CHAPTER IV

RESULTS

4.1 Experiment 1: Effects of Fe toxicity on a Lao rice variety, TDK1

All of growth indicators (plant height, tiller number, root length leaf number and dry weight) showed the similar response to increasing Fe level. Growth was highest in control treatment, with no added Fe. Adding 1000 mg Fe_2SO_4 /kg soil (Fe1000) had no effect on plant growth. Plant growth was depressed by adding 2000 mg Fe_2SO_4 /kg soil (Fe2000).

Plant height

Plant height of the rice variety TDK1 was decreased with increasing Fe levels, with more pronounced effect at Fe2000 (Figure 1). Significant effect ($P < 0.05$) was only observed between Fe0 and Fe2000. The effect of Fe1000 was slight and was not significant from Fe0.

Leaf number

Leaf number of the rice variety TDK1 was decreased with increasing Fe levels, with more pronounced effect at Fe2000 (Figure 2). Significant effect ($P < 0.01$) was only observed between Fe0 and Fe2000. The effect of Fe1000 was slight and was not significant from Fe0.

Root length

Root length of the rice variety TDK1 was decreased with increasing Fe levels, with more pronounced effect at Fe2000 (Figure 3). Significant effect ($P < 0.01$) was only observed between Fe0 and Fe2000. The effect of Fe1000 was slight and was not significant from Fe0.

Tiller number

Tiller number of the rice variety TDK1 was decreased with increasing Fe levels, with more pronounced effect at Fe2000 (Figure 4). Significant effect ($P < 0.05$) was only observed between Fe0 and Fe2000. The effect of Fe1000 was slight and was not significant from Fe0.

Total dry weight

Total dry weight of the rice variety TDK1 was decreased with increasing Fe levels, with more pronounced effect at Fe2000 (Figure 5). Significant effect ($P < 0.01$) was only observed between Fe0 and Fe2000. The effect of Fe1000 was slight and was not significant from Fe0.

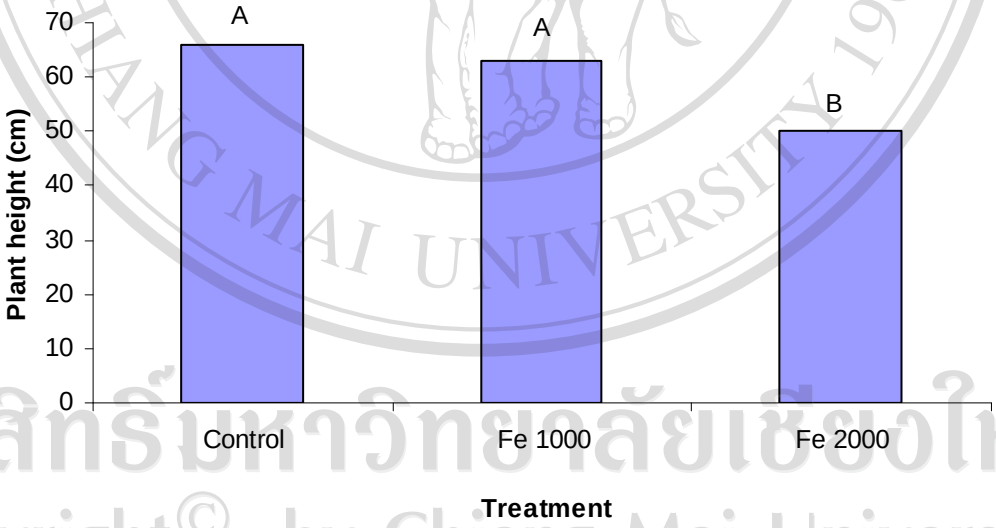

Figure 1. Effect of Fe treatment on plant height of TDK1 at 4 weeks after transplanting. Control = without adding Fe_2SO_4; Fe1000 = adding 1000 mg Fe_2SO_4 per kg soil and Fe2000 = adding 1000 mg Fe_2SO_4 per kg soil. Different letters above the bars indicate significant difference by LSD, at $P < 0.05$.

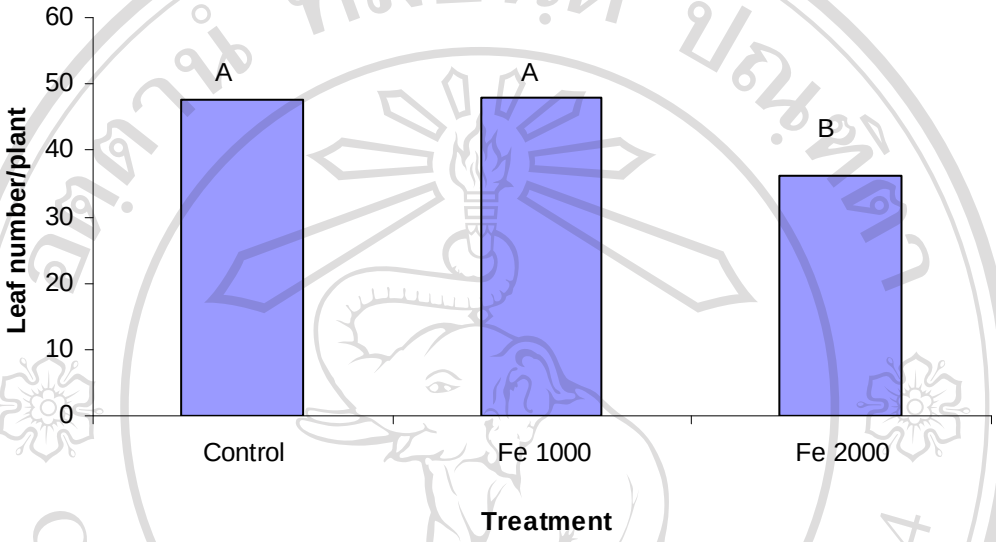

Figure 2. Effect of Fe treatment on leaf numer of TDK1 at 4 weeks after transplanting. Control = without adding Fe_2SO_4; Fe1000 = adding 1000 mg Fe_2SO_4 per kg soil and Fe2000 = adding 1000 mg Fe_2SO_4 per kg soil. Different letters above the bars indicate significant difference by LSD, at $P < 0.05$.

