

## CHAPTER 4

# GENOTYPIC VARIATION IN VEGETATIVE AND REPRODUCTIVE RESPONSES TO LOW BORON IN MAIZE

### 4.1 Introduction

Genotypic variation in responses to low B has been reported in many plants species (see review by Rerkasem and Jamjod, 1997). Physiological impact of low B is also varied. The vegetative response range from inhibition of root growth (Dugger, 1983; Bohnsack and Albert, 1977), poor wood lignification in trees (Dell and Malajczuk, 1994), limited leaf cell expansion (Kirk and Loneragan, 1988; Hu and Brown, 1994), which presumably would result in depressed leaf area, increasing shoot to root ratio as root is more sensitive to B deficiency (Kirk and Loneragan, 1988; Huang *et al.*, 1996). Reproductive growth has been reported affected by B deficiency via development of the flower buds (Rerkasem *et al.*, 1987; Rerkasem, 1991), anther and pollen (Rerkasem *et al.*, 1989; Rerkasem and Loneragan, 1984), the pistil (Xu *et al.*, 1993), pollen germination and the fertilization process (Cheng and Rerkasem, 1993) and seed and fruit (Rerkasem *et al.*, 1987; Rerkasem *et al.*, 1993; Dear and Lipsett, 1987; Keerati-kasikorn *et al.*, 1991). Genotypic variation has been reported only some of these studies, for example in green and black gram (Rerkasem, 1991), peanut (Keerati-kasikorn *et al.*, 1991). In wheat, it has be clearly established that reproductive development, especially formation of the pollen, was most sensitive to B

deficiency (Rerkasem *et al.*, 1993), and that genotypic variation in response to low B in wheat was primarily based on the effect of B on male fertility/sterility (Rerkasem and Jamjod, 1997). In Chapter 3, I found that B deficiency depressed maize grain yield primarily through reproductive development. These earlier experiments also showed maize to differ from wheat in that although pollen development was adversely affected by B deficiency, it was the female flower, *i.e.* the babycorn and silk, that was most sensitive. Understanding how maize genotypes differ in the way in which their vegetative and reproductive development respond to low B will help us not only prevent yield loss in maize grown on low B soils, but should also contribute towards male and female fertility control in hybrid maize seed production. Thus, the objective of this experiment was to determine the response to B and evaluated genotypic variability in response to B deficiency.

#### 4.2 Materials and methods

The experiment was conducted from June to September 2005 at the Multiple Cropping Center, Chiang Mai University, Thailand. The experimental design was a factorial with four replicates x two B levels. Seeds of the seven maize (*Zea mays* L.) genotypes, including: NS1, Pioneer A33, CM, Glutinous Corn, Sweet Corn 7101, White Sweet Corn and NS72 were placed on a moistened paper in aluminum tray until germinated (approximately two days). Germinated seeds were transplanted to pots (0.30 m diameter and 0.30 m deep) filled with washed river quartz sand at 8 seedlings per pot (day 0). During the seven days after transplanting, pots were watered daily with filtered water. Seedlings were thinned to four plants per pot on day 14. Pots were supplied twice daily with complete nutrient solution with two

levels of added B (0 or 20  $\mu\text{M}$  B). The nutrient solution, adapted from Broughton and Dilworth (1971) and Mozafar (1989), consisted of  $\text{KNO}_3$  15000  $\mu\text{M}$ ;  $\text{CaCl}_2$  1000  $\mu\text{M}$ ;  $\text{Mg}(\text{SO}_4)_2 \cdot 7\text{H}_2\text{O}$  2000  $\mu\text{M}$ ;  $\text{KH}_2\text{PO}_4$  1000  $\mu\text{M}$ ; Fe-EDTA 100  $\mu\text{M}$ ;  $\text{K}_2\text{SO}_4$  250  $\mu\text{M}$ ;  $\text{MnSO}_4 \cdot \text{H}_2\text{O}$  9  $\mu\text{M}$ ;  $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$  0.76  $\mu\text{M}$ ;  $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$  0.31  $\mu\text{M}$ ;  $\text{CoSO}_4$  0.1  $\mu\text{M}$  and  $\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$  0.1  $\mu\text{M}$ . All pots were flushed with filtered-water every week to avoid accumulation of salts in the sand.

Plants were harvested during vegetative development at the 5-leaf stage (day 30, Harvest 1), during reproductive growth at anthesis (Harvest 2) and separated into YEB (youngest emerged blade), rest of tops and roots from each pot and dried at 75  $^\circ\text{C}$ . Dry weight were taken B was analysed as described in Chapter 2. Pollen was collected separately at anthesis (tassel, flag leaf and ear leaf) for each pot and applied to the silk of plants in the same pot. At maturity, grain and straw dry weight were determined, yield and yield components were recorded.

#### 4.2 Statistic analysis

The data were analysed statistically by analysis of variance (ANOVA) to test the effect of the factors. Significantly different means were separated at the 0.05 probability level by the least significant difference (LSD). Data of statistical analyses were done by using commercial software (Statistix V. 8, Analytical Software, Inc.).

