Response of Haricot Bean Varieties to Different Levels of Iron Application in Selected Areas of Ethiopia

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Abstract
Haricot bean (*Phaseolus vulgaris* L.) can be an important source of Fe for human nutrition, particularly in regions in which human Fe deficiencies are known to occur. A study using replicated field and greenhouse experiments was conducted in Ethiopia to evaluate the yield and Fe uptake response of different haricot bean varieties (Nasir, Ibado, Hawassa Dume, and Sari-1) to different levels of foliar-applied iron (Fe) fertilizer (0, 1, 2, and 3% solution). Pot experiment results indicated yield, yield components, and tissue Fe concentrations varied among varieties and across soils. The variety Ibado yielded the highest leaf Fe concentration (290.19 mg kg^{-1}) whereas Hawassa Dume had the highest number of pods per plant (7.28) and grain yield (15.85 g per pot). Varieties Sari-1 and Nasir produced the highest number of seeds per pod (4.94) and seed Fe concentration (59.02 mg kg^{-1}), respectively. Levels of Fe fertilization did not significantly influence yield and yield components, but significantly increased both leaf and seed Fe concentrations. Application of 3% $FeSO_4.7H_2O$ produced the highest concentration of both leaf (339.50 mg kg^{-1}) and seed Fe (53.46 mg kg^{-1}). Field experiments revealed that haricot bean varieties significantly varied in yield, yield components, and leaf and seed Fe concentration. Highest grain yield (3099.55 kg ha^{-1}) was observed with variety Hawassa Dume. Production was significantly influenced by planting season and location. Overall, 3% $FeSO_4.7H_2O$ fertilizer application best improved the quality of haricot bean produced.
Keywords: Haricot bean, Iron, Concentration, Varieties, Seed Fe, Leaf Fe

Introduction

Iron is an essential trace element for all organisms (Aref, 2012). It plays a central role in chlorophyll production, photosynthesis, and energy transfer within plants (Singh, 2004). It is essential for human health. Rubio et al. (2009) suggested a dietary Fe intake of 13.161 mg day^{-1} is relatively common in countries where diets include animal protein. Food legumes can also contain appreciable quantities of Fe and it is estimated that forty percent of Fe intake in developing countries is derived from legumes and cereals (Tryphone and Nchimbi-Msolla, 2010). Moreover, Imtiaz et al. (2010) warned that high consumption of cereal-based foods of low Fe content poses a health risk and can slow physiological processes. They suggest an adequate daily dietary intake for young and adults might range from 10 to 60 mg.

Nchimbi-Msolla and Tryphone (2010) reported that haricot bean (*Phaseolus vulgaris* L.) is an important source of minerals and protein and can supply all of the iron that humans require. Haricot bean is one of the principal food and cash crops grown and consumed throughout the world as a grain and vegetable legume (Tryphone and Nchimbi-Msolla, 2010). It plays a vital role in the nutrition of low-income people, especially in developing countries, where it is often the main dietary source of protein, carbohydrate, dietary fiber, and minerals (Prolla et al., 2010). It is one of the most important cash crops and sources of protein for farmers in many lowlands and mid-altitude zones of Ethiopia. The country's export earnings from haricot bean is estimated to be over 85% of export earnings from pulses, with haricot bean ranking third among these commodities and contributing about 9.5% of total export value from Ethiopian agriculture (Katungi et al., 2010). Optimizing growing conditions, production, and quality is therefore crucial to improve the health and income of people, who depend on bean for Fe nutrition and as a cash source.

Haricot bean is sensitive to Fe deficiency in soils (Taber, 2008), though requirements vary among varieties. Additionally, Tryphone and Nchimbi-Msolla (2010) reported that variation in response to Fe application was observed among common bean genotypes in Tanzania. Iron is the fourth most abundant element in the earth's crust with a total content of 100 mg kg^{-1} or >10% in the soils and 50–250 mg kg^{-1} in the plant (Taber, 2008), yet it remains the third most-limiting nutrient for plant growth. This is primarily due to the low solubility of the oxidized ferric form in aerobic environments (Aref, 2012). Munson (1998) reported Fe content of plant >500 mg kg^{-1} as toxic.

Soil test Fe content is considered to be very low when DTPA-Fe is less than 2.5 mg kg^{-1} and marginal when DTPA-Fe is between 2.5 and 4.5 mg kg^{-1}. Values above 4.5 mg kg^{-1} indicate low probability of iron deficiency (Wortmann et al., 2012). Deficiencies of iron in soils is caused by high soil pH (an increase of pH in one unit, e.g., from 6.5 to 7.5, decreases available Fe 1,000 fold), high free soil calcium carbonate ($CaCO_3$), poor aeration (excess CO_2), low organic matter content (Taber, 2008), and high P level (Ronan, 2007). The problem of Fe deficiency in soils and in humans can be addressed by application of Fe containing fertilizers in low Fe soils, and identifying haricot bean varieties that accumulate relatively high levels of Fe, respectively. This could contribute significantly to improving Fe content for consumers who depend on the common bean as major component of their diet.

A soil characterization study carried out in Halaba, Taba and Butajira showed that availability of Fe for crop production was low (Abay et al., 2015). Improving the nutritional quality such as Fe content of haricot bean is an important move in sustaining the health of communities relying on this crop. Cakmak (2002) also indicated that increasing the concentrations of Fe in grains is a high priority research task, and would greatly contribute to the alleviation of Fe deficiencies in human populations worldwide. Iron fertilization is one method to prevent Fe deficiency and increase its concentration in grains (Blair et al., 2009). Moore et al. (2012) also reported that Fe deficiency in beans could be corrected by application of iron sulfate fertilizer. Another report by Govindaraj et al. (2011) indicated that application of fertilizer Fe increased in seeds and stover of wheat and maize. Therefore, this study was conducted to evaluate the response of haricot bean varieties to different foliar application rates of Fe, when grown in soils from different bean growing regions of Ethiopia under greenhouse and field conditions.