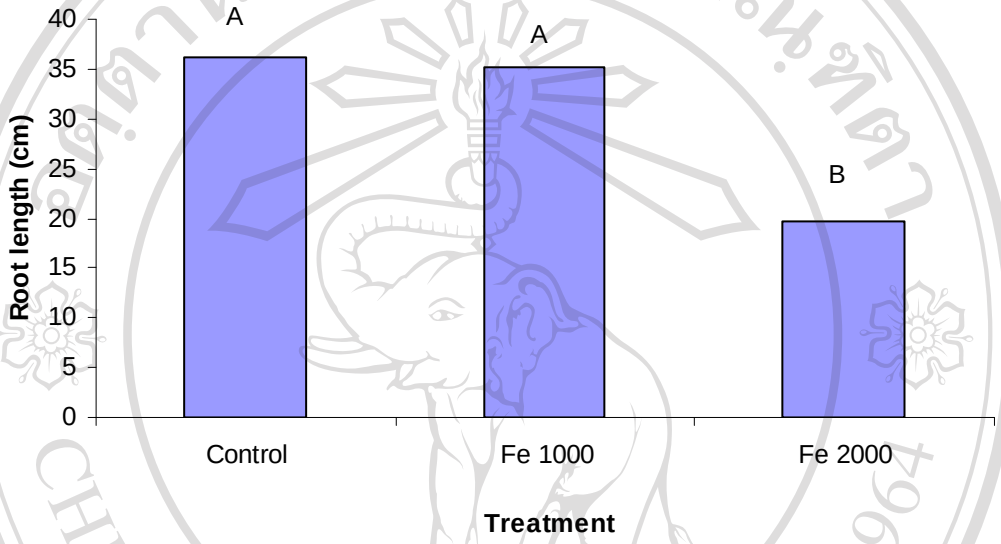

Figure 3. Effect of Fe treatment on root length of TDK1 at 4 weeks after transplanting. Control = without adding Fe_2SO_4; Fe1000 = adding 1000 mg Fe_2SO_4 per kg soil and Fe2000 = adding 1000 mg Fe_2SO_4 per kg soil. Different letters above the bars indicate significant difference by LSD, at $P < 0.05$.

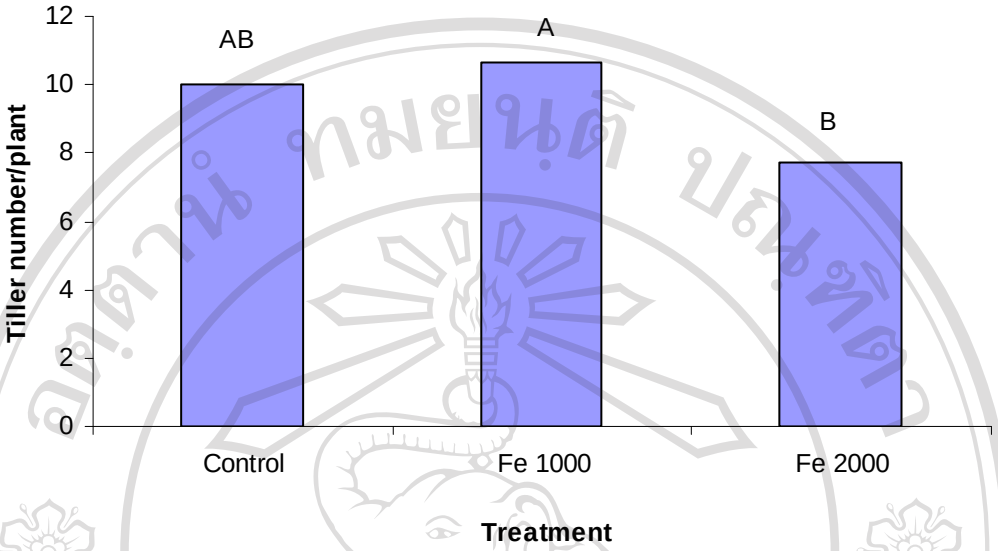

Figure 4. Effect of Fe treatment on tiller number of TDK1 at 4 weeks after transplanting. Control = without adding Fe_2SO_4; Fe1000 = adding 1000 mg Fe_2SO_4 per kg soil and Fe2000 = adding 1000 mg Fe_2SO_4 per kg soil. Different letters above the bars indicate significant difference by LSD, at $P < 0.05$.