### 4.3 Results

#### 4.3.2 Vegetative stage

At early vegetative stage (5-leaf stage) the YEB, shoot and root were all slightly lower in dry weight in B20 than in B0 (Table 4.1). On average of the shoot dry weight, genotypes with highest dry weight were found in CM, NS72 and SC by

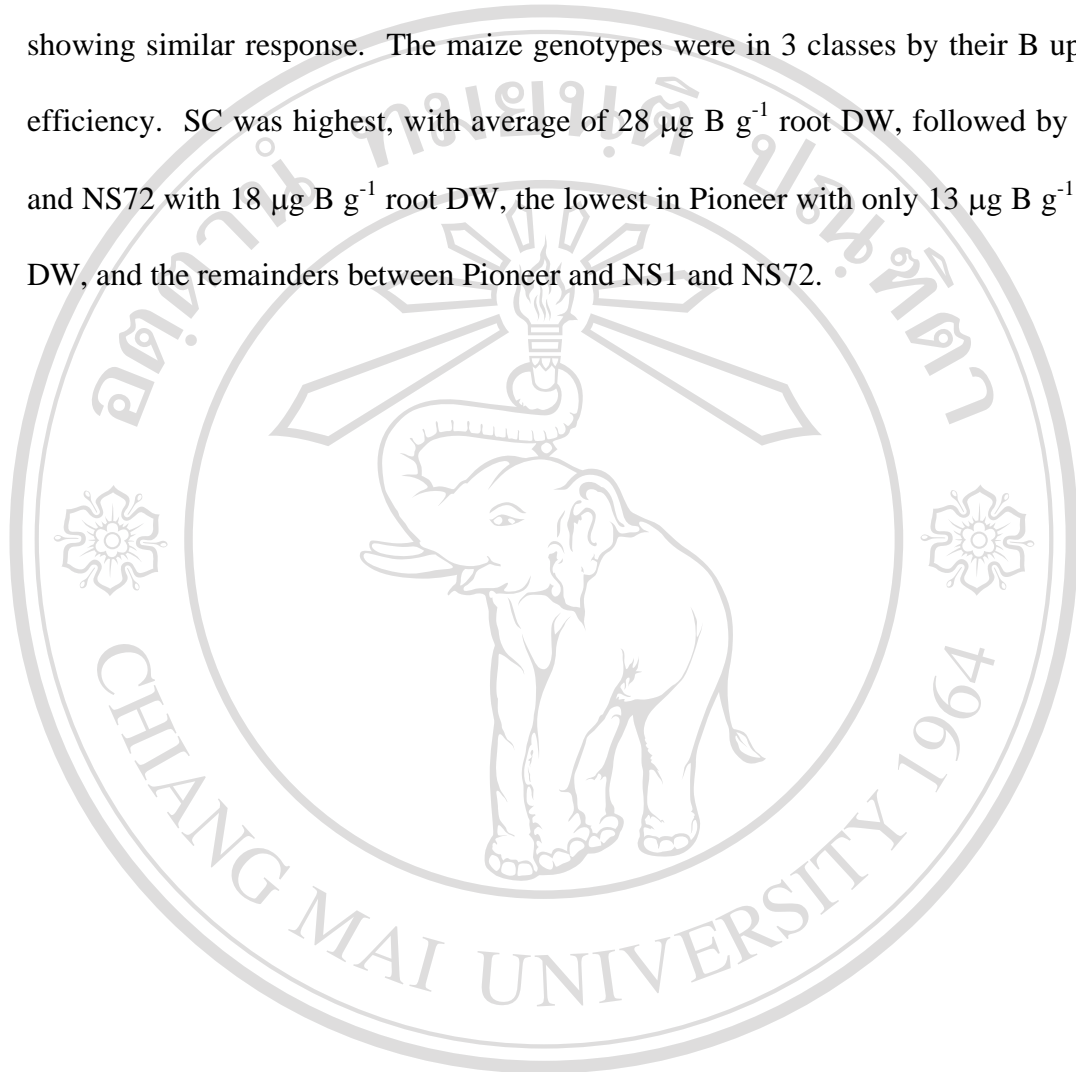
4.3, 4.6 and 4.9 g plant<sup>-1</sup>, respectively whereas the lowest shoot dry weight was 3.3 g plant<sup>-1</sup> in NS1.

B treatment significantly affected the concentration of B in YEB of NS1, GC, CM, SC and WSC but not in Pioneer and NS72 (Figure 4.1). The addition of B significantly increased B concentration in shoot of all genotypes. For example, shoot B concentration increased from about 6 to 9 mg B kg<sup>-1</sup> DW in SC and NS72 increased from about 6 mg B kg<sup>-1</sup> DW to 7 mg B kg<sup>-1</sup> DW. Under B deficiency, genotype did not differ in shoot B of all genotypes in which had about 6 mg B kg<sup>-1</sup> DW. At B20, the highest B concentration in YEB was found in SC was about 9 mg B kg<sup>-1</sup> DW and lowest was found in Pioneer, CM and NS72 was about 6 mg B kg<sup>-1</sup> DW. In case of root, the concentration of B increased with added B in NS1, Pioneer, NS72, SC and WSC but not in CM and GC.

