Improvement for Better Nutrition on Rice through Iron Fertilizer Applications in Acid and Alkaline Soil under Greenhouse Conditions

การเพิ่มปริมาณสารอาหารในเมล็ดข้าวโดยการจัดการการให้ปุ๋ยเหล็กในดินกรดและดินด่าง ภายใต้สภาพโรงเรือนแบบเปิด

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ABSTRACT

Increasing iron (Fe) content in rice is an important global challenge due to high incidence of Fe deficiency in human populations. A pot experiment was conducted to evaluate the effect of Fe fertilizer application on concentration of Fe and Zn in brown rice, as well as grain yield of two rice cultivars (poor and rich Fe content) grown in acid and alkaline soil. The result revealed that Fe fertilizer application increased brown rice grain Fe concentration by 11.7% and 13.5% over control. Foliar Fe application was superior to soil Fe application. The rice cultivar Tabtim Chumpae produced significantly higher the Fe content than those of the cultivar Chainat 1. The response of Fe content has shown greater when the rice crop grown under alkaline soil. The Fe fertilizer application had no significant effect on grain yield. The result suggested that Fe fertilizer application represent a promising agricultural approach to reduce Fe deficiency in countries where rice is a main food crop consumed.

บทคัดย่อ

การเพิ่มความเข้มข้นของธาตุเหล็กในเมล็ดข้าวเป็นอีกความท้าทายที่สำคัญระดับโลก เนื่องจากปัจจุบันประชากรโลกประสบปัญหาร่างกายขาดแคลนธาตุเหล็กเป็นจำนวนมาก งานทดลองนี้จึงมีวัตถุประสงค์เพื่อศึกษาผลของการให้ปุ๋ยเหล็กต่อความเข้มข้นของธาตุเหล็กและธาตุสังกะสีในข้าวกล้องของข้าวสองสายพันธุ์ (มีความเข้มข้นธาตุเหล็กต่ำและสูง) ที่ปลูกในดินกรดและดินด่าง ผลการทดลองพบว่า การให้ปุ๋ยเหล็กช่วยเพิ่มความเข้มข้นของธาตุเหล็กในข้าวกล้องได้ถึง 11.7% และ 13.5% ซึ่งมากกว่าการไม่ให้ปุ๋ยเหล็ก โดยการให้ปุ๋ยเหล็กทางใบเพิ่มปริมาณธาตุเหล็กในข้าวกล้องได้มากกว่าการให้ปุ๋ยเหล็กทางดิน นอกจากนี้ยังพบว่าในข้าวพันธุ์ทับทิมชุมแพ สามารถเพิ่มความเข้มข้นของธาตุเหล็กได้สูงกว่าพันธุ์ชัยนาท1 และมีความเข้มข้นของธาตุเหล็กเพิ่มมากขึ้นเมื่อปลูกในดินด่าง การให้ปุ๋ยเหล็กไม่มีผลต่อผลผลิตข้าวทั้งสองพันธุ์ จากผลการทดลองชี้ให้เห็นว่า การให้ปุ๋ยเหล็กเป็นแนวทางในการจัดการทางการเกษตรที่สามารถช่วยลดการขาดแคลนธาตุเหล็กของมนุษย์ในกลุ่มประเทศที่บริโภคข้าวเป็นอาหารหลักได้

Keywords: Fe fertilizer application, Fe and Zn content in brown rice, Yield
คำสำคัญ: การให้ปุ๋ยเหล็ก ความเข้มข้นของธาตุเหล็กและสังกะสีในข้าวกล้อง ผลผลิต

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Introduction

Rice (*Oryza sativa* L.) is a principal source of food for more than half of the world population, it is the important source of energy (Juliano, 1995; Gregorio et al., 1999), vitamins and rare amino acids for people feeding on the rice as stable (Yong et al., 2008). However, there are low micronutrients content especially iron in grain. Inadequate intake with poor bioavailability of Fe in foods is in general the main cause of global Fe deficiency in humans (Fitzgerald et al., 2009). WHO reported that 3.7 billion people were iron-deficient and the problem was severe enough to cause anemia in 2 billion people. In this data, 40% were non-pregnant women and 50% were pregnant women. It has also been estimated that 31% of children fewer than 5 years olds were anemic, with mostly iron deficiency anemia (Gregorio et al., 1999; Gregorio, 2002; Welch and Graham, 2004).

