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**MICRONUTRIENTS FERTILIZATION AND THEIR CONCENTRATIONS IN PODS
AND SEEDS OF SOYBEAN**

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Abstract

Two field experiments were done at the research field of the Islamic Azad University of Kermanshah province, Iran at 2010-2011. The main objective of this study was to investigation the effects of zinc, iron and manganese applications on their concentrations in pods and seeds of soybean at seed filling period and maturity stages. Usage amounts of fertilizers Zn (0, 20 and 40 kg Zn ha⁻¹), Fe (0, 25 and 50 kg Fe ha⁻¹) and Mn (0, 20 and 40 kg Zn ha⁻¹) from source of ZnSo₄, FeSo₄ and MnSo₄, respectively. At the seed filling period and maturity stages, five plants were selected from each plot randomly. Samples washed with distilled water, dried, weighed, and were incinerated at 550⁰C. Finally, Zn, Fe and Mn concentrations were determined by Atomic Absorption Spectrometry. Based on results obtained, Zn concentration in soybean pods at both stages enhanced by zinc and reduced by iron and did not change by manganese application. Fe concentration was increased by application of 25 kg iron per hectare. With manganese fertilization up to 40 kg per hectare, Mn concentration in pods increased. In addition, zinc application up to 20 kg per hectare increased Mn concentration and in excess amount of zinc, Mn concentration was reduced. Also, Fe concentration in seeds was reduced by manganese use, severely.

Keywords: Iron, Manganese, Maturity, Seed Filling Period, Seeds

INTRODUCTION

In our previous studies we suggested that seeds in soybean plant constitute a system which play the role of final sinks for assimilates derived from the different organs of plant [1 and 2]. Therefore, it is expected that large amounts of micronutrients absorbed by the plant roots will be transferred to pods and ultimately to matured seeds. Processes of micronutrients transport from the soil solution to in the roots and partitioning throughout different parts of plant and finally translocation to the seeds are affected by soil condition [3] and plant growth stage [4]. Naturally, there are differences between micronutrients in the solubility in soil solution [5] and mobile ability in the plant organs [6 and 7] and remobilization from the vegetative tissues to reproductive organs [8]. In this regard, [9] reported that the efficiency of redistribution during germination and seedling stages for zinc, iron and manganese were ranged about 59-90% that depended to the mobility of element. In this study we centralized on zinc, iron and manganese concentrations in pods at seed filling period and maturity stages and in seeds at maturity stage. Indeed. Transfers of the micronutrients from the pods to seeds were studied. Thus, the main aim of this study was to investigation the effects of zinc, iron and manganese

applications on their concentrations in pods and seeds of soybean at seed filling period and maturity stages.

MATERIALS AND METHODS

Micronutrients concentration in plant organs affected by different levels of zinc, iron and manganese fertilization were tested by growing soybean Williams cultivar (*Glycine max* maturity group III) in field conditions at the Islamic Azad University of Kermanshah, Iran (34⁰23' N, 47⁰8' E; 1351 m elevation) for 2 years 2010 and 2011. Before planting, soil samples were collected from experimental area at 0-30 cm depth. The soil texture was silty clay with pH 7.6, total organic matter 2.3%, electrical conductivity (ECe) 0.61 dsm⁻¹, total nitrogen 0.18%, available phosphorus 9.9 mg kg⁻¹, available potassium 563 mg kg⁻¹, zinc 0.71, iron 6.2 and manganese 4.3 mg per kg soil respectively for 2010 and pH 7.4, total organic matter 2.1%, electrical conductivity (ECe) 0.52 dsm⁻¹, total nitrogen 0.14%, available phosphorus 10.1 mg kg⁻¹, available potassium 389 mg kg⁻¹, zinc 0.83, iron 3.6 and manganese 4.0 mg per kg soil respectively for 2011. The experimental design was a factorial experiment based on Randomized Complete Block with three replicates. Soybean seeds were initially soaked in 10% sugar solution, and then each