The B content in the YEB was not different between the B treatments or genotypes (Table 4.2). Shoot B content was higher in B20 than B0 in all genotypes, but no difference was observed in root B content. Most outstanding was differences among the genotypes in their shoot and root B contents. SC was different from all the other genotypes in that it had 1/3 to 55% more B in its shoot and less in the roots. Thus SC had the highest shoot to root ratio of its B content at 4.2 compared with only 2.1-2.6 in the other genotypes (Table 4.3). This difference was also reflected in the ratio of shoot to root in dry weight that was much higher in SC, at 3.5 compared with only 1.8 in Pioneer, which was the lowest (Table 4.3).

Differential response to B among the genotypes was observed in plant height, which showed some toxic effect at B20 (Figure 4.2). The adverse effect at B20 was observed in depressed height some of the genotypes, most notably in NS1 and GC.

Plant B uptake efficiency is defined as whole plant B content per unit root dry weight. The B uptake efficiency was clearly high in B20 than in B0, with genotypes showing similar response. The maize genotypes were in 3 classes by their B uptake efficiency. SC was highest, with average of  $28 \mu\text{g B g}^{-1}$  root DW, followed by NS1 and NS72 with  $18 \mu\text{g B g}^{-1}$  root DW, the lowest in Pioneer with only  $13 \mu\text{g B g}^{-1}$  root DW, and the remainders between Pioneer and NS1 and NS72.



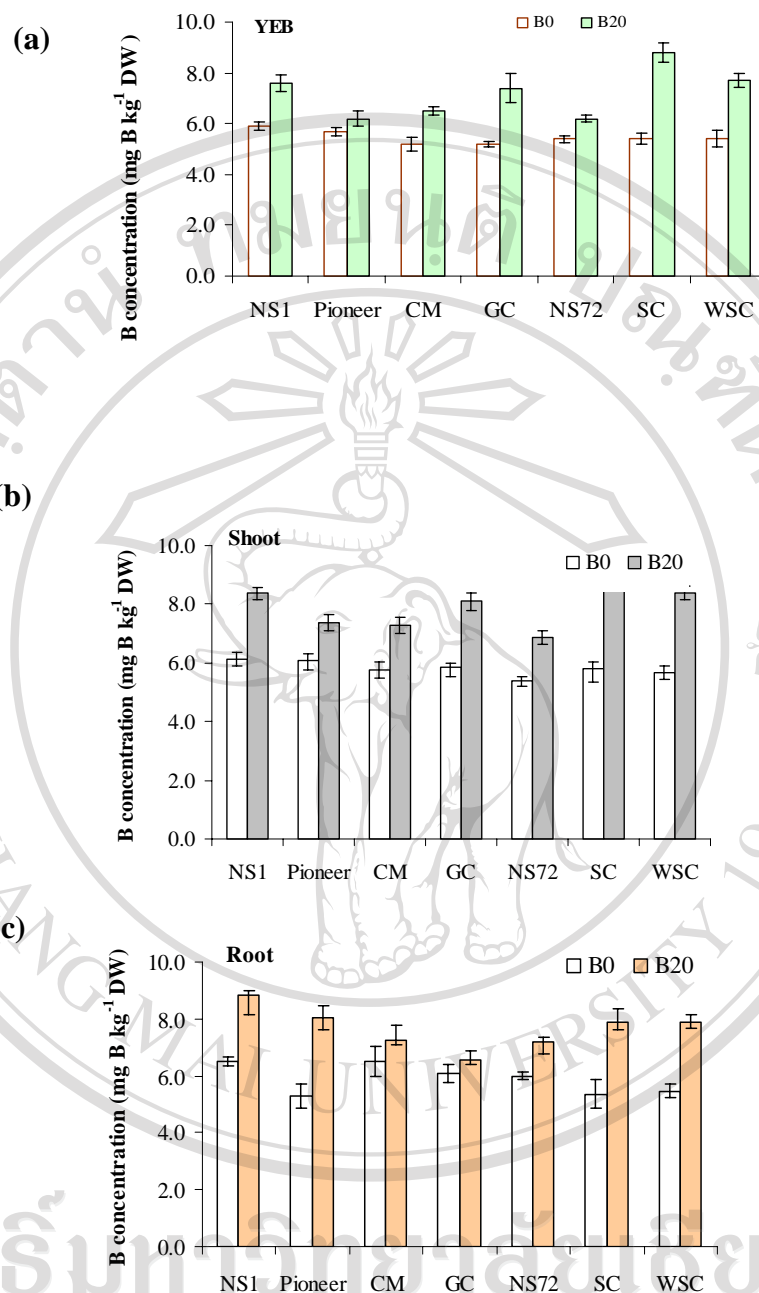
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**Table 4.1** Dry weight (g plant<sup>-1</sup>) of the YEB, total shoot and root of the seven genotypes of maize at two levels of B supply at the 5-leaf stage.