Materials and Methods

Experiments were conducted on farmers' fields and in greenhouse conditions. Factor A was Fe fertilizer with four levels (0, 1, 2, and 3% solution) and factor B was haricot bean of four varieties (Nasir, Ibado, Hawassa Dume, and Sari-1). Ferrous sulphate solutions of 1, 2, and 3% were prepared by adding 1, 2, and 3 kg of ferrous sulfate into 100 L of water, respectively (Incitec Pivot, 2003). The experimental sites were located near Halaba (Latitude: 07^{o}20'34.5" and Longitude: 38^{o}06'30.0"), Taba (Latitude: 07^{o}01'01.9" and Longitude: 37^{0}53'57.6"), and Butajira (Latitude: 08^{o}12'25.9" and Longitude: 38^{o}27'33.2"), and soils (0 to 30 cm depth) were also collected from these sites for the greenhouse experiment. The types of soils were: Haplic Luvisols (Humic) in Butajira; Andic Lixisols (Humic) in Halaba; and Haplic Lixisols (Siltic) in Taba. The soil properties of each site are indicated in Table 1.

Table 1. Physico-chemical properties of the experimental soils

Soil properties	Butajira soil	Halaba soil	Taba soil
Textural class	Clay loam	Clay loam	Clay loam
pH (H_2O)	7.4	7.70	7.47
Org. C (%)	2.05	2.35	2.35
DTPA Zn (mg kg^{-1})	0.49	1.33	1.80
DTPA Fe (mg kg^{-1})	1.26	1.60	1.50
Total N (%)	0.34	0.24	0.16
Available P (mg kg^{-1})	12.13	10.00	14.30

The source of Fe was heptahydrated iron sulphate ($FeSO_4.7H_2O$). This was sprayed three times, at 15 day intervals, starting at 15 days after planting at a volume of 100 L ha^{-1} per each application date. Nitrogen and P were equally applied for all treatments. Phosphorus was applied just before planting as TSP at 50 mg P kg^{-1} soil (0.25g P per pot) for the pot experiment and 20 kg P ha^{-1} for the field experiment. Nitrogen was also applied just before planting as urea at 0.045g N per pot band at 18 kg Nha^{-1} for the field experiment. Four haricot bean seeds per pot were sown, thinned to two seedlings at 10 days after sowing. For the field experiment, haricot bean was planted in rows with a spacing of 10 cm between plants and 40 cm between rows during the belg (short rain, which normally occurs during the months of March, April and May) and meher (long rain, which normally occurs during the months of June, July and August) seasons, with a plot size of 4 × 4 m. The greenhouse experiment was conducted in pots with 5 kg soil per pot. All appropriate management practices were carried out equally for all treatments. Plant height, number of pods per plant, number of seeds per pot, 1000 seed weight, and biomass and grain yields were collected. Three fully developed leaves at the top of the plant during initial flowering, along with seeds, were collected from each treatment pot and plot, dried in an oven at 70 ^{o}C for 24 h, then ground using a rotating sample mill. The ground plant materials were digested and analyzed for Fe content according to the following procedures. A sample of 0.5 g was weighed using five digit sensitive balance into the digestion tube. Six milliliters of nitric acid (HNO_3) was added to each tube. The tubes were placed in a digestion block at 90 ^{o}C for 45 min. Five milliliters of hydrogen peroxide (H_2O_2) was added, in two splits (3 ml and 2 ml) while the samples were in the digestion block, and digested for another 65 min. Three milliliters of 6 *M* hydrogen chloride (HCl) was then added and the samples were digested until the solution had turned completely clear (after about 5 minutes). The tubes were removed from the block, cooled for 20 minutes, and shaken using a vortex.The digests were then transferred into dram vials, brought to 25 ml with deionized water, and stored. The concentration of Fe was analyzed using a Microwave Plasma Atomic Emission Spectrometer (MPAES) at 259.940 nm. 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 two seasons and three locations were combined.

Results and Discussion

Pot experiments

The pot experiment indicated that yield, yield components and tissue Fe concentrations of haricot bean varied significantly across soils (Table 2). The tallest plants (40.32 cm) and highest seed Fe concentration (59.04 mg kg^{-1}) were recorded in Taba soil, whereas the lowest values, 38.01 cm and 35.63 mg kg^{-1}, respectively, were observed in Halaba soil. However, highest values of number of pods per plant (6.8), grain yield (16.44a $kgha^{-1}$) and leaf Fe concentration (302.97 mg kg^{-1}) were obtained in Halaba soil, and the lowest values of number of pods per plant (5.5) and seeds per pod (4.0) were recorded in Taba soil. The highest number of seeds per pod (4.4) and the lowest value of leaf Fe concentration (224.37 mg kg^{-1}) were observed in Butajira soil. Yield, yield components and tissue Fe concentrations also varied significantly among haricot bean varieties (Table 3) indicating haricot bean varieties vary in yield potential and Fe accumulating capability. This is in agreement with Gebre-Egziabher et al. (2014), who reported that haricot bean varieties varied significantly in number of pods per plant and seeds per pod, and grain yield in northern Ethiopia. Bhargava et al. (2008) reported that the accumulation of Fe varies greatly among crop varieties. The tallest plants (43.97 cm), highest leaf Fe concentration (290.19 mg kg^{-1}), and fewest pods per plant (4.8) and seeds per pod (3.3) were observed with Ibado. Hawassa Dume produced the highest pods per plant (7.3) and grain yield (15.85 $kgha^{-1}$), whereas Sari-1 and Nasir produced the highest seeds per pod (4.9) and seed Fe concentration (59.02 mg kg^{-1}), respectively. Though Hawassa Dume yielded the highest grain (15.85 g per pot), it is almost equal to the yield of Nasir (15.75 g per pot). The lowest leaf and seed Fe, 235.68 and 44.56 mg kg^{-1}, respectively, were recorded with Hawassa Dume. Moraghan et al. (2002) similarly reported in pot experiment that yield and tissue Fe concentrations varied among haricot bean varieties in the Netherlands. Tryphone and Nchimbi-Msolla (2010) likewise reported that Fe concentrations in leaf and seed varied among haricot bean genotypes. They rated genotypes with leaf Fe concentrations ranging from 163.7 to 270 mg kg^{-1} as low, 271 to 310 mg kg^{-1} as moderate, and 311 to 485.6 mg kg^{-1} as high; and rated seed iron content ranging from 23.6 to 42.0 mg kg^{-1} as low, 43.0 to 59.6 mg kg^{-1} as moderate, and 60.4 to 105.5 mg kg^{-1} as high. The findings of this experiment using soils collected in Ethiopia revealed leaf and seed Fe concentrations consistent with the low to moderate ratings. Thus, Hawassa Dume had low and moderate leaf and seed Fe

Table 2. Haricot bean production as influenced by soil type.