kg of seeds inoculated with 2 grams of *Bradyrhizobium japonicum*. Before planting, fertilizers were used as follows: 200 kg P₂O₅/ha and 50 kgN/ha and mixed with soil and land was ploughed once and harrowed twice. The plots consisted of six rows, 5 m in length spacing 60 cm apart. The distance between plants within a row was 5 cm and plant density achieved by over planting and thinning at V3 stage. Usage amounts of fertilizers zinc (0, 20, 40 kg.ha⁻¹ from ZnSo₄ source); iron (0, 25, 50 kg.ha⁻¹ from FeSo₄ source) and manganese (0, 20, 40 kg.ha⁻¹ from MnSo₄ source) were calculated based on plots area surface; next, fertilizers were mixed with soft soil at the ratio of 1:5 and placed on furrows made manually next to the stacks. At the seed filling period and maturity stages based on [10], five plants were selected from each plot randomly. Samples (pods and seeds, separately) washed with distilled water and were dried in the oven at 70^{0C} for 48 hours, weighed, and incinerated at 550^{0C}. Dry ash samples were soluble in concentrated HNO₃ and HCLO₄. Zn, Fe and Mn contents were determined by Atomic Absorption Spectrometry (AAS) according to [11]. All of data obtained from the measurements were evaluated statistically by MSTATC software. Combined variance analysis was performed after Bartlet test for checking uniformity of

data variance ($P=0.05$). LSD (Least Significant Difference Test) was used to means comparison. Excel software (version of 2007) was used to construct diagrams.

RESULTS AND DISCUSSION

Combined analysis of variance the impact of zinc, iron and manganese fertilizations on their concentrations in soybean pods and seeds at seed filling period and maturity stages was shown in **Table (1)**. Zn concentration in soybean pods at seed filling period stage affected by zinc, iron and manganese applications. Also, iron and manganese used had significantly effects on Fe concentration in pods at seed filling period stage. While, zinc application had no significantly effect on Fe concentration. Impact of zinc, iron, manganese and interaction effects of zinc with manganese (Zn×Mn) and iron with manganese (Fe×Mn) on Mn concentration in pods of soybean at seed filling period stage were significant. At maturity stage, Zn concentration influenced by zinc and iron applied. In contrast, manganese use had no effect on Zn concentration. Changes in Fe concentration in pods was affected by zinc at 5% level and iron and manganese at 1% levels. In addition, Mn concentration in pods at maturity stage was influenced by iron and manganese fertilizations at 1% levels ($P<0.01$). Zinc

applied had no significant effect on manganese concentration in pods at maturity stage. In soybean seeds, changes in Zn and Fe concentration were affected by zinc use. Iron application had significantly effects on Zn, Fe and Mn concentrations at maturity stage (**Table 1**). Impact of manganese fertilization on Fe and Mn concentrations in seed at maturity stage was significant at 1% levels. But Zn and Mn concentrations were not affected by manganese and zinc fertilizations, respectively. Interaction effects of zinc, iron and manganese on their concentrations in pod at seed filling period and maturity stages was shown in **Figure 1**. These results indicated that Zn concentration in soybean pods at both stages (seed filling period and maturity) enhanced by zinc and reduced by iron and did not change by manganese application. A similar result was reported by [12] that emphasized High concentrations of iron prevent the absorption of zinc and there is an antagonistic effect between these two elements. Furthermore, Fe concentration increased by application of 25 kg iron per hectare and with excess amounts of this element, Fe concentration was decreased. Zinc fertilization up to 40 kg per hectare had slightly effect on Fe concentration in pods, while Fe concentration in seeds reduced by manganese use, severely. Antagonistic effects

between iron and manganese were emphasized by [13]. Little changes in Mn concentrations in pods were observed when zinc was used. At maturity stage, iron fertilization up to 25 kg per hectare increased Mn concentration in soybean pods. Iron used in little amount, amended absorption and transfer of manganese in plant [14]. The effect of manganese application on Mn concentration was similar to what was observed in the impact of zinc fertilization on Zn concentration. Based on results obtained, with manganese fertilization up to 40 kg per hectare, Mn concentration in pods increased. This increase at seed filling period stage was more than the maturity stage. Similar findings have also been reported in other researches [15 and 16]. Zinc fertilization increased Zn concentration and decreased Fe concentration in soybean seed at maturity stage (**Figure 2**). In the other side, zinc application up to 20 kg per hectare increased Mn concentration and in excess amount of zinc, Mn concentration was reduced. Similar results were observed for the effects of iron application on Mn concentration. While, Fe concentration in soybean seed increased at different levels of iron application from 0 to 50 kg Fe ha⁻¹ (Fig 2). Iron and manganese fertilizations did not cause significant changes in Zn concentration. However, [17] reported that transfer of zinc to