Genotype	Added B (μM)		mean
	B0	B20	
<u>YEB</u>			
NS1	0.55	0.41	0.48
Pioneer	0.60	0.42	0.51
CM	0.63	0.49	0.56
GC	0.54	0.40	0.47
NS72	0.46	0.53	0.50
SC	0.54	0.46	0.50
WSC	0.46	0.40	0.43
mean	0.54	0.44	
F-test	B **	G <sup>NS</sup>	BxG <sup>NS</sup>
LSD <sub>0.05</sub>	0.06	-	-
<u>Total shoot</u>			
NS1	3.72	2.93	3.32C
Pioneer	4.01	3.33	3.67BC
CM	4.82	3.74	4.28AB
GC	4.55	3.03	3.79BC
NS72	4.34	4.76	4.55A
SC	5.14	4.66	4.90A
WSC	4.19	3.32	3.76BC
mean	4.40A	3.68B	
F-test	B <sup>***</sup>	G <sup>***</sup>	BxG <sup>NS</sup>
LSD <sub>0.05</sub>	0.37	0.69	-
<u>Root</u>			
NS1	1.60	1.34	1.47CD
Pioneer	2.30	1.89	2.09A
CM	2.28	1.58	1.93AB
GC	2.09	1.45	1.77ABCD
NS72	1.95	1.66	1.80BC
SC	1.69	1.23	1.46D
WSC	1.73	1.67	1.70BCD
mean	1.95A	1.54B	
F-test	B <sup>***</sup>	G <sup>***</sup>	BxG <sup>NS</sup>
LSD <sub>0.05</sub>	0.18	0.17	-

NS= not significant, \*\* and \*\*\* significant at P<0.01 and 0.001, respectively.

Number followed with the different letters indicated significantly different by LSD ( $P < 0.05$ ).



**Figure 4.1** Boron concentration (mg B kg<sup>-1</sup> DW) in plant parts of seven genotypes of maize at two levels of boron supply at the 5-leaf stage. Bars represent mean of four replicates ( $\pm$ SE).

Analysis of variance

Effects	B	G	BxG
F-test (a) YEB	**	NS	NS
(b) Shoot	***	***	NS
(c) Root	***	***	NS

**Table 4.2** Boron content ( $\mu\text{g plant}^{-1}$ ) in the YEB, total shoot and root of the seven genotypes of maize at two levels of B supply at the 5-leaf stage.

Genotype	Added B ( $\mu\text{M}$ )		mean
	B0	B20	
	<u>YEB</u>		
NS1	3.4	3.1	3.2
Pioneer	3.4	2.6	3.0
CM	3.2	3.1	3.2
GC	2.8	3.0	2.9
NS72	2.7	2.2	3.0
SC	2.8	4.2	3.5
WSC	2.4	3.1	2.8
mean	3.0	3.2	
F-test	B <sup>NS</sup>	G <sup>NS</sup>	BxG <sup>NS</sup>
	<u>Total shoot</u>		
NS1	23.2	26.1	24.7B
Pioneer	24.7	27.4	26.1B
CM	29.6	29.7	29.6B
GC	27.3	26.0	26.7B
NS72	23.9	35.4	29.6B
SC	32.2	44.3	38.3A
WSC	24.1	29.4	26.7B
mean	26.4B	31.2A	
F-test	B <sup>***</sup>	G <sup>***</sup>	BxG <sup>NS</sup>
LSD <sub>0.05</sub>	3.5	6.53	-
	<u>Root</u>		
NS1	10.4	12.1	11.3AB
Pioneer	12.2	15.1	13.7A
CM	14.9	11.6	13.3A
GC	12.7	9.6	11.2AB
NS72	11.6	11.9	11.8AB
SC	8.9	9.5	9.2B
WSC	9.3	13.1	11.2AB
mean	11.5	11.8	
F-test	B <sup>NS</sup>	G <sup>*</sup>	BxG <sup>NS</sup>
LSD <sub>0.05</sub>	-	2.7	-

NS= not significant, \*and \*\*\* significant at  $P < 0.05$  and  $0.001$ , respectively.

The difference between genotypes in the same column is indicated by upper case letters.

