4.29	0.66	0.61	0.75
	2C	133-143	9.8	1.53	1.21	1.19	0.27	0.17	0.20
	3C	143-169	9.12	3.70	24.4	1.35	0.66	0.52	1.26
	А	0-50	8.32	4.46	47.7	0.31	0.43	0.20	0.63
	В	50-80	8.5	1.96	42.3	0.33	0.21	0.05	0.08
	Bk	80-109	9.2	1.63	37.3	4.35	0.70	0.70	0.73
	Ck	109-140	9.7	0.46	1.20	1.21	0.26	0.16	0.22
	2B	140-151	9.55	0.19	40.5	5.20	0.71	0.60	0.22
	2C	151-187	9.54	0.16	32.4	0.75	0.31	0.32	0.18
Alage	Ар	0-20	6.9	1.88	18.6	0.57	0.34	0.28	0.1
	Bh	20-48	7.65	2.43	32.7	0.41	0.25	0.18	0.01
	BC	48-87	8.22	1.17	25.0	0.33	0.30	0.09	0.16
	Ck	87-137	8.46	0.71	12.6	0.17	0.10	0.09	0.02
	C ₁	137-175	9.3	0.37	14.6	0.39	0.25	0.02	0.04
	C_2	175-197	9.5	0.28	16.9	0.67	0.07	0.05	3.44
J. Andegna	Ар	0-30	7.61	2.86	45.7	1.79	1.67	0.90	8.89
	В	30-60	7.66	1.66	33.8	1.46	1.07	0.32	1.28
	Е	60-92	7.70	1.61	32.4	0.51	1.06	0.28	1.27
	Bt_1	92-165	7.46	1.87	50.4	0.95	1.68	0.77	3.42
	E'	165-205	8.01	1.54	40	1.21	0.87	0.19	1.41
	Ар	0-23	7.6	2.42	47.8	0.82	0.33	1.33	12.1
	Ē	23-52	7.43	1.61	29.7	0.42	1.40	0.7	4.46
	Bt_1	52-92	7.33	1.70	39.3	0.88	1.43	1.15	4.97
	Bt ₂	92-146	8.02	2.18	48.3	0.21	1.50	0.18	0.63
	Ē	146-175	7.59	1.95	40.2	0.36	1.51	0.23	0.32
	Bt ₃	175-200	8.19	1.78	31.0	0.07	1.21	0.28	0.17

Table 3 Available	micronutrients	of soils in the	e study sites	(cont'd)
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Application of NPZn and NPFe did not significantly (p>0.05) influence haricot bean yield compared to NP fertilizers (Tables 4), which is in agreement with the findings reported by Zhao et al. (2011) on dry weight of wheat. McKenzie et al. (2000) also reported that fertilization of Zn did not significantly increase seed yield of dry bean in Canada. However, application of Zn with NP significantly increased grain yield compared to the no fertilizer treatment, but did not significantly increased over the NP, and NPFe treatments. Though not significant, haricot bean fertilization with NPFe increase grain yield compared to the no fertilizer. Similarly, Malakouti (2008) and Rastogi et al. (2014) reported an increase in grain yield of wheat and linseed due to application of Zn and Fe in Iran and India, respectively. Heidarian et al. (2011) also reported an increase in yield and yield components of soy bean due to fertilization of Zn and Fe. Fertilization of haricot bean only with NP did not significantly increase grain yield over the no fertilizer, and with NPZn treatments. Application of NP, NPZn, and NPFe significantly increased grain yield over NPZnFe, Zn only, and Fe only treatments. Lower grain yield was observed at application of NPZnFe than the yield obtained at application of NP, NPZn, NPFe, and with no fertilizer, which could be attributed to antagonism between Fe and Zn that occurs through competition between Zn²⁺ and Fe²⁺ during uptake, and competitive inhibition between Zn and Fe during unloading in the xylem (Alloway, 2008; Kabata-Pendias, 2001). The Fe only treatment produced significantly lower grain yield than the no fertilizer indicating application of Fe alone affected the balance of NP in the soil (Poshtmasari et al., 2008). The Zn only treatment also yielded low grain though not significantly lower than the no fertilizer treatment. The highest grain yield (16.60 g/pot), which is significantly higher than the yield obtained with no fertilizer (14.73 g/pot) was observed at application of NPZn, while the lowest grain yield (12.80 g/pot) was observed with Fe only treatment. Plant height also followed similar trend with grain yield (Table 4). Fertilization of haricot bean only with NP did not increase plant height compared to the no fertilizer treatment. Combined application of Zn and NP significantly increased plant height over the no fertilizer, NP, Zn only and Fe only treatments. A similar