Location of the soils	Plant height (cm)	No of pods per plant	No of seeds per pod	Grain yield (g/pot)	Leaf Fe (mg kg^{-1})	Seed Fe (mg kg^{-1})
Butajira	38.22b	6.1b	4.4a	16.29a	224.37c	54.51b
Taba	40.32a	5.5c	4.0b	11.15b	262.40b	59.04a
Halaba	38.01b	6.8a	4.3ab	16.44a	302.97a	35.63c
CV (%)	9.89	18.30	19.87	22.81	14.69	14.09
LSD (5%)	1.56	0.55	0.34	1.34	15.67	2.84
			P value			
Variety (Var)	<.0001	<.0001	<.0001	0.0005	<.0001	<.0001
Fe	0.2365	0.2968	0.9718	0.6308	<.0001	0.0004
Var × Fe	0.3374	0.1019	0.5587	0.1307	0.0109	<.0001
Soil	0.0066	<.0001	0.0369	<.0001	<.0001	<.0001
Variety × Soil	0.3083	0.1007	0.1255	0.0725	0.0020	<.0001
Fe × Soil	0.0238	0.4883	0.5084	0.1080	0.0023	<.0001
Var × Fe × Soil	0.0090	0.0473	0.5412	0.2411	0.0661	<.0001

Means followed by the same letter(s) within a column are not significantly different at $P \leq 0.05$ according to least significant difference (LSD) test.

concentrations, respectively, whereas Ibado and Nasir had moderate leaf and seed Fe concentrations, respectively. Therefore, varieties Nasir and Hawassa Dume as seed, and Ibado as leaf are recommended to be consumed for better Fe supply. Oyelude et al. (2012) reported that consumption of *Phaseolus vulgaris* leaf could assist to meet part of the daily requirement of iron in the diet. The authors also reported that the phytates level in *Phaseolus vulgaris* leaf does not hinder availability of iron.

Table 3. Yield, yield components and tissue Fe concentration as influenced by variety

Yield components and Tissue Fe	Variety				CV (%)	LSD (5%)
	Nasir	Ibado	Hawassa Dume	Sari-1		
Plant height (cm)	36.99b	43.97a	36.94b	37.51b	9.89	1.80
No of pods per plant	6.64b	4.81d	7.28a	5.89c	18.30	0.53
No of seeds per pod	4.39b	3.31c	4.36b	4.94a	19.87	0.40
Grain yield (g/pot)	15.75a	12.65c	15.85a	14.24b	22.81	1.55
Leaf Fe ((mg kg^{-1}))	270.21b	290.19a	235.68c	256.90b	14.69	18.09
Seed Fe ((mg kg^{-1}))	59.02a	44.78c	44.56c	50.55b	14.09	3.28

Means followed by the same letter(s) within a row are not significantly different at P ≤ 0.05 according to least significant difference (LSD) test.

The application of different levels of Fe fertilizer did not significantly influence yield and yield components of haricot bean varieties, but it significantly increased both leaf and seed Fe concentrations (Table 4) indicating that the varieties are relatively nonresponsive, irrespective of Fe deficiencies, as suggested above (Hawassa Dume had low leaf Fe concentrations). Anu et al. (2014) reported that foliar application of Fe increased Fe content in linseed shoot. The highest concentrations of both leaf (339.50 mg kg^{-1}) and seed Fe (59.46 mg kg^{-1}) were observed at application of the highest level of Fe fertilizer (3% $FeSO_4.7H_2O$). A similar finding was reported by Zhao et al. (2011) for wheat. In the Ethiopian haricot bean experiment, the highest seed Fe concentration was 59.46 mg kg^{-1}, which is enough for 4.5 days per person (Rubio et al. 2009), i.e. 1 kg of haricot bean produced at application of 3% $FeSO_4.7H_2O$ can supply a single person's Fe needs for 4.5 days.

Table 4. Yield, yield components and tissue Fe concentration as influenced by increasing levels of Fe fertilizer

Yield components and Tissue Fe	$FeSO_4.7H_2O$ (%)				CV (%)	LSD (5%)
	0	1	2	3		
Plant height (cm)	37.89a	39.49a	38.58a	39.46a	9.89	1.80
No of pods per plant	6.17a	6.00a	6.00a	6.44a	18.30	0.53
No of seeds per pod	4.28a	4.19a	4.25a	4.28a	19.87	0.40
Grain yield (g/pot)	14.94a	13.99a	14.34a	14.71a	22.81	1.55
Leaf Fe (mg kg^{-1})	183.04d	240.17c	290.27b	339.50a	14.69	18.09
Seed Fe (mg kg^{-1})	46.23d	50.81c	54.41b	59.46a	14.09	3.28

Means followed by the same letter(s) within a row are not significantly different at P ≤ 0.05 according to least significant difference (LSD) test.

The interaction between variety and levels of Fe fertilizer significantly ($p<0.0001$) influenced concentrations of Fe in tissues of haricot bean (Fig.1) indicating varieties vary in Fe fertilizer requirements to accumulate the same amount of Fe in their tissues. The highest tissue Fe concentrations of Nasir and Sari-1 were obtained at application of 3% $FeSO_4.7H_2O$, while the lowest values were observed with no Fe fertilizer. On the other hand, Ibado and Hawassa Dume produced the highest leaf and seed Fe concentrations at applications of 3 and 2% $FeSO_4.7H_2O$, respectively, while the lowest values were observed with no Fe fertilizer. Therefore, application of 3% $FeSO_4.7H_2O$ is recommended to produce quality product from Nasir and Sari-1.On the other hand, application of FeSO4.7H_2O at 3% (if leaf is considered) and 2% (if seed is considered) are recommended for quality production from Ibado and Hawassa Dume, respectively. The concentrations of Fe in leaves were by far higher than that of seeds in all varieties, locations, and at different levels of Fe fertilizers suggesting that concentrations of plant minerals vary between plant tissues (Nchimbi-Msolla and Tryphone, 2010).