other organs of plants enhanced by manganese fertilization. Different levels of manganese applications increased Mn concentration and decreased Fe concentration in soybean seeds at maturity stages, and the changes in the application of more than 20 kg Mn ha⁻¹ were intensified.

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Table 1: Combined analysis of variance the impact of zinc, iron and manganese fertilizations on their concentrations in soybean pods and seeds at seed filling period and maturity stages

Source of variation	df	Concentration in seeds								
		Seed filling period stage			Maturity stage			Maturity stage		
		[Zn]	[Fe]	[Mn]	[Zn]	[Fe]	[Mn]	[Zn]	[Fe]	[Mn]
Year (Y)	1	0.1	462.7	24.8	3.7	198.0	162.0	13.5	3.1	1561.1
Zn	2	2260.7**	3.8 ^{ns}	23.5*	615.5**	24.5*	8.3 ^{ns}	4874.1**	208.3**	30.1 ^{ns}
Y×Zn	2	89.1**	68.4**	129.1**	344.6**	96.9**	48.6**	61.6**	222.3**	271.7**
Fe	2	71.9**	3342.6**	136.9**	105.4**	1714.7**	378.8**	90.1**	8864.3**	177.8**
Y×Fe	2	79.2**	413.7**	21.6*	45.2*	521.8**	326.3**	3.9 ^{ns}	3131.3**	75.3**
Zn×Fe	4	7.9 ^{ns}	24.1**	4.3 ^{ns}	4.2 ^{ns}	40.7**	28.9*	6.2 ^{ns}	43.7**	35.6*
Y×Zn×Fe	4	9.7*	16.6**	30.7**	9.6 ^{ns}	12.3 ^{ns}	24.9*	38.1**	31.7**	44.4**
Mn	2	19.2**	208.7**	6543.7**	13.7 ^{ns}	81.4**	191.9**	15.7 ^{ns}	111.2**	24570.9**
Y×Mn	2	20.7**	77.5**	133.6**	16.1 ^{ns}	23.1*	702.0**	0.9 ^{ns}	97.5*	162.8**
Zn×Mn	4	6.9 ^{ns}	5.2 ^{ns}	17.8*	4.2 ^{ns}	2.1 ^{ns}	5.5 ^{ns}	1.2 ^{ns}	7.4 ^{ns}	7.5 ^{ns}
Y×Zn×Mn	4	2.6 ^{ns}	2.6 ^{ns}	30.2**	4.4 ^{ns}	3.1 ^{ns}	8.2 ^{ns}	5.5 ^{ns}	10.2 ^{ns}	83.2**
Fe×Mn	4	2.3 ^{ns}	9.2*	14.4**	3.6 ^{ns}	4.3 ^{ns}	14.8 ^{ns}	7.9 ^{ns}	25.4*	16.7 ^{ns}
Y×Fe×Mn	4	1.2 ^{ns}	12.6**	9.0*	0.6 ^{ns}	3.5 ^{ns}	18.3 ^{ns}	20.6**	15.7 ^{ns}	31.9*
Zn×Fe×Mn	8	1.8 ^{ns}	2.8 ^{ns}	10.1*	1.8 ^{ns}	1.9 ^{ns}	3.6 ^{ns}	3.8 ^{ns}	6.1 ^{ns}	4.1 ^{ns}
Y×Zn×Fe×Mn	8	2.6 ^{ns}	5.8 ^{ns}	4.9 ^{ns}	1.4 ^{ns}	2.8 ^{ns}	9.7 ^{ns}	3.8 ^{ns}	3.7 ^{ns}	17.6 ^{ns}
Error	104	3.7	3.2	2.9	5.3	6.8	8.5	5.1	7.3	10.2
Coefficient of variation (%)	-	6.4	6.6	8.3	10.6	10.2	12.6	7.1	9.7	7.7