finding was also reported by Hamsa and Puttaiah (2012), who indicated that application of Zn significantly increased plant height. Plants grown with application of Zn and Fe with NP were also significantly taller than those grown with no fertilizer. Combined applications of Zn with NP, Fe with NP, and Zn and Fe with NP produced statistically equal height. The tallest plants (38.22 cm) were observed with NPZn, while the shortest plants (34.61cm) were obtained with Fe only treatment. Number of pods per plant was also significantly higher with NP, ZnNP, and FeNP than those with no fertilizer. On the other hand, NPZnFe, Zn only and Fe only produced statistically equal pods with the no fertilizer treatment. Though not significant, application of Zn increased number of pods per plant compared to the no fertilizer, which is in agreement with the finding reported by Nadergoli et al. (2011). This finding could be attributed to the positive effect of Zn on the formation of stamens and pollens that could increase fertility of flowers to produce more pods (Nadergoli et al., 2011). The most pods per plant (7.50) were observed with application of NP, while the least pods per plant (6.22) were obtained with Fe only treatment. Fertilization of haricot bean with NP alone or combined with Zn and/or Fe did not significantly increase seeds per pod compared to the no fertilizer treatment. Except the Fe only treatment, all treatments including the no fertilizer one produced statistically equal number of seeds per pod. Combined application of NP with Zn and/or Fe, yielded lower seeds per pod than individual application of the nutrients. The Fe only treatment produced significantly higher number seeds per pod than NP with Zn and /or Fe treatments. The most seeds per pod (5.11) were observed with Fe only treatment, whereas the least seeds per pod (4.44) were obtained with NPZnFe treatments, indicating Fe has positive effect in seed filling.

Application of Zn either alone or combined with NP and Fe produced significantly higher tissue Zn concentration than the other treatments and this is in line with the findings reported by Zhao et al. (2011) and Narwal et al. (2010) on wheat. Similar findings also were reported elsewhere by Talukder et al. (2009) and Tryphone and Nchimbi-Msolla (2010) on haricot bean in Tanzania. The highest value of leaf Zn (52.36 mg kg⁻¹) was observed with Zn only treatment whereas the lowest value (15.75 mg kg⁻¹) was obtained with NPFe treatment. The seed Zn values obtained with applications of NPZn, NPZnFe, and Zn only were 29.94, 29.58, and 29.89 mg kg⁻¹, respectively, and are highest and statistically equal, whereas the lowest seed Zn (26.22 mg kg⁻¹) was obtained with application of Fe only. The treatments with no fertilizer, NP, NPFe, and Fe only produced statistically equal tissue Zn concentrations, which are significantly lower than the values obtained with those with Zn fertilizer. Application of Fe either individually or combined with NP and Zn yielded significantly higher tissue Fe concentrations than the rest of the treatments. All treatments with no Fe produced statistically equal tissue Fe concentrations, which were significantly lower than those with Fe fertilizer. The highest leaf and seed Fe concentrations, 293.83 and 55.79 mg kg⁻¹, respectively, were observed with NPZnFe and Fe only, respectively, whereas the lowest values of leaf and seed Fe, 162.09 and 42.76 mg kg⁻¹, respectively, were obtained with application of Zn only. Leaf Fe concentrations were by far higher in all treatments than grain Fe concentrations as reported by Nchimbi-Msolla and Tryphone (2010) suggesting that plant mineral concentrations vary between plant tissues, whereas leaf and grain Zn concentrations were relatively closer. With application of Zn fertilizer, leaf Zn was higher than seed Zn but the reverse was true in the absence of Zn fertilizer. Combined application of Zn and Fe with NP significantly increased the concentrations of Zn and Fe in leaf and seed of haricot beans. Kobraee et al. (2011) also reported that application of Zn and Fe increased concentrations of these nutrients in leaf and seed of soy bean.