The interaction between variety and soil significantly affected tissue Fe concentrations (Fig. 3). Nchimbi-Msolla and Tryphone (2010) likewise reported that a significant difference in Fe concentration was observed among cultivars of haricot bean between locations in Tanzania. In the Ethiopian experiment, the highest leaf Fe concentrations in all varieties were observed at Halaba, with the lowest values recorded in Taba, although in Ibado the lowest value was observed at Butajira (Fig. 3). The highest leaf Fe concentrations in all varieties imply that leaves can be consumed as a vegetable at Halaba to supply or supplement Fe (Tryphone and Nchimbi-Msolla, 2010). In contrast, the highest seed Fe concentration in all varieties were observed at Taba, except Ibado, for which highest seed Fe was observed at Butajira. This finding indicates that though differences in tissue Fe concentrations were observed among varieties, Nasir, Hawassa Dume, and Sari-1 showed a similar trend for storing Fe in their tissues across locations, while Ibado differed.

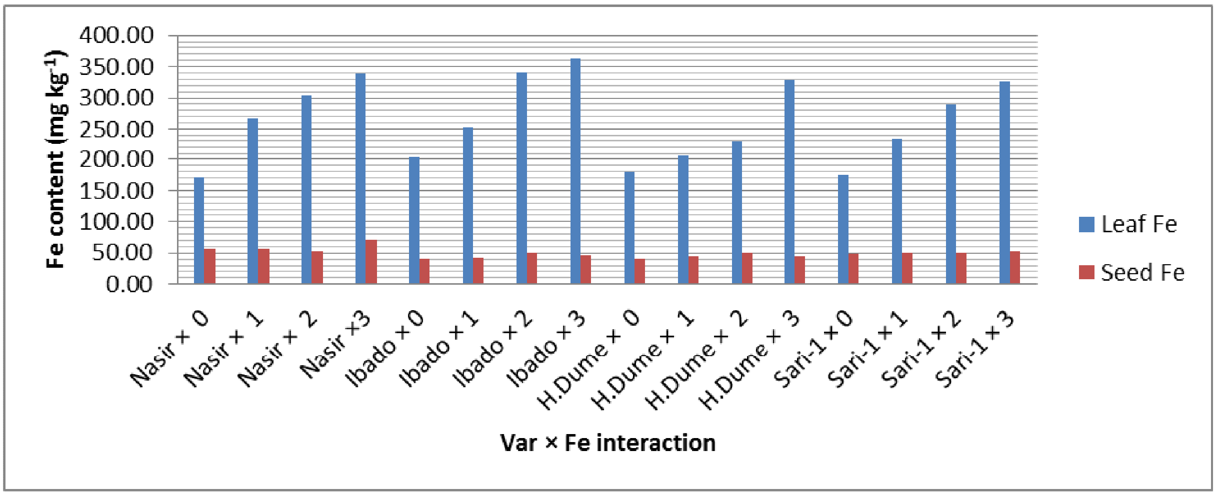

Fig. 1. Leaf and seed Fe concentrations as influenced by interaction between haricot bean varieties and different levels of Fe fertilizer

The interaction between Fe levels and soil significantly ($p<0.0001$) influenced tissue Fe concentrations (Fig. 2). At all levels of Fe fertilizer including zero (control), the highest concentrations of leaf Fe were obtained in Halaba soil, while the lowest values were recorded in Butajira soil. Highest seed Fe concentrations with zero and at 3% Fe fertilizer were observed in Taba soil, but at 1 and 2% Fe fertilizer, the values were highest in Butajira. In contrast, to leaf Fe, the lowest seed Fe concentrations were observed in Halaba soil. This indicates that leaf and seed Fe did not correlate within locations. The finding also showed that the same amount of Fe fertilizer is recommended in all locations to achieve highest leaf Fe concentrations, but the amount required to get the highest seed Fe concentration would vary according to location. At all levels of Fe fertilizer, the highest leaf and the lowest seed Fe concentrations for all varieties were observed in Halaba soil, while the lowest values of leaf Fe were observed in Butajira soil, but for Ibado the lowest leaf Fe concentration was observed in Taba soil (Fig. 4). Nasir at 0 and 1%, Ibado at 2 and 3%, Hawassa Dume at 1 and 2%, and Sari-1 at 2% $FeSO_4.7H_2O$ produced the highest seed Fe concentrations in Butajira, and at the rest of Fe levels produced the highest seed Fe in Taba soil. At zero level of Fe fertilizer, all varieties gave the highest seed Fe concentration in Taba soil, except Nasir, which was highest in Butajira soil. Therefore, this result revealed that at all levels of Fe fertilizer application, quality production of haricot bean may occur at Butajira and Taba in terms of seed Fe concentration, and at Halaba in terms of leaf Fe concentration.

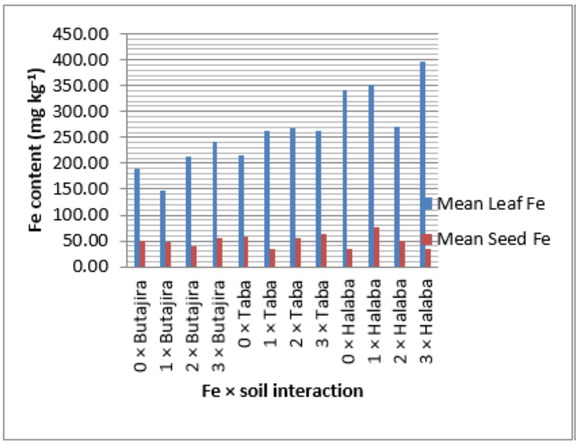

Fig. 2. Leaf and seed Fe concentrations as influenced by Fe fertilizer levels × soil interaction

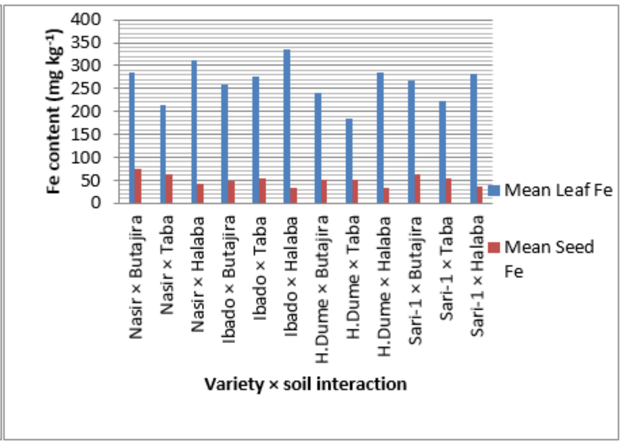

Fig. 3. Leaf and seed Fe concentrations as influenced by variety × soil interaction