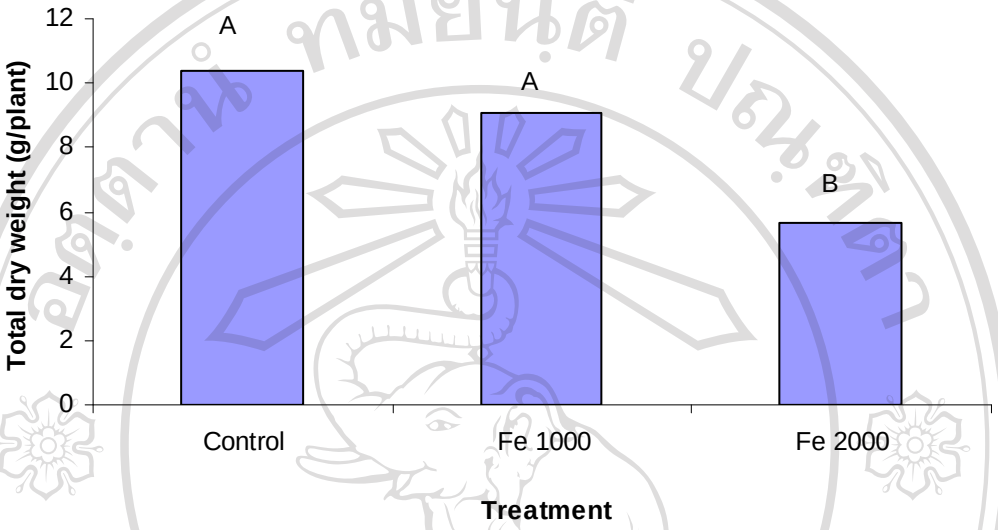

Figure 5. Effect of Fe treatment on total dry weight of TDK1 at 4 weeks after transplanting. Control = without adding Fe_2SO_4; Fe1000 = adding 1000 mg Fe_2SO_4 per kg soil and Fe2000 = adding 1000 mg Fe_2SO_4 per kg soil. Different letters above the bars indicate significant difference by LSD, at $P < 0.05$.

Root length correlated positively with plant dry weight ($P < 0.01$), plant height ($P < 0.01$), leaf number ($P < 0.01$) and tiller number ($P < 0.05$) (Table 1). Dry weight also correlated well with plant height ($P < 0.01$) and leaf number ($P < 0.01$) although it did not correlate with tiller number ($P < 0.05$), nor did tiller number correlate with plant height ($P < 0.05$)

Table 1. Correlation between different responses to Fe toxicity

parameter	DW	Plant height	Leaf number	Tiller number
Plant height	0.926**			
Leaf number	0.883**	0.772**		
Tiller number	0.679*	0.554^{ns}	0.756**	
Root length	0.942**	0.974**	0.821**	0.651*

NS= correlation not significant at $P < 0.05$, * significant at $P < 0.05$; ** significant at P<0.01.

4.2 Experiment 2: Screening Fe toxicity tolerance in different rice varieties.

This experiment found the variation among Lao rice varieties in their response to Fe in solution culture, with sufficient (20 ppm Fe, Fe20) and toxic (150 ppm Fe, Fe150) levels. At Fe20 the rice plants of all varieties grow very well, there was no symptom of Fe toxicity. At Fe150, the rice varieties showed different effects of Fe toxicity. In general, at Fe150 there was classic leaf symptom of tiny brown spots on the lower leaves starting from the tips and spreading toward the bases of the leaves. Responses to Fe toxicity were measured in Fe150 as percentage of measurement in Fe20. Ranking of varieties in their relative responses to Fe, however, changed with the different growth parameters.

Dry weight

The effect of Fe toxicity on total dry weight was clearly observed at two weeks after Fe treatment. There was significant difference ($P < 0.05$) in dry weight responses to Fe toxicity, measured as dry weight in Fe150 relative to Fe20, here termed 'relative dry weight' (Figure 6). RD10 had the highest relative dry weight among the 9 rice varieties, followed by TDK5, TDK11 and TDK6, with only slightly lower relative dry weight. The remaining 5 varieties had significantly lower relative dry weight than RD10, with TDK1, MNG and TDK7 having the lowest relative dry weight of less than 40% compared with more than 60% in RD10.

Plant height

The effect of Fe toxicity on plant height was clear at two weeks after treatment. There was significant difference ($P < 0.01$) in the plant height response to Fe toxicity among the 9 rice varieties, measured as height in Fe150 relative to Fe20, here termed 'relative height' (Figure 7). TDK5 and TDK6 had the highest and MNG

the lowest relative height. The relative height of the other 6 varieties, namely, TDK10, IR70617, RD10, TDK11, TDK1 and TDK7, is falling in between.

Tiller number

After two weeks of treatment, tiller number of the 9 different rice varieties was affected different by Fe toxicity, as measured by number of tillers in Fe150 relative to Fe20, here termed 'relative tiller number'. Iron toxicity clearly depressed tillering, with relative tiller number ranging from 20-30%. There was significant difference among the rice varieties ($P < 0.05$) in their tillering response to Fe toxicity (Figure 8). TDK10 had the highest relative tiller number at more than 30%, with TDK1 and TDK7 having significantly lower relative tiller number at 20% ($P < 0.05$). The other 6 varieties, namely, TDK5, TDK11, RD10, IR70617, TDK6 and MNG fell into the intermediate class in their tillering response to Fe toxicity.

Root length

After two weeks of treatment, root length of the 9 different rice varieties was affected differently by Fe toxicity, as measured by root length in Fe150 relative to Fe20, here termed 'relative root length. Iron toxicity clearly depressed root length ($P < 0.01$), with relative root length ranging from 65-90%, with marked differentiation among the varieties into TDK1 with the highest relative root length, followed by TDK11, TDK6 and RD10 with slightly lower relative root length. Of the remaining 5 varieties, TDK7 and TDK10 had significantly lower relative root length than TDK1, followed by IR70617, and even lower relative root length in TDK5 and MNG.