Table 4 Yield, yield components and tissue concentrations of Fe and Zn as influenced by Fe and Zn fertilization								
Treatment	Grain	Mean plant	Mean no	Mean	Leaf Zn	Seed Zn	Leaf Fe	Seed Fe
	yield	height	of pods	no of	$(mg kg^{-1})$	$(mg kg^{-1})$	$(mg kg^{-1})$	$(mg kg^{-1})$
	(g/pot)	(cm)	per plant	seeds				
				per pod				
No fertilizer	14.73bcd	34.81c	6.39c	4.83ab	20.53c	26.71b	186.15b	44.11cd
NP	15.86ab	34.58c	7.50a	4.83ab	16.97cd	27.06b	181.04b	45.99c
NPZn	16.60a	38.22a	7.39a	4.67b	44.75b	29.94a	178.12b	44.94cd
NPFe	15.31ab	36.39abc	7.17ab	4.67b	15.75d	27.06b	274.63a	52.12b
NPZnFe	13.32de	37.14ab	6.61bc	4.44b	51.40a	29.58a	293.83a	54.99a
Zn only	13.75cde	35.11bc	6.94abc	4.78ab	52.36a	29.89a	162.09b	42.76d
Fe only	12.80e	34.61c	6.22c	5.11a	18.58cd	26.22b	291.56a	55.79a
CV (%)	19.25	8.98	15.99	12.69	17.78	5.18	18.84	7.54
LSD (5%)	1.8672	2.1349	0.731	0.400	4.7546	0.9639	27.968	2.4341
P-value								
[@] Trt	0.0006	0.0034	0.0029	0.0663	<.0001	<.0001	<.0001	<.0001
Soil	<.0001	0.0001	<.0001	0.0123	<.0001	<.0001	<.0001	<.0001
Trt*Soil	0.1241	0.0136	0.1199	0.5226	<.0001	0.0016	0.0028	<.0001

Table 4 Yield, yield components and tissue concentrations of Fe and Zn as influenced by Fe and Zn fertilization

[@]Trt=Treatment

Haricot bean yield, yield components and tissue Zn and Fe concentrations varied significantly across soils of the different locations (Table 5). Similar findings were reported by Nchimbi-Msolla and Tryphone (2010) in Tanzania on haricot bean. The grain yields observed in soils of Alage and Tenkaka Umbulo are significantly higher than the values obtained in soils of the other locations. The highest grain yield (18.73 g/pot) was observed in Alage soils and this value was significantly higher than the values obtained in soils of other locations, except in soils of Tenkaka Umbulo. The lowest grain yield (11.94 g/pot) was observed in Taba soils and this value is not significantly different from the values obtained in soils of Kontela, Huletegna Choroko, and Jole Andegna. The tallest plants (38.83 cm) were observed in Tenkaka soils, while the shortest (33.95 cm) were obtained in Huletegna Choroko soils. The tallest plants are significantly different from the plants grown in the rest of the locations. Number of pods per plant followed a similar trend with grain yield and plant height. The highest number of pods per plant (8.05) was observed in soils of Tenkaka Umbulo and this value is statistically equal with the value obtained in Alage soils, but higher than the values observed in soils of the rest of the locations. The lowest number of pods per plant (5.62) was obtained in Kontela soils and this value is significantly lower than the value obtained in soils of all of the rest of the locations. The number of seeds per pod observed in soils of Alage and Taba are significantly higher than the values obtained in soils of the other locations. The highest value (5.14) was produced in soils of Taba, whereas the lowest one was yielded in soils of Huletegna Choroko.