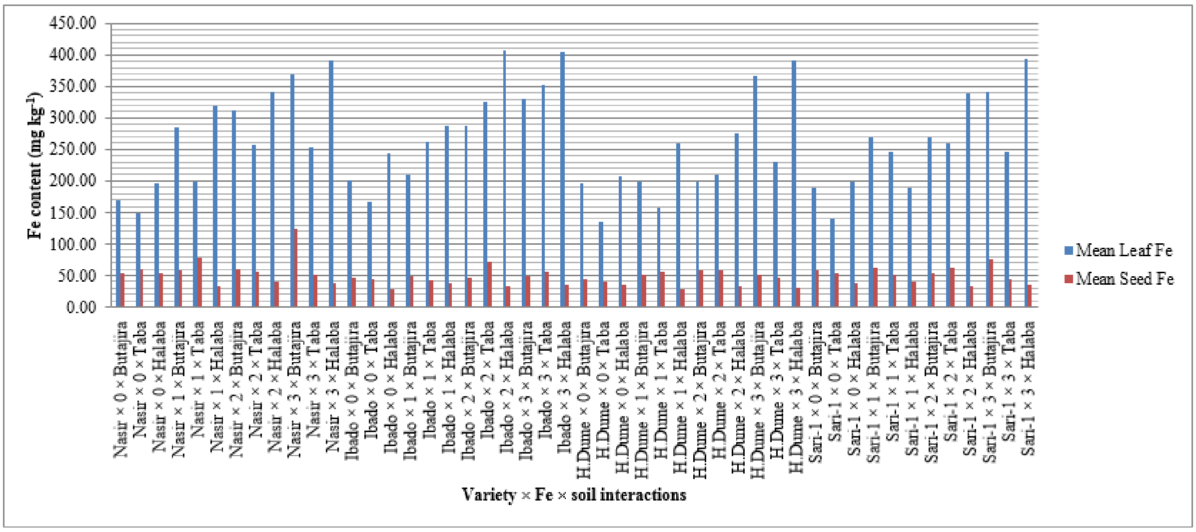

Fig. 4. Leaf and seed Fe as influenced by varieties × Fe fertilizer levels × locations interactions

Field experiments

Like the pot experiments, the field experiments showed significant variation between haricot bean varieties in yield parameters and tissue Fe concentrations (Table 6). Sebuwufu (2013) reported similar finding on yield and yield components of haricot bean. Ghanbari et al. (2013) also reported that haricot bean genotypes varied in tissue Fe concentration. The tallest plant (46.88 cm), highest biomass (7558.6 kg ha^{-1}) and thousand seeds weight (485.49 g) were observed with Ibado, whereas the lowest values, 40.96 cm, 6371.5 kg ha^{-1} and 190.71g, respectively, were observed with Sari-1. The highest grain yield (3099.55 kg ha-1) was observed with Hawassa Dume, which is almost equal to the yield of Nasir (3068.67 kg ha^{-1}), but significantly higher than that of Ibado and Sari-1. Ibado yielded the lowest values in terms of number of branches per plant (4.30), pods per plant (11.66) and seeds per pod (3.95), while the highest values, 4.82, 19.84 and 5.64, respectively, were observed with Nasir. The highest leaf Fe (167.12 mg kg^{-1}) and seed Fe (86.46 mg kg^{-1}) were observed with Ibado and Nasir, respectively.

Application of different levels of Fe significantly increased tissue Fe concentrations and 1000 seed weight, but did not influence grain yield and other yield components (Table 5). The highest seed weight (337.78 g) was observed at 3% $FeSO_4.7H_2O$, while the lowest value (240.76 g) was observed with zero Fe. The highest leaf and seed Fe concentrations, 195.60 and 89.18 mg kg^{-1}, respectively, were observed at 3% $FeSO_4.7H_2O$, while the lowest values, 149.89 and 28.72 mg kg^{-1}, respectively, were obtained at zero Fe fertilizer. Similarly, Yadav et al. (2013) reported a significantly increased fruit yield by application of Fe fertilizer to peach trees in India, and Kobraee et al. (2011) reported increased yield and tissue Fe concentration by application of Fe fertilizer to soybean in Iran. In contrast, Imakumbili et al. (2010) reported application of Fe fertilizer did not significantly affect seed Fe concentration of bean in Tanzania. Yet, in this Ethiopian field study, all interactions with different levels of Fe fertilizer did not significantly influence yield parameters and concentrations of tissue Fe. Application of different levels of Fe fertilizer in different varieties, locations, and seasons did not significantly influence haricot bean production, both in quality and quantity.

All yield, yield components and tissue Fe concentrations varied significantly between locations (Table 7). The highest values of plant height (46.69 cm), number of pods per plant (20.21), grain yield (3014.49 kg ha^{-1}), and seed Fe concentration (119.79 mg kg^{-1}) were observed at Butajira, whereas the lowest values, 40.77 cm, 13.42, 2620.01 kg ha^{-1} and 55.94 mg kg^{-1}, respectively, were observed at Halaba. Contrary to the field experiment, the highest seed Fe concentration was observed in Taba soil in the pot experiment. This could be because Fe absorption in Taba fields might be more affected by environmental factors than in Butajira. However, under controlled conditions, where environmental factors have less effect, Fe absorption was higher in Taba than in Butajira that could be attributed to the higher soil Fe content of Taba soils. The highest biomass yield (7382.8 kg ha^{-1}) was observed at Halaba but not significantly different from the value (7119.1 kg ha^{-1}) observed at Butajira. The highest leaf Fe concentration (217 mg kg^{-1}) was also observed at Halaba, which was significantly higher than that at Butajira and Taba, while the lowest (91.24 mg kg^{-1}) value was observed at Taba. Generally, addition of Fe resulted in best haricot bean production at Butajira, in terms of both quantity and quality, which might be attributable to lower available Fe and higher total nitrogen in the experimental soil as compared to the other locations.

Growing season also significantly influenced haricot bean production (Table 7). Sebuwufu (2013) likewise reported that seed Fe varied greatly across years. Thus, the observed differences in Fe across growing season may be due to dilution effects associated with seed yield. Plant height, number of pods per plant, number of seeds per pod, grain yield, biomass yield and leaf Fe concentration were significantly higher in belg (short rain) season. Haricot bean production was better in belg than meher (long rain) season in terms of quantity, but there was no difference between the two seasons in quality with respect to seed Fe concentration. If concentration of Fe in leaf is considered, however, belg season is recommended (especially where haricot bean leaves can be consumed). The number of branches per plant, 1000 seed weight, and concentration of Fe in seed showed no statistical difference between the two seasons, though a higher value of seed Fe was recorded during belg season (Table 7). The poor bean production in meher might be attributable to the higher rainfall (Ogola, 1991).