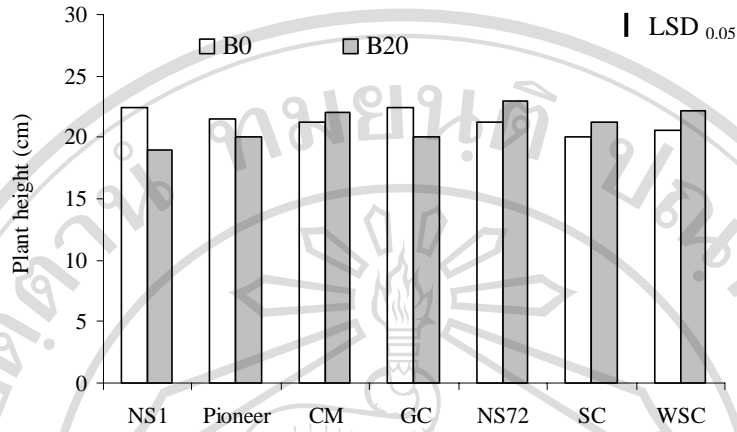


**Table 4.3** The ratio between shoot and root for dry weight and B content of seven genotypes of maize at two levels of B supply at the 5-leaf stage.

Genotype	Added B ( $\mu\text{M}$ )		mean
	B0	B20	
	<u>Shoot DW: root DW ratio</u>		
NS1	2.6	2.4	2.5B
Pioneer	1.9	1.8	1.8C
CM	2.1	2.5	2.3BC
GC	2.3	2.2	2.2BC
NS72	2.4	3.0	2.7B
SC	3.1	2.9	3.5A
WSC	2.4	2.1	2.2BC
mean	2.4	2.5	
F-test	NS	G <sup>***</sup>	BxG <sup>NS</sup>
LSD <sub>0.05</sub>	-	0.6	
	<u>B content shoot: root</u>		
NS1	2.4	2.5	2.5B
Pioneer	2.3	1.9	2.1B
CM	2.1	2.7	2.4B
GC	2.2	2.9	2.6B
NS72	2.2	3.1	2.6B
SC	3.8	4.6	4.2A
WSC	2.6	2.2	2.4B
mean	2.5	2.8	
F-test	B <sup>NS</sup>	G <sup>***</sup>	BxG <sup>NS</sup>
LSD 0.05	-	0.70	-

NS = not significant \* and \*\*\* significant at  $p < 0.05$  and  $0.001$ , respectively.

The difference between genotypes in the same column is indicated by upper case letters.



**Figure 4.2** Plant height (cm) of the seven genotypes of maize at two levels of B supply at the 5-leaf stage. The vertical bars represents the  $LSD_{0.05}$  for G x B interaction.

Analysis of variance

Effects	B	G	BxG
F-test	NS	NS	*

**Table 4.4** Boron uptake ( $\mu\text{g B g}^{-1}$  root DW) of seven genotypes of maize at two levels of B supply at the 5-leaf stage (H1).

Genotype	Added B ( $\mu\text{M}$ )		mean
	B0	B20	
	<u>Whole plant</u>		
NS1	15.7	21.4	18.6B
Pioneer	11.4	14.8	13.1C
CM	13.5	19.3	16.4BC
GC	13.4	18.8	16.1BC
NS72	13.2	22.3	17.7B
SC	19.3	36.3	27.8A
WSC	14.0	17.7	15.8BC
mean	14.3B	21.5A	
F-test	B***	G***	BxG <sup>NS</sup>
LSD <sub>0.05</sub>	2.4	4.6	-

NS = not significant \*\*\* significant at  $P < 0.001$ .

Mean value followed with the different letters indicated significantly different by LSD ( $P < 0.05$ )

### 4.3.2 Anthesis stage

#### 4.3.2.1 Dry weight, boron concentration and the content of B

By anthesis genotypes responded differently to B in dry weight of plant parts, namely tassel, ear leaf, flag leaf, silk, young ear, husk and also root (Figure 4.3 and

Figure 4.4), but this did not show in the B effect on total shoot dry weight (Table 4.5).

The effect of B, however, was consistent on the different plant parts only in some genotypes. Most notable was SC, which exhibited no effect of B on dry weight of all of its parts that were measured. Pioneer showed almost the same lack of response to B, except that it showed a slight effect of B deficiency on dry weight of ear leaf.

Differential effects of B on the male (tassel) and female (silk and young ear) flowers were observed in many of the genotypes. WSC represents those genotypes which showed moderate effect of B deficiency on tassel dry weight but were most markedly

affected by B deficiency on dry weight of silk and young ear. Opposite to this was GC, which showed the biggest depression of tassel dry weight in B0, by only moderate effects on silk and young ear. NS1, on the other hand, had significantly higher dry weight of silk and young ear in B20 than in B0, but the B effect was reverse on its tassel dry weight, which was higher in B0. Differential response to low B in the silk and young ear, which were both parts of the female flower, was found in one genotype. NS72 was among those with the biggest effect of B deficiency on silk dry weight, whereas its young ear dry weight was only moderately depressed in B0 compared with B20.

Increasing B from B0 to B20 depressed dry weight of flag leaf (Figure 4.3, b) and ear leaf (Figure 4.3, c) of NS1 in NS72, but increasing them in some genotypes and having no significant effect in others. Increasing B had the biggest effect in increasing flag leaf and ear leaf dry weight in WSC, moderately in CM and no significant effect in SC, GC and NS72. Among the seven genotypes of maize, B deficiency markedly depressed the dry weight of root only in CM. In NS1 the root dry weight was actually lower in B20 than in B0 (Figure 4.4, c). Dry weight of husk was lower in B20 than B0 in Pioneer and GC and but showing little or no difference between the two B treatments in the other genotypes. The ratio of shoot root dry weight showed no response to B (Table 4.5).