Iron is one of the most abundant metals in the earth's crust, its availability to plant roots is very low. Fe availability depends on the soil redox potential and pH. In soil with low pH, Fe^{3+} is freed from the oxide, and becomes high solubility, more available for uptake by roots and absorbed available (Mengel and Kirkby, 1987; Marschner, 1995; Takahashi et al., 2001), if the iron concentration exceeds the optimum, resulting in toxic effects on plants. Whereas, the solubility of the Fe^{2+} and Fe^{3+} will be reduced when soil pH increases. In high pH, in soil has high Ca^{2+}concentration can evictions Fe^{3+} result in a high calcium chelate, Fe is readily oxidized and is predominately in the form of insoluble ferric oxides that resulting Fe^{3+} is unavailable to plants appeared a yellow card symptoms (Lindsay et al., 1967; Julian et al., 1983), and some staple crops, like rice, are especially susceptible to Fe deficiency due to 30% of the world's cropland is too alkaline for optimal plant growth (Marschner, 1995; Takahashi et al., 2001).

Such conditions as mentioned above, soil iron application maybe less efficient to plant, thus foliar iron application is the one method that could be much efficient and increase iron absorption by plant (Horesh and Levy, 1981).

Objective of this study

The objective of this research was to investigate the effect of different iron fertilizer application on iron content in grain and yield of two rice cultivars grown in acid and alkaline soil under open greenhouse conditions.

Materials and methods

Location and soil

The pot experiments were conducted in the Agronomy Experimental Farm, Department of Plant Science and Agricultural Resources, Faculty of Agriculture, Khon Kaen University (latitude 16º 28'N, longitude 102º48'E, 200 m above sea level) during wet season (June – December) 2017. A composite representative soil sample of acid and alkaline soil was collected from famous field at 0-30 cm depth and analyzed for physico-chemical properties as shown in Table 1.

Table 1 Soil physico-chemical properties contained in a plastic pot prior to rice seeding

Parameters	Acid soil	Alkaline soil
Physical characteristics		
Sand (%)	77.00	43.93
Silt (%)	15.00	24.07
Clay (%)	8.00	32.00
Textural class	Sandy loam	Clay loam
Chemical Characteristics		
pH (1:1 soil:water)	4.78	7.57
Total N (%)	0.043	0.114
Available P (mg/kg)	7.94	37.11
Exchangeable K (mg/kg)	40.28	90.63
Exchangeable Ca (mg/kg)	365	6390
Available Fe (mg/kg)	68.19	19.85
Organic matter (%)	0.634	2.611
CEC (c mol (+)/kg)	4.85	31.52

Soil preparation and seeding

The acid and alkaline soil used in the experiment was sun dried for 1 week, grinding to smaller size and contain in plastic pots 11 kg pot^{-1}. The plastic pots size was 24 cm in diameter and 20 cm in height. One seed of each rice cultivar was seeded into the soil at 3 cm depth.

Weather

The total amount of rainfall was 781.92 mm during the growing period. The highest maximum temperature (32.97 ^{o}C) was observed in August. While, the lowest minimum temperature (16.85 ^{o}C) exhibited in December 2017 as Table 2.

Table 2 Climatic data of the open greenhouse experimental site during the entire growing period in wet season 2017

Month	Rainfall (mm)	Temperature (°C)			Relative humidity (%)	Sunlight (h day^{-1})
		Minimum	Maximum	Average		
July	207.80	23.92	31.40	27.66	95	3.43
August	211.62	24.10	32.80	28.45	95	5.51
September	143.90	24.10	32.97	28.54	96	7.42
October	218.20	22.63	31.81	27.22	95	6.08
November	0.40	20.47	31.91	26.19	88	7.51
December	0.00	16.85	29.99	23.42	89	7.96
Total	781.92	132.07	190.88	161.48	558	37.91
Mean	130.32	22.01	31.81	26.91	93	6.32