Table 5. Effect of Fe fertilization on production and tissue Fe concentration of haricot bean

$FeSO_4.7H_2O$ (%)	Plant height (cm)	no of branches per plant	No of pods per plant	No of seeds per pod	Grain yield (kg ha^{-1})	Biomass (kg ha^{-1})	1000 seeds weight (g)	Leaf Fe (mg kg^{-1})	Seed Fe (mg kg^{-1})
0	45.42a	4.68a	17.26a	5.18a	2792.93a	7072.5a	240.76b	149.89d	28.72d
1	45.20a	4.59a	16.17a	5.25a	2754.04a	6914.1a	241.87b	164.71c	49.53c
2	45.62a	4.58a	16.94a	5.09a	2744.2a	6933.6a	249.86b	175.58b	67.67b
3	45.22a	4.65a	16.69a	5.2a	2846.12a	6927.1a	337.78a	195.60a	89.18a
CV (%)	10.74	16.14	22.86	12.88	20.29	18.62	14.20	21.35	16.41
LSD (5%)	1.35	0.21	1.09	0.19	160.48	368.25	86.81	9.29	20.32
P value									
*V	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.3164	0.8880
Fe	0.0037	0.799	0.2513	0.3742	0.5874	0.8146	0.0806	0.0112	0.0453
V × Fe	0.2943	0.086	0.0562	0.1498	0.6749	0.1970	0.0176	0.9867	0.3958
@L	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.0535	<.0001	<.0001
V × L	<.0001	<.0001	<.0001	0.3644	<.0001	0.0204	0.0087	0.6135	0.6509
Fe × L	0.5200	0.8618	0.6790	0.0298	0.9475	0.1880	0.1069	0.5685	0.5381
V × Fe × L	0.6760	0.3486	0.6686	0.8620	0.5631	0.2544	0.0027	0.7482	0.1828
#S	<.0001	0.1725	<.0001	<.0001	<.0001	<.0001	0.6309	<.0001	0.6477
V × S	<.0001	0.745	0.0006	0.018	<.0001	0.2163	1.0000	0.1337	0.6271
Fe × S	0.2122	0.9670	0.6680	0.8117	0.2484	0.9912	1.0000	0.5359	0.4807
V × Fe × S	0.7508	0.3960	0.4864	0.7205	0.6678	0.9979	1.0000	0.9681	0.2481
L × S	<.0001	0.0078	<.0001	0.8494	<.0001	0.1051	1.0000	<.0001	<.0001
V × L × S	0.0002	0.0123	0.0016	0.1164	<.0001	0.9448	1.0000	0.7913	0.5150

Means followed by the same letter(s) within a column are not significantly different at $P \leq 0.05$ according to least significant difference (LSD) test.

*V= variety; @L=location; #S=season

Table 6. Effect of varieties on production and tissue Fe concentration of haricot bean

Parameters	Variety				CV (%)	LSD (5%)
	Nasir	Ibado	Hawassa Dume	Sari-1		
Plant height (cm)	45.50b	46.88a	43.80c	40.96d	10.74	1.35
No of branches per plant	4.82a	4.30 c	4.56b	4.81a	16.14	0.21
No of pods per plant	19.84a	11.66c	18.01b	17.56b	22.86	1.09
No of seeds per pod	5.64a	3.95b	5.50a	5.60a	12.88	0.19
Grain yield (kg ha^{-1})	3068.67a	2709.72b	3099.55a	2359.34c	20.29	160.48
Biomass (kg ha^{-1})	6931.4b	7558.6a	6985.7b	6371.5c	18.62	368.25
1000 seeds weight (g)	194.7b	485.49a	199.36b	190.71b	14.20	86.81
Leaf Fe (mg kg^{-1})	152.54b	167.12a	138.32c	148.80b	21.35	9.29
Seed Fe (mg kg^{-1})	86.46a	59.27b	60.69b	64.69b	16.41	20.32

Means followed by the same letter(s) within a row are not significantly different at $P \leq 0.05$ according to least significant difference (LSD) test.

Table 7. Influence of experimental location and season on production and tissue Fe concentration of haricot bean

	Location					Season		CV (%)	LSD (5%)
Yield components and tissue Fe	Butajira	Taba	Halaba	CV (%)	LSD (5%)	Belg	Meher		
Plant height (cm)	46.69a	45.40b	40.77c	10.74	1.17	48.27a	40.30b	10.74	9556
No of branches per plant	4.53b	4.35b	4.99a	16.14	0.18	4.68a	4.57a	16.14	0.15
No of pods per plant	20.21a	16.67b	13.42c	22.86	0.94	18.81a	14.72b	22.86	0.77
No of seeds per pod	5.17b	5.59a	4.80c	12.88	0.16	5.43a	4.94b	12.88	0.13
Grain yield (kg ha^{-1})	3014.5a	2718.5b	2620.01b	20.29	138.98	3253.2a	2315.5b	20.29	113.44
Biomass (kg ha^{-1})	7119.1a	6383.5b	7382.8a	18.62	1.97	9282.4a	4641.2b	18.62	260.39
1000 seeds weight (g)	231.69b	320.04a	250.97a	14.20	75.18	275.07a	260.07a	14.20	61.38
Leaf Fe (mg kg^{-1})	151.35b	91.24c	217.00a	21.35	8.05	179.43a	126.96b	21.35	6.57
Seed Fe (mg kg^{-1})	119.79a	72.61b	55.94b	16.41	17.6	84.4a	81.11a	16.41	14.37

Means followed by the same letter(s) within a row are not significantly different at $P \leq 0.05$ according to least significant difference (LSD) test.

The interaction between variety and season significantly influenced plant height (P<0.0001), number of pods per plant (P=0.0006) and grain yield (P<0.0001). All varieties better performed in terms of plant height, number of pods per plant and grain yield during belg than meher season (Fig. 5). The other yield components and concentrations of tissue Fe were not significantly influenced by the interactions of variety and season, though the concentration of leaf Fe in all varieties was higher during belg than meher. The concentration of seed Fe in Ibado and Hawassa Dume was higher during belg, while seed Fe in Nasir and Sari-1 was higher in meher season.