Leaf number

The effects of Fe toxicity in leaf number, pronounced after two weeks of treatment. The 9 different rice varieties was affected different by Fe toxicity, while measured as a result of leaf number in Fe150 relative to Fe20, here termed 'relative leaf number. Iron toxicity obviously depressed leaf number ($P < 0.01$), with relative leaf number ranging from 27-45%, with marked differentiation among the varieties into RD10, and however four rice varieties were not significant difference as TDK5, TDK10, TDK11 and IR70617. MNG with the lowest relative leaf number and two rice varieties as TDK1 and TDK7 were not significant difference with MNG.

Leaf Bronzing

The effect of Fe toxicity on leaf symptom was obvious at two weeks after treatment. There was significant difference ($P < 0.01$) in the leaf symptom response to Fe toxicity among the 9 rice varieties measured as leaf bronzing index (%LBI)' (Figure 11). The LBI values in the different rice varieties ranged from 27-58%, with marked differentiation among the varieties into TDK10 and RD10 with the lowest in %LBI. Of the remaining rice varieties, TDK7 and MNG had the highest %LBI value, followed by TDK6, TDK1, TDK5, IR70617, and TDK11, in that order.

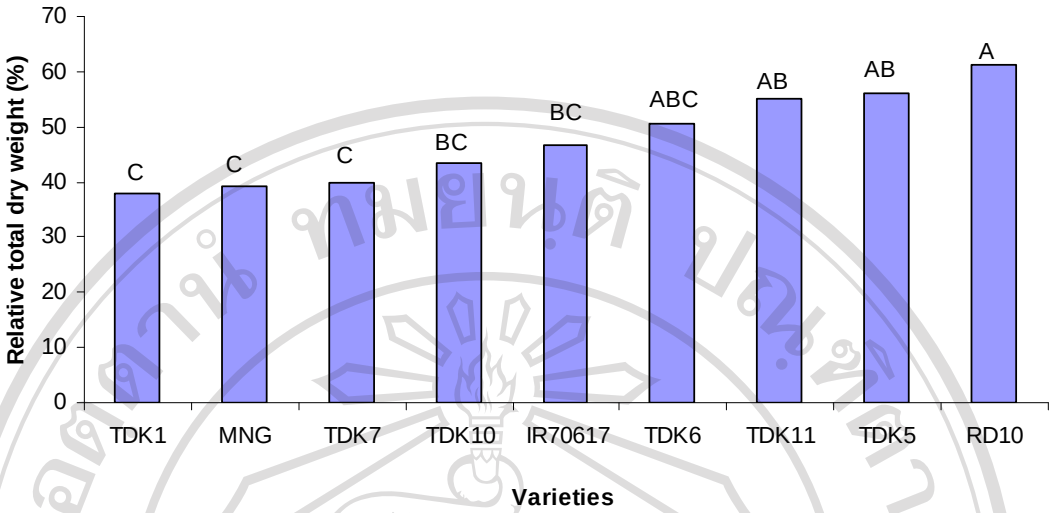

Figure 6. Total dry weight in Fe150 relative to Fe20, % in 9 rice varieties. Different letters above bars designate significant difference by LSD at $P < 0.05$.

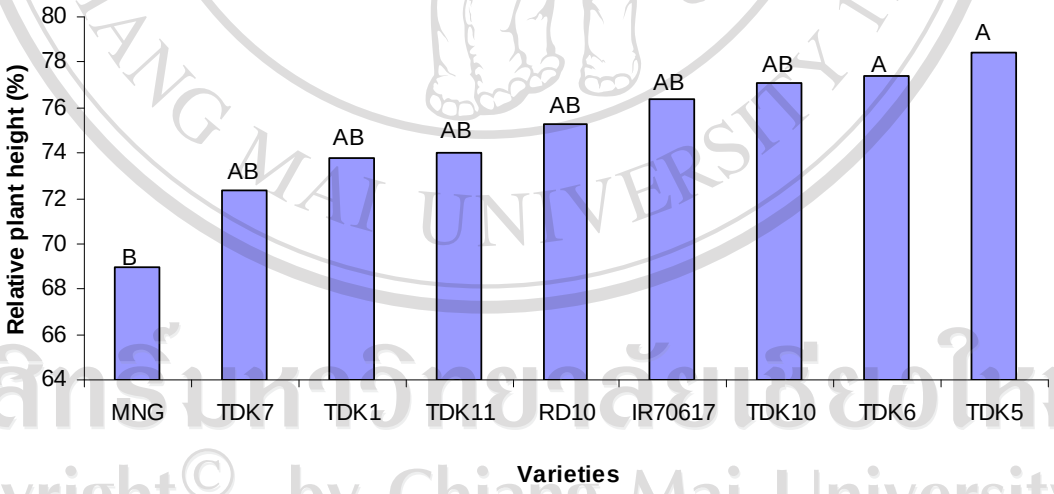

Figure 7. Plant height in Fe150 relative to Fe20, % in 9 rice varieties. Different letters above bars designate significant difference by LSD at $P < 0.05$.