Experimental design and Fe application

A pot experiment was arranged as a split–split plot design with four replications under open greenhouse conditions. The main-plot consists of acid soil and alkaline soil. The sub-plot consists of iron fertilizer application methods, viz. 1) control (no iron), 2) foliar solution of Fe (1.5 g Fe L^{-1}) using iron sulfate powder [$FeSO_4.7H_2O$ (1% Fe)] dissolved in distilled water was sprayed with a hand-held pump sprayer at the rate of 1,000 L ha^{-1} on plant leaves at two weeks after flowering, 3) soil application with Fe (5 kg Fe ha^{-1}) using iron sulfate powder [$FeSO_4.7H_2O$ (1% Fe)] sown into the soil at panicle initiation, and 4) soil applied iron sulphate powder at rate of 2.5 kg ha^{-1} at panicle initiation combined with foliar iron sulphate solution at rate of 500 L ha^{-1} at 2 weeks after flowering. Two rice cultivars namely, chainat 1 (grain low Fe content) and Tabtim Chumpae (grain high Fe content) were assigned as sub-sub plot.

Plant management

Water was applied by hand irrigation for seeds germination and during seedling growth at field capacity. Then, standing water was maintained at 5 cm depth above soil surface at 20 days after seeding until 10 days prior to harvest. For weed control, hand weeding was taken at 20 days after seeding. Fertilizer grade 46-0-0 (N) and 0-0-60 (K_2O) were applied at rate of 112.5 kg ha^{-1} and 37.5 kg ha^{-1} in acid soil, and 112.5 kg ha^{-1} and 18.75 kg ha^{-1} in alkaline soil two times at 30 days after seeding and at panicle initiation growth stage for all pots. Hand weeding was undertaken at 14, 30, 60 and 90 day after seeding.

Determination of yield and yield components

From each pot was measured to classify their panicle number, grain number per panicle, 1,000 grain weight, filled grain percentage and grain yield per pot at harvest time.

Iron and zinc content measurement in grain

Brown rice grain of 1 gram was analyzed for iron and zinc content by extracting brown rice flour with hydrocolic acid. Then the samples were read by Atomic Absorption Spectrophotometer (AAS), comparing the concentration of standard solution of iron and zinc (Zarcinas et al., 1987).

Statistical analysis

All the data, the analysis of variance (ANOVA) was used to distinguish the differences among treatments and compared the different means of each treatment by using STATISTIC8 program and comparison of mean square by LSD method at $P \leq 0.05$ levels.

Result

Fe content in brown rice grain

The Fe content in brown rice grain was significantly different among Fe fertilizer application methods at harvest. Soil application or foliar application produced significantly higher Fe content than that of non-Fe fertilizer control (Table 3). The cultivar Tabtim Chumpae gave significantly greater Fe content than those of the cultivar Chainat 1 (Table 3). For soil types, Fe content was significant higher when rice grown in an acid soil than those of alkaline soil in the present study (Table 3).

In the present experiment, an interaction effect between soil types and Fe fertilizer application methods was observed on Fe content in brown rice grain (Figure 1a). All of Fe fertilizer application methods under alkaline soil gave significant highest Fe content when compared with the control. While, the minimum Fe content was shown in foliar Fe fertilizer application method in acid soil.

Moreover, an interaction effect among soil types, Fe fertilizer application methods and cultivars also were noticed on Fe content in brown rice grain (Figure 1b). All of Fe fertilizer application methods under alkaline soil in the cultivar Chainat 1 gave significant highest Fe content when compared with the control, but in the cultivar Tabtim Chumpae, the foliar combined soil Fe application method gave lowest Fe content when compared with the control.

Table 3 Iron and zinc content in brown rice grain as affected by different iron fertilizer application methods of two rice cultivars grown under acid and alkaline soils in open greenhouse conditions

Treatment	**Iron content (mg/kg)**	**Zinc content (mg/kg)**
Soil type (S)		
Acid soil	32.10 a	26.41
Alkaline soil	28.32 b	24.16
F-test	*	ns
Iron fertilizer application method (F)		
Non-iron (control)	28.63 b	26.14
Foliar application	32.49 a	24.34
Soil application	31.97 a	25.62
Foliar combined soil application	27.74 b	25.04
F-test	**	ns
Rice cultivars (C)		
CNT1	27.85 b	26.14 a
Tubtim Chumpae	32.57 a	24.43 b
F-test	**	*
Interaction		
S x F	**	ns
S x C	ns	ns
F x C	ns	ns
S x F x C	*	ns

*, ** and ns = significant at 0.05 level, 0.01 level and not significant, respectively. Mean in the same column with different letters are significantly different at $p \leq 0.05$, as determined by LSD.