The interaction between variety and location significantly (P<0.0001) affected plant height, number of branches per plant, number of pods per plant, and grain and biomass yields (Table 5). A similar finding was reported on pods per plant of haricot bean in Uganda by Sebuwufu (2013). The strong interactions between varieties and locations indicate that the different varieties responded differently to local farm conditions, which could be attributable to the inherent genetic potential of the varieties and differences in soil conditions. All varieties performed better in plant height, number of pods per plant, and grain yield at Butajira, except Ibado, which yielded higher at Taba. The interaction between variety and location did not significantly influence tissue concentration of Fe, although the highest values of leaf and seed Fe of all varieties were observed at Halaba and Butajira, respectively (Fig. 6). A similar finding was reported by Imakumbili et al. (2010), on bean in Tanzania.

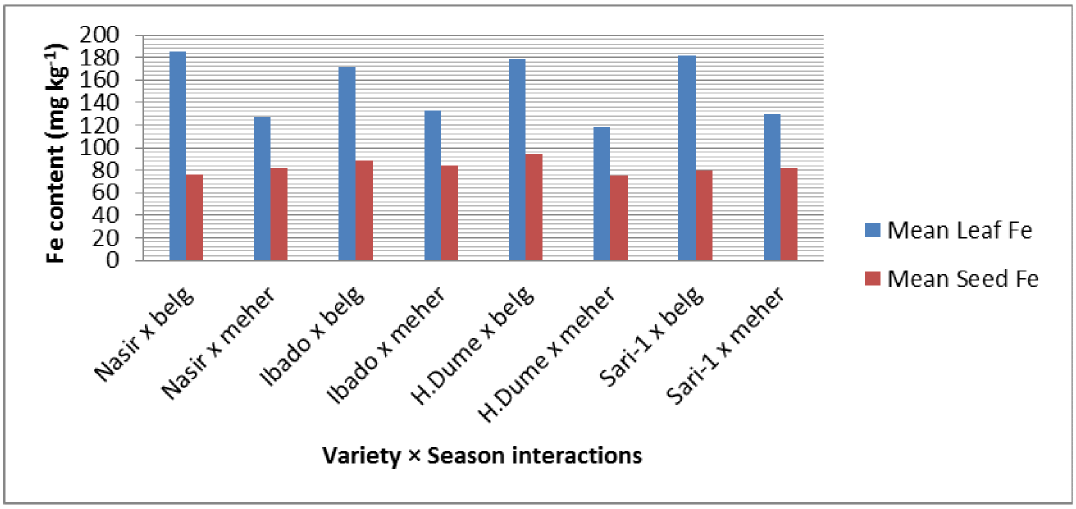

Fig. 5. Effect of variety × season on production and tissue Fe concentration of haricot bean

The interaction between location and season significantly (P<0.0001) influenced plant height, number of branches and pods per plant, grain yield, and concentrations of leaf and seed Fe. At Butajira, the tallest plant, highest number of branches and pods per plant were observed during meher season though the values during both seasons were relatively close. Conversely, grain yield and concentrations of leaf and seed Fe were higher during belg season (Fig. 7). At Taba, yield and yield components were higher during belg season, whereas tissue Fe concentrations were higher during meher season. The lower yield in Taba during meher season could be attributed to the higher rainfall in this season than belg (Ogola, 1991). In contrast, haricot bean performed better both in quantity and quality of production during belg season at Butajira and Halaba (Fig. 7). The concentrations of Fe were by far higher in leaves than in seeds of all varieties, at all locations, and levels of Fe fertilizers, suggesting that concentrations of plant minerals vary between plant tissues.

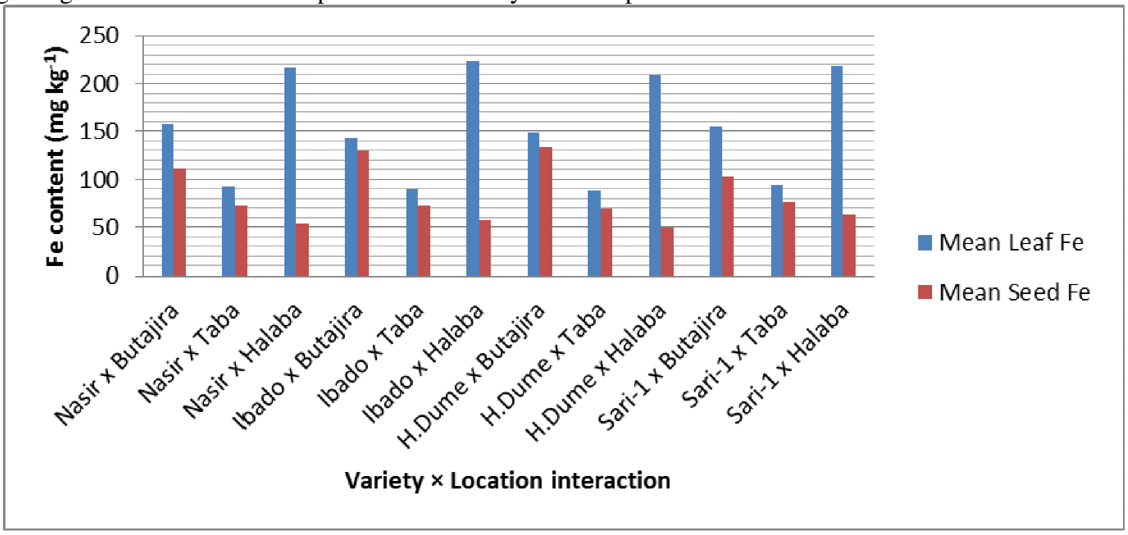

Fig. 6. Influence of variety × location on production and tissue Fe concentration of haricot bean