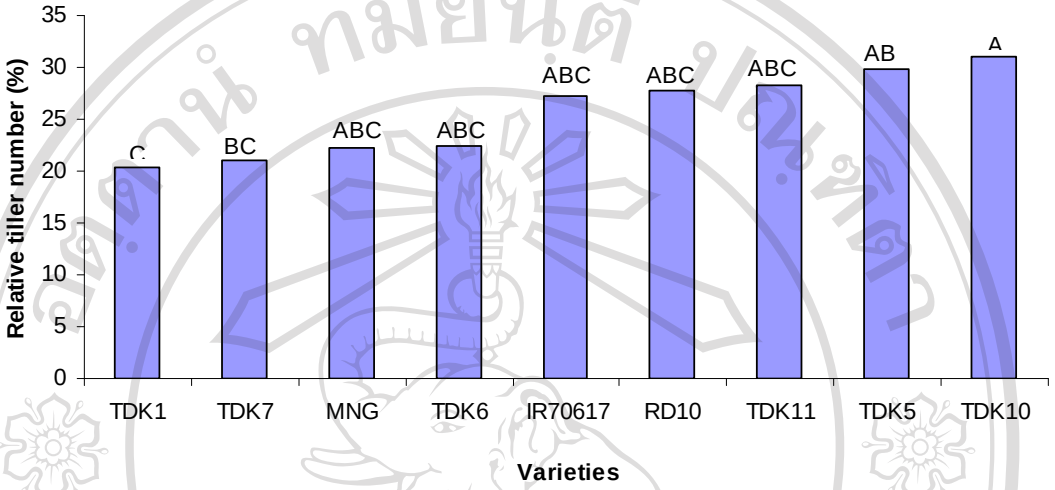

Figure 8. Tiller number in Fe150 relative to Fe20, % in 9 rice varieties. Different letters above bars designate significant difference by LSD at $P < 0.05$.

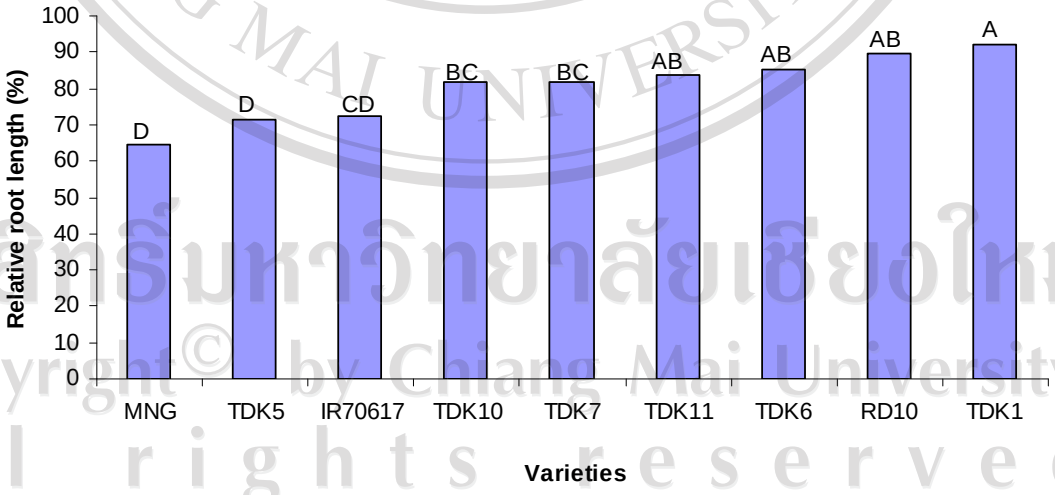

Figure 9. Root length in Fe150 relative to Fe20, % in 9 rice varieties. Different letters above bars designate significant difference by LSD at $P < 0.05$.

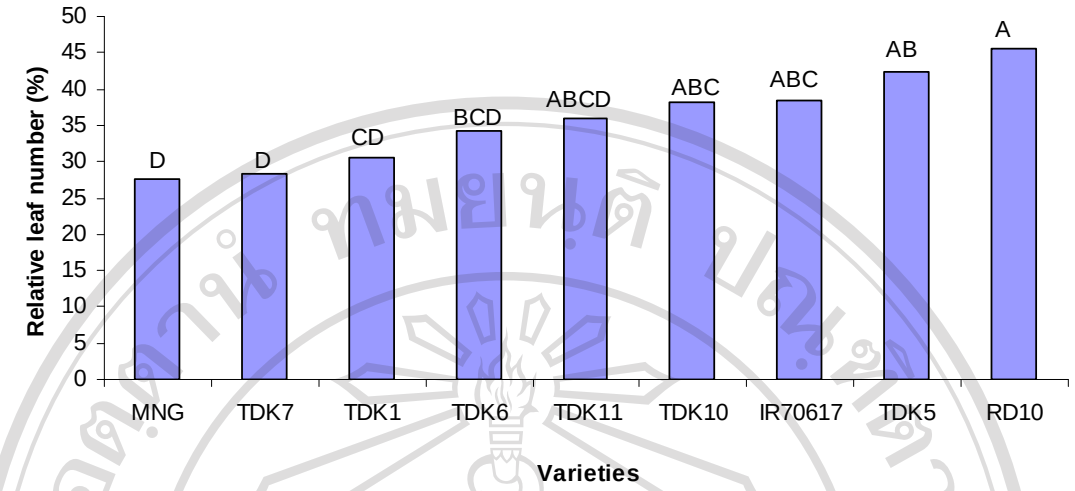

Figure 10. Leaf number in Fe150 relative to Fe20, % in 9 rice varieties. Different letters above bars designate significant difference by LSD at $P < 0.05$.