Increasing B increased B concentration in reproductive tissues of all genotypes, but there was a strong interaction ( $P < 0.01$ ) between B and genotypes on their effects on B concentration in different reproductive tissues. Generally, B concentration in various plant parts such as tassel, flag leaf, ear leaf, silk and pollen of all maize genotypes were significantly increased with added B (Figure 4.5 and 4.6;

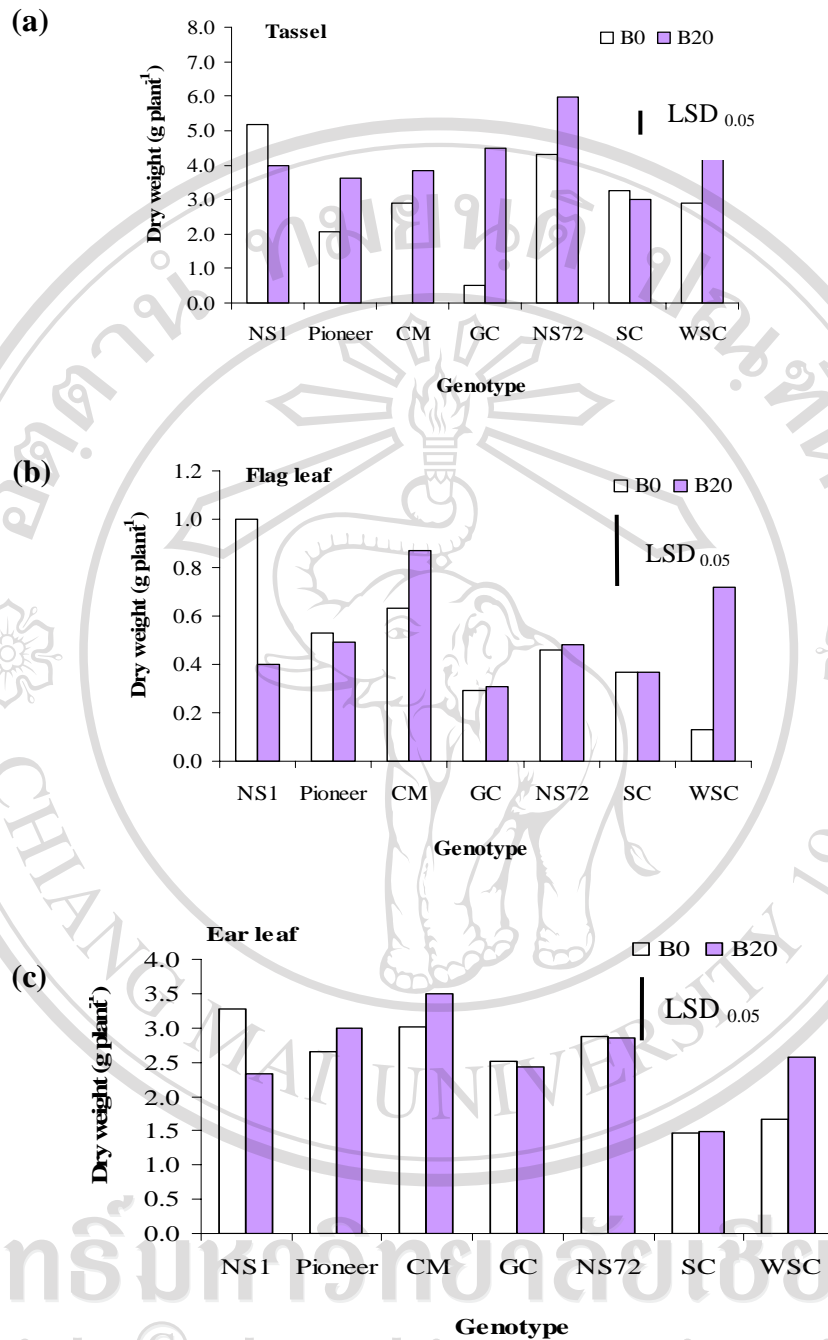
Table 4.6). Some genotypes maintained higher B concentration in B0 in their silk and pollen than others. In B0, SC and WSC had about 7 mg B kg<sup>-1</sup> in their silk whereas others five genotypes had only about 5 mg B kg<sup>-1</sup> or lower (Figure 4.6, a). SC also had highest B concentration in the young ear in B0, but also did Pioneer. B concentration in the pollen in B0 was highest in SC and Pioneer, which were twice as much as that in NS1, NS72 and WSC, which had the lowest pollen B concentration in B0. No significant difference between genotypes was observed in the concentration of B in flag leaf, ear leaf, shoot (Figure 4.7) and roots (Table 4.6). Genotypes differed in their husk B concentration at B0, with CM and SC having the highest B and GC and WSC the lowest B concentration.