Zn content in brown rice grain

The application of Fe into the soil may be affected on Zn availability to the plant. Zn micronutrient also plays on important role for better health. Zn deficiency in humans is particularly problematic for women and children in developing countries, especially in children, such as impairments in physical growth, immune system, and learning ability, and causing DNA damage and cancer development (Boonchuay et al., 2013). In the present study, Fe fertilizer application method and soil type data not show significant difference on Zn content in brown rice grain, but significant difference was detected in the rice cultivars. The cultivars Chainat 1 produced significantly higher Zn content than those of the cultivar Tabtim Chumpae (Table 2).

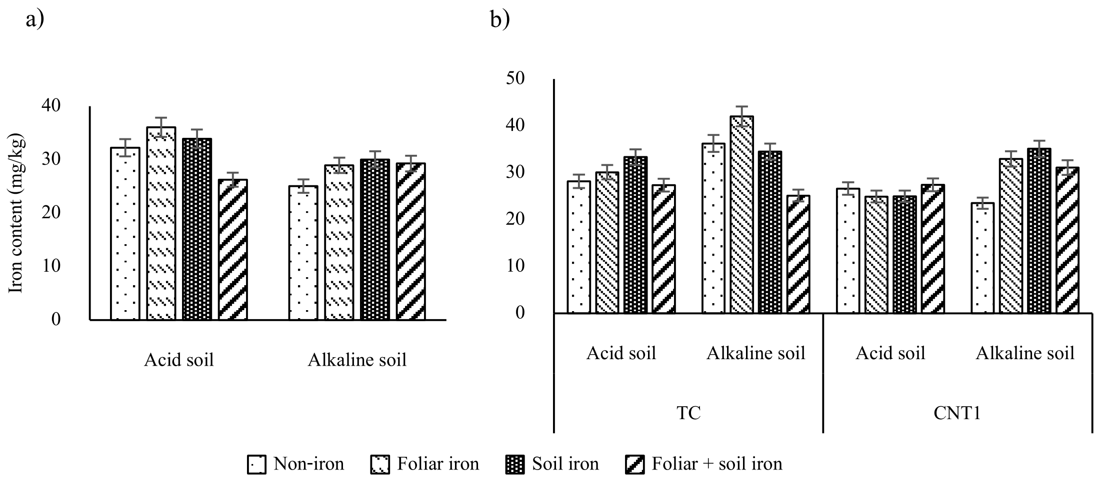

Figure 1 The interaction effect between soil types and iron fertilizer application methods (a) and interaction effect between soil types, rice cultivars and iron fertilizer application methods (b) for iron concentration in brown rice grain.

Yield and yield components

The panicle number and grain number per panicle were significantly affected by the soil types (Table 4). The maximum panicle number was obtained in the acid soil. While, the highest grain number per panicle was observed in the alkaline soil.

In the present experiment found that, the rice cultivars had a significant effect on grain yield, 1,000 grain weight and grain number per panicle (Table 4). The maximum grain yield and grain number per panicle were observed in the cultivar Tabtim Chumpae, while the maximum 1,000 grain weight was observed in the Chainat 1 cultivar.

In the present experiment, an interaction effect between soil types and Fe fertilizer application methods was observed on the panicle number (Table 4). The maximum panicle number was observed in soil Fe application and foliar combined soil Fe application methods in acid soil when compared with the control, but alkaline soil did not show the difference (Figure 2b).

An interaction between soil types and rice cultivars was observed on the panicle number and grain yield (Table 4). The maximum panicle number was observed in the acid soil in the cultivar Tabtim Chumpae (Figure 2c). While, the maximum grain yield was observed in alkaline soil in the cultivar Chainat 1 (Figure 3a).