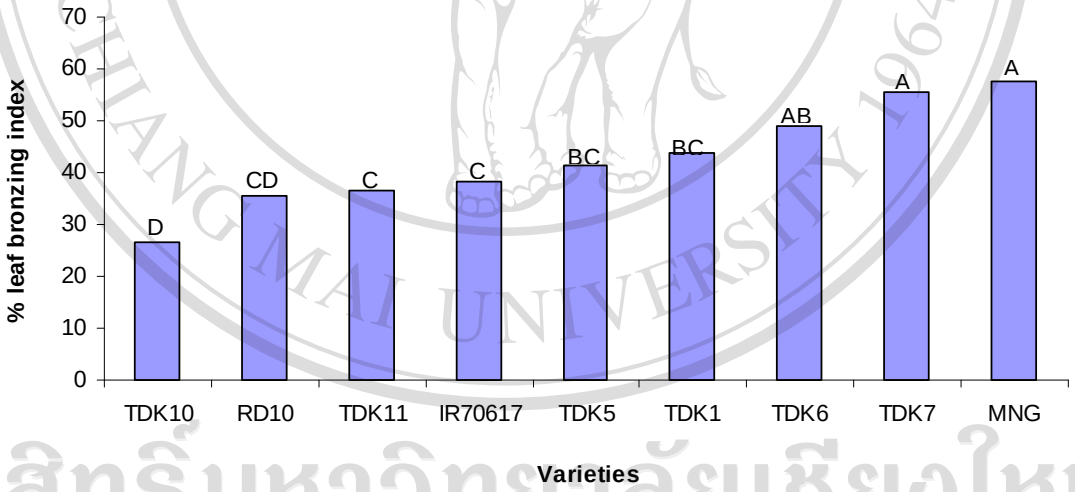

Figure11. %LBI in Fe150 in 9 rice varieties. Different letters above bars designate significant difference by LSD at $P < 0.05$.

Correlation between different responses to iron toxicity

Correlation between responses to Fe toxicity in the different parameters of growth showed differential responses in different growth processes (Table 2). Relative tiller number was not significantly ($P < 0.05$) correlated with relative dry weight and relative root length, but it was significantly correlated with relative plant height, LBI and relative leaf number. The LBI was not significantly ($P < 0.05$) correlated with relative dry weight and relative root length, it was significantly correlated with relative tiller number, relative plant height and relative leaf number. Relative plant height was significantly ($P < 0.05$) correlated with all parameters of Fe toxicity response. Relative leaf number was significantly ($P < 0.05$) correlated with relative plant height, LBI and relative tiller number, but it was not significantly correlated with relative dry weight and relative root length.

Table 2. Correlation between different responses to Fe toxicity

Response to Fe toxicity[1]	RDW	RPH	LBI	RLN	RRL
RPH	0.377*				
LBI	ns	-0.575**			
RLN	ns	0.747**	-0.54**		
RRL	0.327*	0.324*	ns	ns	
RTN	ns	0.777**	-0.44**	0.940**	ns

[1]RDW = dry weight in Fe150 relative to Fe20; RPH = height in Fe150 relative to Fe20; LBI = leaf bronzing index; RLN = leaf number in Fe150 relative to Fe20; RRL = root length in Fe150 relative to Fe20; RTN = tiller number in Fe150 relative to Fe20. ns = not significant, $P < 0.05$; * significant at $P < 0.05$ and and ** significant at $P < 0.01$.

Fig.12: All 9 rice varieties at 20 mg Fe L^{-1} (Fe20) 10 days after treatment.

Fig. 13: All 9 rice varieties at 150 mg Fe L^{-1} (Fe150) 10 days after treatment.