Tissue Zn and Fe concentrations also varied across locations. Significantly higher leaf Zn was produced in soils of Alage than the other locations. The highest leaf Zn (45.46 mg kg⁻¹) was observed in Alage soils, while the lowest value (23.76 mg kg⁻¹) was obtained in Kontela soils. The seed Zn concentrations observed in soils of Huletegna Choroko and Tenkaka Umbulo, 30.16 and 30.76 mg kg⁻¹, respectively, are significantly higher than the values obtained in soils of the other locations. The highest seed Zn (30.76 mg kg⁻¹) was observed in soils of Tenkaka Umbulo, while the lowest value (24.93 mg kg⁻¹) was produced in Alage soils. The lowest seed Zn is significantly lower than the values obtained in soils of all the other locations. The highest leaf Fe concentrations obtained in soils of Alage and Jole Andegna are significantly higher than the values observed in soils of Jole Andegna, whereas the lowest value (191.63 mg kg⁻¹) was obtained in soils of Tenkaka Umbulo. The highest seed Fe (59.50 mg kg⁻¹), which is significantly higher than the values obtained in soils of the other locations, was observed in soils of Taba, while the lowest value (41.83 mg kg⁻¹) was observed in soils of Tenkaka Umbulo.

According to Govindaraj et al. (2011), sufficiency range of Fe and Zn concentrations in critical stages of pulses, are 50-250 and 20-40 mg kg⁻¹, respectively. Hodges (2010) also reported that deficiency of Fe in most plant species occurred when the Fe content of leaves was below 10- 80 mg kg⁻¹. Accordingly, Fe, Zn concentrations in the leaves of haricot bean with all treatments in all locations were sufficient.

Soils	Grain	Mean	Mean no	Mean no	Leaf Zn	Seed Zn	Leaf Fe	Seed Fe
	yield	plant	of pods	of seeds	$(mg kg^{-1})$	$(mg kg^{-1})$	$(mg kg^{-1})$	$(mg kg^{-1})$
	(g/pot)	height	per plant	per pod				
		(cm)						
Alage soil	18.73a	34.86bc	7.48ab	4.90ab	45.46a	24.93e	258.13a	44.99c
[*] J. A. soil	13.66b	35.05bc	6.43c	4.62bc	37.10b	27.61c	279.22a	51.03b
[@] H.C. soil	13.54b	33.95c	6.43c	4.48c	29.98c	30.16a	217.65b	49.45b
[#] T. U. soil	17.08a	38.83a	8.05a	4.71bc	25.67cd	30.76a	191.63c	41.83d
Taba soil	11.94b	35.93bc	7.33b	5.14a	26.89cd	28.75b	197.28bc	59.50a
Kontela soil	12.79b	36.40b	5.62d	4.71bc	23.76d	26.18d	199.60bc	45.22c
CV (%)	19.25	8.98	15.99	12.69	17.78	5.18	18.84	7.54
LSD (5%)	1.7287	1.9766	0.6764	0.371	4.4019	0.8924	25.893	2.2535

Table 5 Yield, yield components and tissue concentrations of Fe and Zn as inf	fluenced by soil types
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^(a)H. C. =Huletegna Choroko; [#]T. U. =Tenkaka Umbulo; ^{*}J.A. =Jole Andegna

Conclusion

Micronutrients, especially, Fe, Zn and Cu were low in most of the studied soils indicating the need for application of site specific fertilizers containing these nutrients. Application of NPZn and NPFe did not significantly (p<0.05) influence haricot bean yield compared to NP fertilizers. However, application of Zn with NP significantly increased grain yield compared to the no fertilizer treatment. Though not significant, haricot bean fertilization with NPFe also increased grain yield compared to the no fertilizer. Application of Zn either alone or combined with NP and Fe produced significantly higher tissue Zn concentration than the other treatments. Fertilization of Fe either alone or combined with NP and Zn also produced significantly higher tissue Fe concentration than the other treatments. Haricot bean yield, yield components and tissue mineral concentrations varied significantly across soils. The increased grain yield due to application of NPZn (compared to both NP and no fertilizer) and NPFe (compared to no fertilizer), and the increased tissue Fe and Zn due to

application of the nutrients indicate that fertilization of haricot beans with Fe and Zn is important in the soils of the study sites especially for quality production. However, further studies that include different haricot bean genotypes and levels of Fe and Zn are required for sound conclusions.

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