An interaction between Fe fertilizer application methods and rice cultivars was observed on the panicle number and grain yield (Table 4). The maximum panicle number was observed in foliar Fe fertilizer application method in the cultivar Chainat 1 when compared with the control, while the cultivar Tabtim Chumpae, the maximum panicle number was observed in soil Fe application and foliar combined soil Fe application methods (Figure 2a). While, the maximum grain yield was observed in all Fe fertilizer application methods in the cultivar Tabtim Chumpae when compared with the control, but the cultivar Chainat 1 did not show difference (Figure 3b).

Moreover, an interaction effect among soil types, Fe fertilizer application methods and rice cultivars also were noticed on 1,000 grain weight (Table 4). The cultivar Chainat 1, the soil Fe fertilizer application gave lowest 1,000 grain weight in the acid soil when compared with the control. While, foliar Fe fertilizer application gave lowest 1,000 grain weight in acid soil in the cultivar Tabtim Chumpae (did not show the data).

Table 4 Yield and yield component as affected by different iron fertilizer application methods of two rice cultivars grown under acid and alkaline soils in open greenhouse conditions

Treatment	Panicle (no. plant^{-1})	Grain (no. panicle^{-1})	1,000 grains weight (g)	Filled grain (%)	Grain yield (g pot^{-1})
Soil type (S)					
Acid soil	11.38 a	86.42 b	24.72	75.55	22.53
Alkaline soil	8.33 b	111.39 a	25.53	79.00	23.86
F-test	*	*	ns	ns	ns
Iron fertilizer application (F)					
Non-iron (control)	8.58	93.69	25.70	78.23	22.01
Foliar application	9.83	96.32	24.57	74.74	24.64
Soil application	10.42	106.81	24.60	76.71	22.34
Foliar combined soil application	10.58	98.81	25.64	79.38	23.79
F-test	ns	ns	ns	ns	ns
Rice cultivar (C)					
CNT1	10.21	90.18 b	27.16 a	77.06	22.25 b
Tubtim Chumpae	9.50	107.64 a	23.09 b	77.48	24.14 a
F-test	ns	**	**	ns	*
Interaction					
S x F	*	ns	ns	ns	ns
S x C	**	ns	ns	ns	**
F x C	**	ns	ns	ns	**
S x F x C	ns	ns	*	ns	ns

*, ** and ns = significant at 0.05 level, 0.01 level and not significant, respectively. Mean in the same column with different letters are significantly different at $p \leq 0.05$, as determined by LSD.