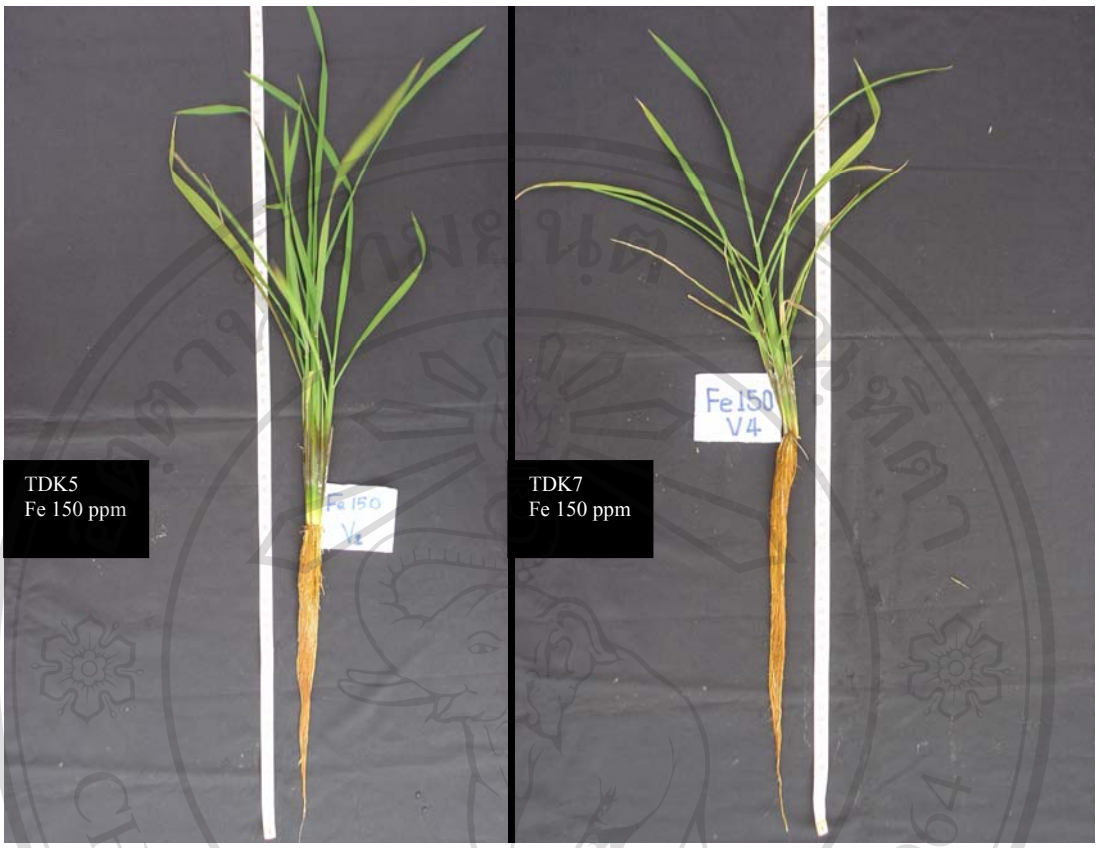

Fig. 14: Comparing between Fe toxicity sensitive rice variety (TDK7) and Fe toxicity tolerant (TDK5) rice variety

4.3 Experiment 3: Growth and yield of rice varieties with different sensitivity to Fe toxicity in the field with Fe toxicity problem in Vientiane, Laos.

Harvest 1

Leaf Bronzing

The Fe toxicity symptoms of the rice varieties TDK5, TDK7 and TDK10 appeared on the lower leaves, starting at the tips. Later, the entire leaf turns brown, purple, yellow and orange. TDK5 had lower Leaf Bronzing Index (LBI) than TDK7 and TDK10 (Table 3). Root dipping with Zn had no significant effect on LBI of all 3 varieties. Foliar Zn had not yet been applied when leaf bronzing was assessed.

Table 3. Leaf bronzing index (%) in 3 rice varieties with 2 Zn treatments in a lowland rice field affected by Fe toxicity, at one month after transplanting.

Variety	Zn application treatment		Mean
	Control	Dipped	
TDK5	36.25	38.75	37.50B
TDK7	51.25	52.50	51.87A
TDK10	48.75	52.50	50.62A
Mean	45.41	47.91	
F-test	Variety**	Treatment^{ns}	Variety x Treatment^{ns}

** Significant at $P < 0.01$; ns = not significant at $P < 0.05$
Different letters after numbers designate significant difference by $LSD_{0.05}$;
$LSD_{0.05}$ for comparing between variety means = 5.023

Control = no Zn application, Dipped = dipping roots seedling in 1% w/v $ZnSO_4$ solution. The mean in same column followed by different capital letters are significant different at $p \leq 0.05$.

Plant height

The result of this experiment showed no significant interaction between rice varieties and Zn treatment ($P < 0.05$). However, there were significant differences between rice varieties. TDK10 tended to be taller than TDK5 and TDK7 (Table4). There was also significant Zn treatment effect, in which spraying tended to increase plant height and root dipping tended to decrease plant height.

Table 4. Plant height (cm) in 3 rice varieties with 2 Zn treatments in a lowland rice field affected by Fe toxicity, at one month after transplanting.

Variety	Zn application treatment		Mean
	Control	Dipped	
TDK5	43.45	42.70	43.07AB
TDK7	44.05	36.20	40.12B
TDK10	44.15	44.60	44.37A
Mean	43.88a	41.16b	
F-test	Variety*	Treatment*	Variety x Treatment^{ns}

* Significant at $P < 0.05$; ns = not significant at $P < 0.05$
Different letters after numbers designate significant difference by $LSD_{0.05}$
$LSD_{0.05}$ for comparing between variety means = 3.315
$LSD_{0.05}$ for comparing between Treatment means = 2.707

Control = no Zn application, Dipped = dipping roots seedling in 1% w/v $ZnSO_4$ solution. The mean in same column followed by different capital letters are significant different at $p \leq 0.05$, the mean in same row followed by different small letters are significant different at $p \leq 0.05$.

Tiller number

There was no significant effect ($P < 0.05$) of rice varieties and Zn treatments on tiller number (Table5).

Table 5. Tiller number (Tiller/plant) in 3 rice varieties with 2 Zn treatments in a lowland rice field affected by Fe toxicity, at one month after transplanting.

Variety	Zn application treatment		Mean
	Control	Dipped	
TDK5	5.10	5.20	5.15
TDK7	5.00	5.00	5.00
TDK10	4.70	4.30	4.50
Mean	4.93	4.83	
F-test	Variety^{ns}	Treatment^{ns}	Variety x Treatment^{ns}

ns = not significant at $P < 0.05$

Control = no Zn application, Dipped = dipping roots seedling in 1% w/v $ZnSO_4$ solution.

Harvest 2

Plant height

By harvest 2 there was no interaction ($P < 0.05$) between the effect of rice varieties and Zn treatment. However, there were significant ($P < 0.01$) differences in height between the 3 rice varieties. TDK5 was taller than TDK7 and TDK10 (Table6). There was also significant Zn treatment effect, in which both Zn spraying and root dipping tended to decrease plant height.

Table 6. Plant height (cm) in 3 rice varieties with 3 Zn treatments in a lowland rice field affected by Fe toxicity, at two months after transplanting.