There was highly significant interaction between B and genotypes on their effects on the content of B in tassel, ear leaf, husk, young ear and root (Figure 4.8 and 4.9; Table 4.6). Tassel B in B0 was lowest in GC (about 3 µg plant<sup>-1</sup>) whereas SC and NS1 in the same B treatment had about seven times as much B (Figure 4.8, a). Increasing B supply from B0 to B20 increased B content in the tassel of all genotypes, but magnitude of increase was highest in GC. The content of B in ear leaf in B0 did not differ much among the seven genotypes, although increasing B to B20 increased the tassel B content differently, with the biggest increase in CM, NS1 and NS72, and smallest in SC (Figure 4.8, b).

The root B content in B0 was lowest in SC (Figure 4.8, c). The other genotypes all had higher B contents in their roots in B0, especially Pioneer and CM, which had about five times as much root B as SC. Increasing B to B20 had little effect on root B content of SC, NS1 and NS71, but increasing it most markedly (by 60%) and significantly in CM.

The interaction between B and genotype on the content of B in shoot, flag leaf and silk were not significant (Table 4.7). However, there was significant interaction between genotype and B treatment on their effect on total B uptake (shoot+ root). In B0, SC and WSC were genotypes that accumulated less B, which was about half or less of the total B content in the other genotypes. Total plant B content increased with increasing B from B0 to B20. SC was the least responsive, its total B in B20 was only about double that in B0. By contrast, the total B in B20 other genotypes were triple to quadruple that in B0. As the result, SC had the lowest total B content in B20 that was only one third of the other genotypes.

The ratio of B content in shoot: root of most genotypes increased with added B ranging from about 3.6-7.1 to 10.7-22.1 excepted in CM which B treatment did not affect (Table 4.7). Comparisons of B efficiency were made between seven maize genotypes were showed in Figure 4.10. Without B, all genotypes showed ability to uptake B similarly, however, SC had a slightly better root efficiency about  $40 \mu\text{g B g}^{-1}$  root DW whereas the others less than approximately  $24\text{-}25 \mu\text{g B g}^{-1}$  root DW excepted in NS1 took up  $30 \mu\text{g B g}^{-1}$  root DW.

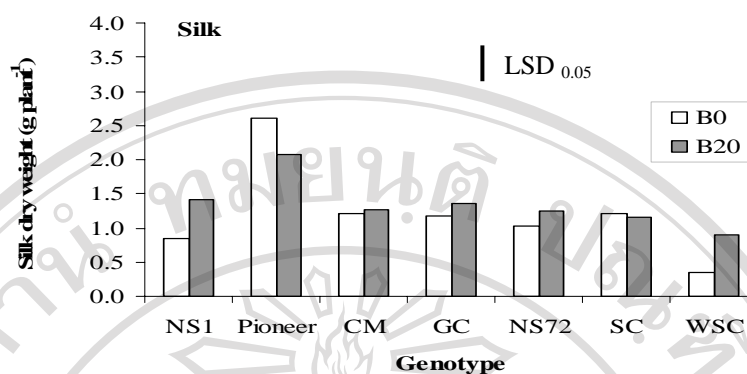


**Figure 4.3** Dry weight (g plant<sup>-1</sup>) in plant parts of seven genotypes of maize at two levels of B supply at anthesis. The vertical bars represents the LSD<sub>0.05</sub> for G x B interaction.

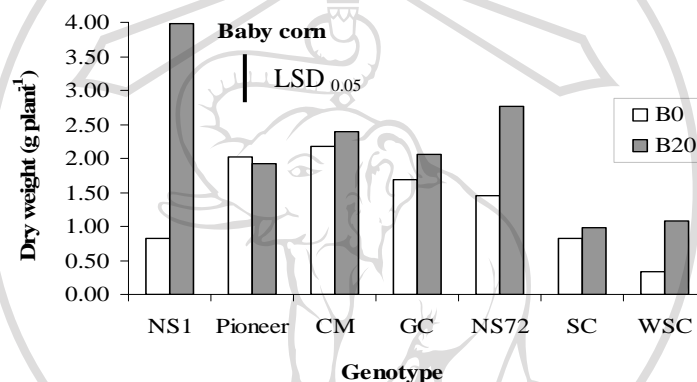
Analysis of variance

Effects	B	G	BxG
F-test (a) Tassel	NS	**	*
(b) Flag leaf	NS	**	**
(c) Ear leaf	NS	***	*

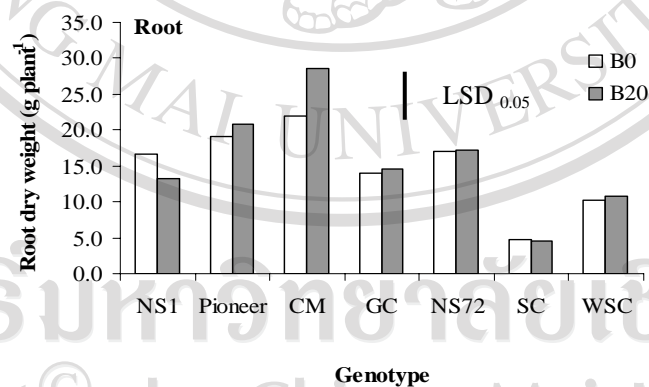
(a)



(b)



(c)



**Figure 4.4** Dry weight ( $\text{g plant}^{-1}$ ) in plant parts of the seven genotypes of maize at two levels of B supply at anthesis. The vertical bars represents the  $\text{LSD}_{0.05}$  for G x B interaction.