-ns, * and **: non-significant, significant at 5% and 1% levels of probability, respectively

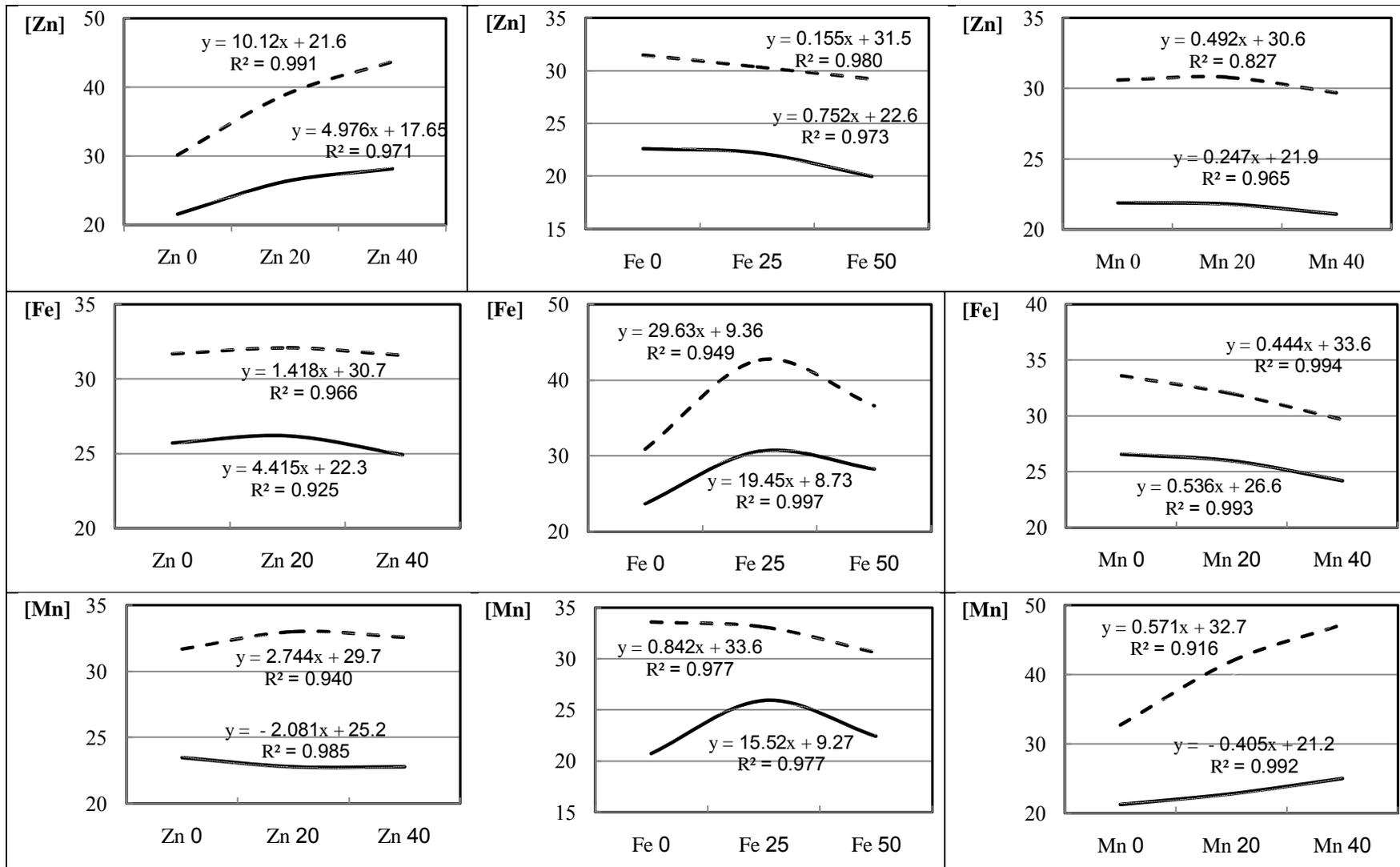


Figure 1- The impact of different levels of zinc, iron and manganese applications on Zn, Fe and Mn concentrations (mg.kg⁻¹ dry weight) in soybean pods at seed filling period stage (-----) and maturity stage (————)