Variety	Zn application treatment			Mean
	Control	Spray	Dipped	
TDK5	91.25	85.30	78.90	85.15A
TDK7	72.10	66.30	62.50	66.96B
TDK10	85.10	66.15	59.50	70.25B
Mean	82.81a	72.58b	66.96b	
F-test	Variety**	Treatment**	Variety x Treatment^{ns}	

** Significant at $P < 0.01$; ns = not significant at $P < 0.05$
Different letters after numbers designate significant difference by $LSD_{0.05}$
$LSD_{0.05}$ for comparing between variety means = 6.972
$LSD_{0.05}$ for comparing between Treatment means = 7.232

Control = no Zn application, Dipped = dipping roots seedling in 1% w/v $ZnSO_4$ solution and Spray = spraying 0.5% w/v $ZnSO_4$ solution on the top of rice leaves at 30 days after transplanting. The mean in same column followed by different capital letters are significant different, the mean in same row followed by different small letters are significant different at $P \leq 0.05$.

Tiller number

There was no significant effect ($P < 0.05$) of rice varieties and Zn treatments on tiller number (Table 7).

Table 7. Tiller number (Tiller/plant) in 3 rice varieties with 3 Zn treatments in a lowland rice field affected by Fe toxicity, at two months after transplanting.

Variety	Zn application treatment			Mean
	Control	Spray	Dipped	
TDK5	10.95	10.20	13.00	11.38A
TDK7	10.95	9.95	11.00	10.63AB
TDK10	9.65	7.55	10.40	9.20B
Mean	10.51ab	9.23b	11.46a	
F-test	Variety*	Treatment*	Variety x Treatment^{ns}	

* Significant at $P < 0.05$; ns = not significant at $P < 0.05$
Different letters after numbers designate significant difference by $LSD_{0.05}$
$LSD_{0.05}$ for comparing between variety means = 2.159
$LSD_{0.05}$ for comparing between Treatment means = 3.099

Control = no Zn application, Dipped = dipping roots seedling in 1% w/v $ZnSO_4$ solution and Spray = spraying 0.5% w/v $ZnSO_4$ solution on the top of rice leaves at 30 days after transplanting. The mean in same column followed by different capital letters are significant different, the mean in same row followed by different small letters are significant different at $p \leq 0.05$.

Total dry weight

Significant ($P < 0.05$) difference in plant dry weight was only found between the rice varieties, with no effect of Zn treatments (Table 8) TDK5 had higher dry weight than TDK7 and TDK10 with all of the Zn treatments

Table 8. Total dry weight (g/m^2) in 3 rice varieties with 3 Zn treatments in a lowland rice field affected by Fe toxicity, at two months after transplanting.

Variety	Zn application treatment			Mean
	Control	Spray	Dipped	
TDK5	73.83	77.69	73.92	75.15A
TDK7	58.03	60.18	65.65	61.29B
TDK10	62.16	51.29	62.75	58.73B
Mean	64.67	63.05	67.44	
F-test	Variety*	Treatment^{ns}	Variety x Treatment^{ns}	

* Significant at $P < 0.05$; ns = not significant at $P < 0.05$
Different letters after numbers designate significant difference by $LSD_{0.05}$
$LSD_{0.05}$ for comparing between variety means = 11.407

Control = no Zn application, Dipped = dipping roots seedling in 1% w/v $ZnSO_4$ solution and Spray = spraying 0.5% w/v $ZnSO_4$ solution on the top of rice leaves at 30 days after transplanting. The mean in same column followed by different capital letters are significant different, the mean in same row followed by different small letters are significant different at $p \leq 0.05$.

Grain yield

There was no significant interaction between the effect of rice varieties and Zn treatments on grain yield ($P < 0.05$). It was found that the rice varieties were significant different ($P < 0.05$), TDK5 (272.35 g/m^2) with marked differentiation among the rice varieties into TDK5 with the highest grain yield, followed by TDK10 (254 g/m^2). However, TDK10 was not significant differences ($P < 0.05$) with TDK7 (249 g/m^2) and TDK7 was the lowest in this experiment (Table 9).

Based on the result of data analysis showed there were significant differences ($P < 0.05$) between Zn treatments. The root dipping with Zn treatment (dipping roots seedling in 1% Zn sulfate solution 10 g L^{-1}) gave the highest grain yield was the highest (279 g/m^2). Zinc spray treatment (spraying 0.5% $ZnSO_4.7H_2O$ on the top of rice leaves at 30 days after transplanting) gave grain yield (252 g/m^2) that was not significantly different from, control treatment (no added Zn) (245 g/m^2) (Table 9)

Table 9. Grain yield (g/m^2) in 3 rice varieties with 3 Zn treatments in a lowland rice field affected by Fe toxicity.

Variety	Zn application treatment			Mean
	Control	Spray	Dipped	
TDK5	266.82	261.39	288.85	272.35A
TDK7	231.64	250.96	265.37	249.32B
TDK10	235.85	242.80	282.93	253.86AB
Mean	244.77b	251.72b	279.05a	
F-test	Variety*	Treatment*	Variety x Treatment^{ns}	

* Significant at $P < 0.05$; ns = not significant at $P < 0.05$
Different letters after numbers designate significant difference by $LSD_{0.05}$
$LSD_{0.05}$ for comparing between variety means = 17.118
$LSD_{0.05}$ for comparing between Treatment means = 24.624

Control = no Zn application, Dipped = dipping roots seedling in 1% w/v $ZnSO_4$ solution and Spray = spraying 0.5% w/v $ZnSO_4$ solution on the top of rice leaves at 30 days after transplanting. The mean in same column followed by different capital letters are significant different, the mean in same row followed by different small letters are significant different at $p \leq 0.05$.