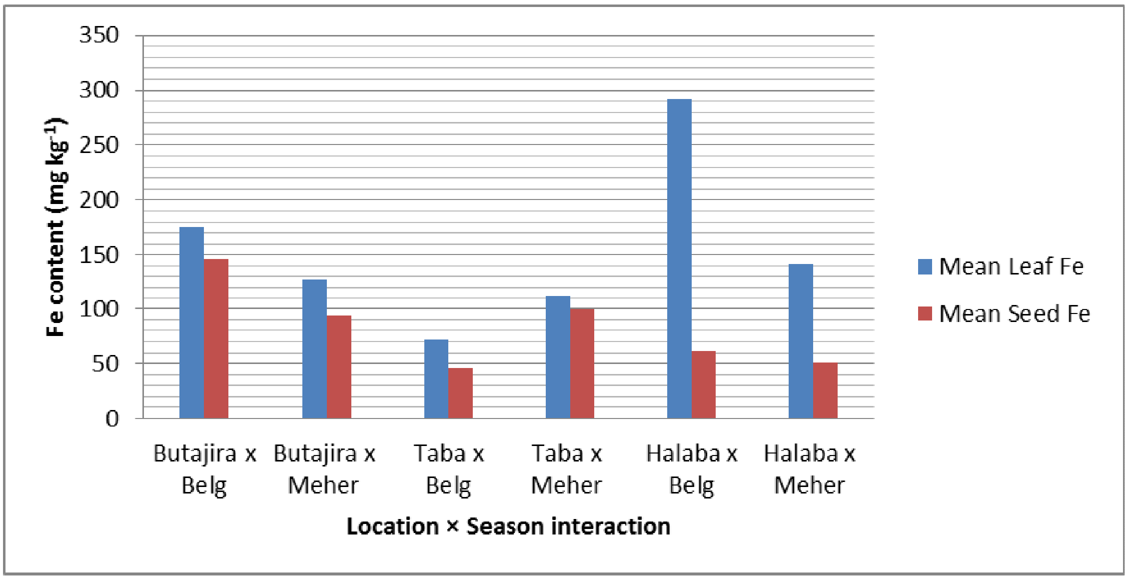

Fig.7. Influence of location × season on production and tissue Fe concentration of haricot bean

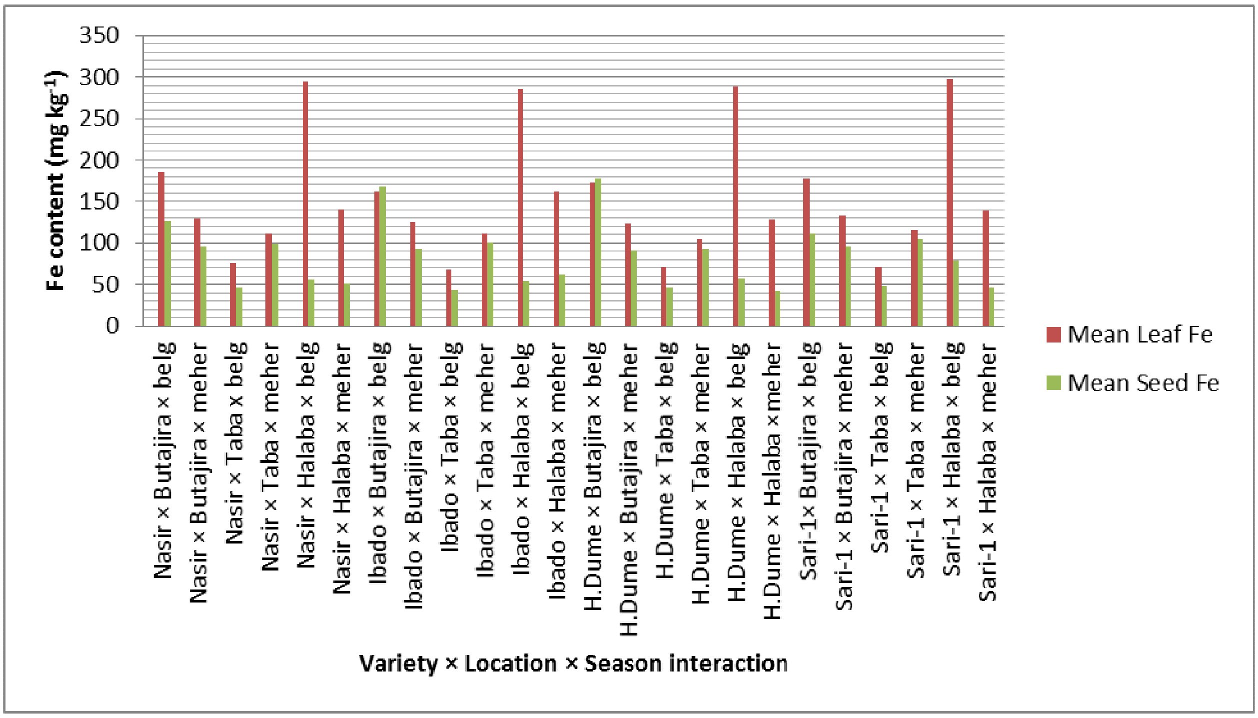

Fig. 8. Influence of variety × location × season on production and tissue Fe concentration of haricot bean

The interaction between varieties, location, and growing season significantly affected plant height (P=0.0002), number of branches (P=0.0123) and pods per plant (P=0.0016), and grain yield ($p<0.0001$) of haricot bean. At Halaba and Taba, all varieties performed better in all yield and yield components during belg season. At Butajira; Nasir, Hawassa Dume, and Sari-1 performed better in plant height during meher season, but their performance in grain yield was better during belg season. Conversely, Ibado performed better in plant height during belg but yielded higher grain during meher season. Tissue Fe concentrations were not significantly affected by the interaction of variety × location × season, although all varieties produced higher leaf and seed Fe during belg season at Butajira and Halaba, respectively. At Taba, however, all varieties resulted in higher tissue Fe concentrations during meher season (Fig. 8).

Conclusions

Both pot and field experiments revealed that all yield, yield components and tissue Fe concentrations varied significantly across locations. Generally, Fe fertilization resulted in best haricot bean production at Butajira both in quantity and quality, which could be attributed to lower available Fe and higher total nitrogen in the experimental soil as compared to the other locations. Yield, yield components and tissue Fe concentrations also varied significantly among haricot bean varieties. Nasir produced the highest grain yield and seed Fe concentration and was found to be best variety both in quality and quantity. The application of different levels of Fe fertilizer did not significantly influence yield and yield components of haricot bean varieties, but it significantly increased tissue Fe concentrations, the highest values being observed at 3% $FeSO_4.7H_2O$.

This study revealed that tissue Fe concentration varies among haricot bean varieties, and Fe fertilization enriches haricot bean with the element and hence improves its quality. Consequently, Nasir and 3% $FeSO_4.7H_2O$ were found to be the best variety and rate, respectively, for quality production of haricot bean. Therefore, growing Nasir at 3% $FeSO_4.7H_2O$ can contribute to alleviate the deficiency of Fe in people who consume the crop as a major component of their diet.

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