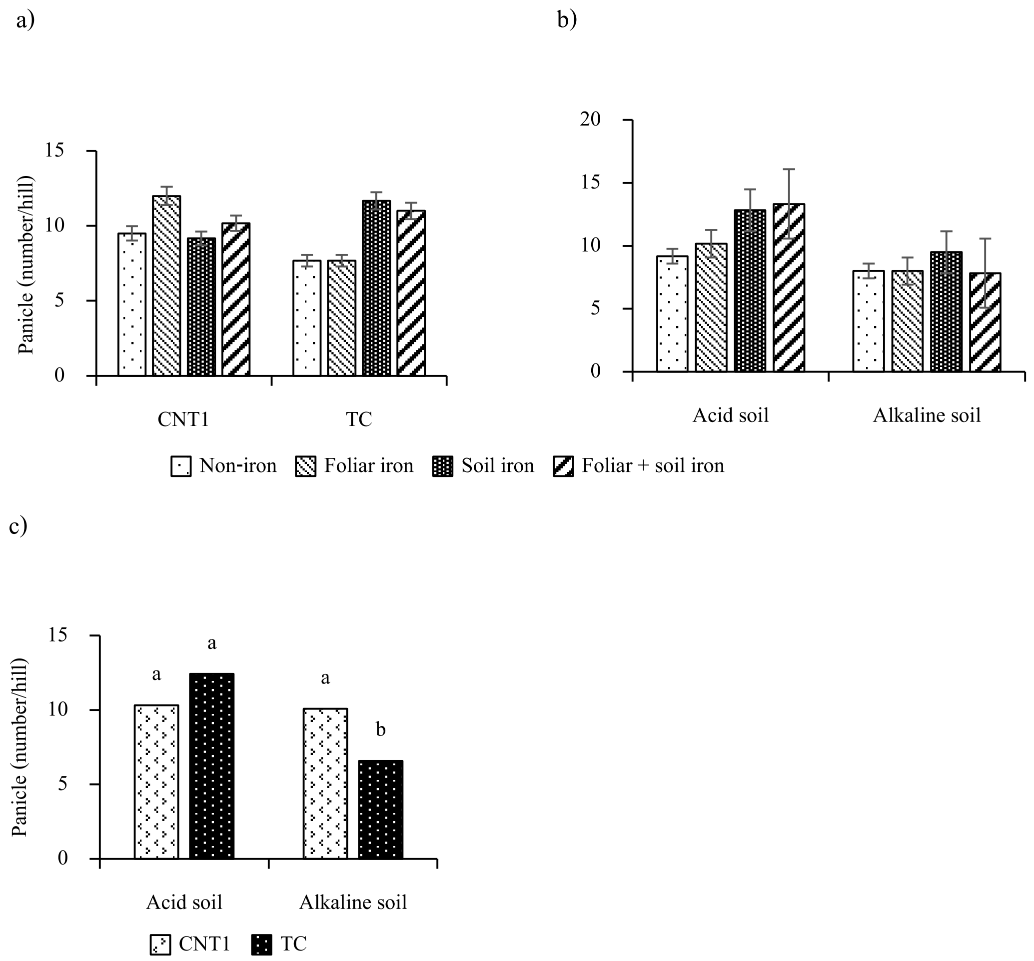

Figure 2 The interaction means between Fe fertilizer applications and rice cultivars (a), between Fe fertilizer application and soil types (b), and between soil types and rice cultivars (c) for number of panicle per hill. The same letter was not significant different at $p \leq 0.05$, as determined by LSD.

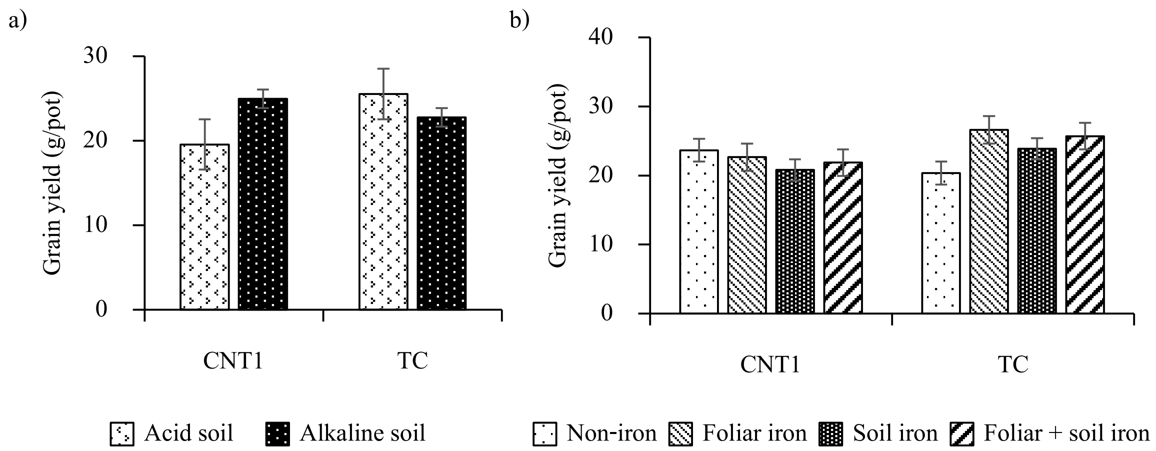

Figure 3 The interaction between soil types and rice cultivars (a), and between Fe fertilizer application and rice cultivars (b) for grain yield.