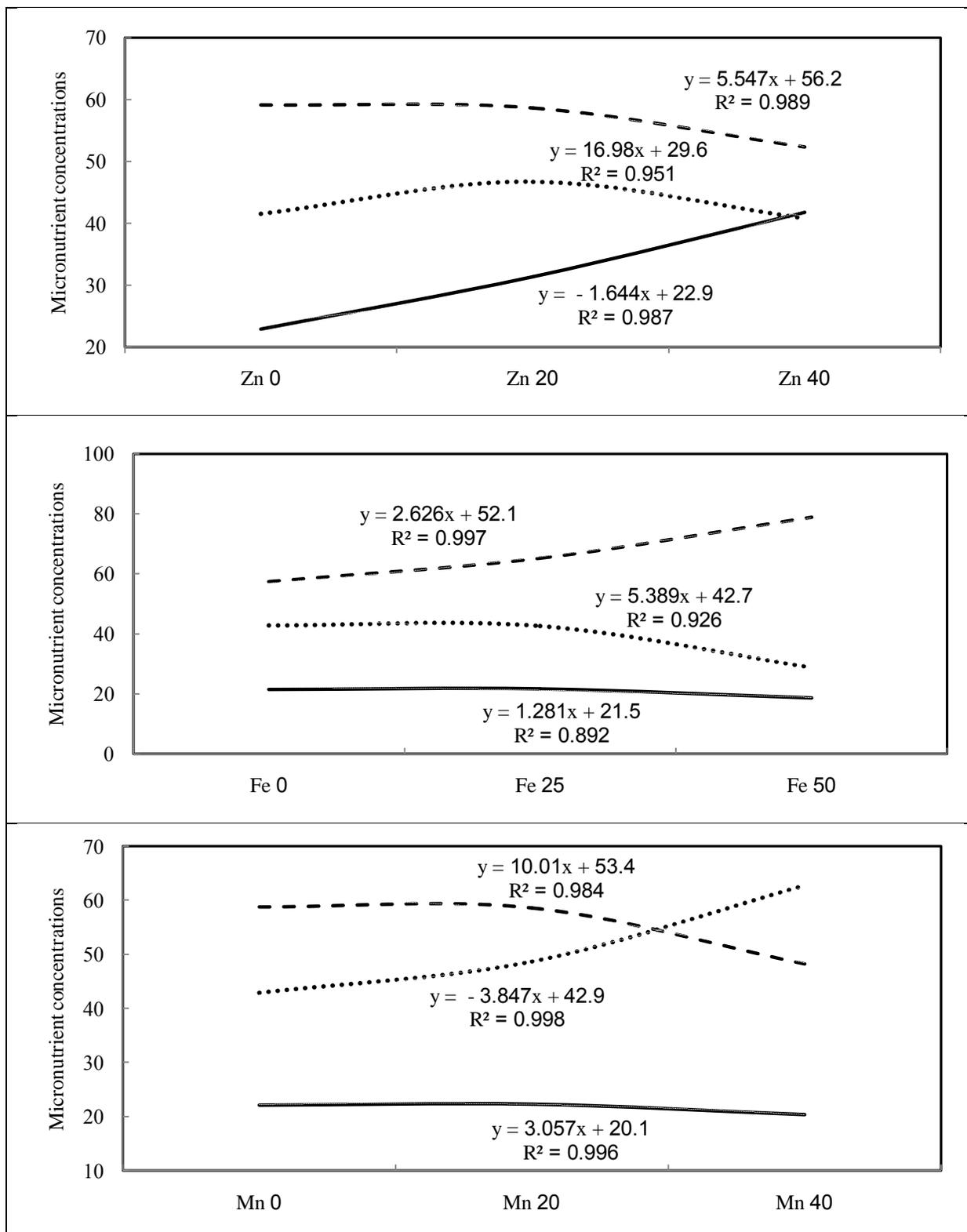


Figure 2- The impact of different levels of zinc, iron and manganese applications on Zn (———), Fe (- - - - -) and Mn (.....) concentrations (mg.kg⁻¹ dry weight) in soybean seed at maturity stage