Discussion

Fe and Zn content in brown rice

The Fe fertilizer application was demonstrated as a short-term efficient approach to increase the Fe in brown rice. In the present experiment, single soil Fe application or single foliar application increased Fe content in brown rice grain by 11.7% and 13.5%, over control (non-Fe fertilizer), respectively. He et al. (2013) reported that foliar FeSO4.EDTA-FeNa, HEDTA-Fe, and DTPA-Fe sprays could remarkably improved the Fe concentration of polished rice, and the foliar application of DTPA-Fe significantly increased Fe concentration by 20.4%. The other research by Wei et al. (2012) stated that the foliar application of $FeSO_4$ plus nicotinamine (NA), and $FeSO_4$ plus $ZnSO_4$ increased the polished rice Fe concentration by 17.0%, 30.0% and 27.1%, respectively.

In general, foliar application of Fe was considered more effective than soil application, because soil application the soluble Fe was rapidly conversion into unavailable solid Fe (III) forms (Fageria et al., 2011). The Fe foliar applied increased Fe content in brown rice greater than soil application was reported by the previous research (Aciksoz et al., 2011). Due to leaf-applied substances can enter the leaf either by penetration of the cuticle or via the stomatal pathway, thus foliar fertilizer application is the one most effective application is increasingly used to moderate micronutrient deficiencies (Yong et al., 2008).

In the present experiment, the cultivar Tabtim Chumpae (Fe-rich cultivar) produced significantly higher the Fe content in brown rice grain than those of the Chainat 1 cultivar (Fe-poor cultivar). However, it revealed that the Chainat 1 cultivar gave higher Fe content in brown rice in Fe fertilizer application methods than those of non-Fe fertilizer application methods (control). Previous research has suggested that the foliar Fe application could improve the Fe content in brown rice grain of both common cultivars and Fe-rich cultivars (Yuan et al., 2012). The variations in term of increase Fe content in grain might come from the genetic control of ability absorption and seed deposition of Fe application (Wei et al., 2012)

In recent studies, it has been shown that Fe content was greatly response when rice received Fe fertilizer under alkaline soil rather than acid soil. This was probably attributed to acid soil consist of more available Fe than that of alkaline soil. In addition, when foliar Fe applied to calcareous soils the process of conversion of Fe into unavailable forms was not appeared.

For Zn content in brown rice grain, iron sulfate powder application [$FeSO_4.7H_2O$ (1% Fe)] did not show significantly increased Zn content in brown rice as compared to control in the present study. Similarly, with previous research by Wei et al. (2012) who reported that additional of $FeSO_4$ to foliar Fe application had no significant difference on Zn concentration in both of brown rice and polish rice grain, an addition of $FeSO_4$+NA+Zn to foliar Fe application increased both Fe and Zn content in brown rice and polish rice grain. However, previously study indicated that application of optimum Fe level could increase to the highest Zn level in rice and wheat grains (Yong et al. 2008; Pol et al., 2005). In this study, the cultivar Chainat 1 produced significantly higher Zn content than those of Tabtim Chumpae cultivar. Wei et al. (2012) reported that the cultivar different in Fe and Zn concentration was observed and may be due to the genetic control of leaf absorption and seed absorption of foliar application.

Grain yield

In the present experiment, the Fe fertilizer application methods did not show a significant difference on grain yield. This indicates that Fe fertilizer application in the form of [$FeSO_4.7H_2O$ (1% Fe)] can improved nutritional value of rice grain (Fe content), but not in term of grain yield per unit of land areas, similarly to has been reported by Yong et al. (2008) and Zhang et al. (2009). This might be due to Fe concentration in the soil prior application is sufficient for plant growth, similarly have been reported by Wei et al. (2012).

Conclusion

Nowadays, an anemia is the important problem is concern, thus increasing iron concentration in rice grains is the better satisfy human demands. In this study found that foliar and soil iron applications method could increase Fe content in brown rice grain. All Fe fertilizer applications; soil Fe application, Foliar Fe application and soil combined foliar Fe application could increase iron content in rice grain of the cultivar Chainat 1 and Tabtim Chumpae when grown in alkaline soil. Whereas, Fe fertilizer applications had not effected on grain yield. However, this present study found that all Fe fertilizer application methods could increase grain yield in the cultivar Tabtim Chumpae as compared to control. Thus, Fe fertilizer applications especially foliar Fe application might be a rapid method for improving the iron content in rice grain.

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