#### Analysis of variance

Effects	B	G	BxG
F-test (a) Silk	NS	***	*
(b) Young ear	***	***	***
(c) Root	NS	***	*

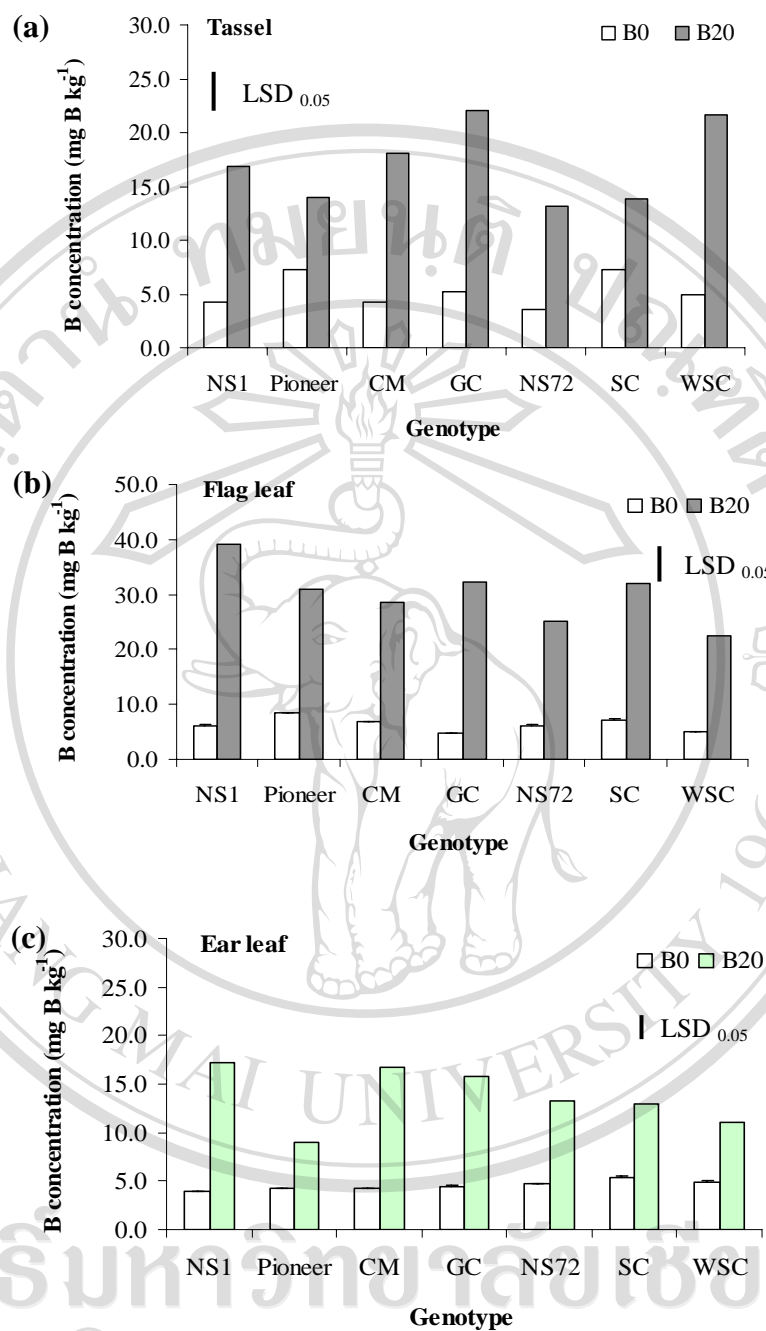


**Table 4.5** Dry weight (g plant<sup>-1</sup>) in total shoot, husk and shoot: root ratio dry weight of seven genotypes of maize at two levels of B supply at anthesis

Genotype	Added B ( $\mu\text{M}$ )		mean
	0	20	
	<u>Total shoot</u>		
NS1	97.1	89.9	93.5B
Pioneer	72.8	83.9	78.4CD
CM	98.0	118.6	108.3A
GC	68.7	76.9	72.8D
NS72	82.6	93.8	88.2BC
SC	22.5	23.0	22.8F
WSC	46.6	66.2	56.4E
mean	69.8B	78.9A	
F-test	B**	G***	BxG <sup>NS</sup>
LSD <sub>0.05</sub>	6.5	12.2	-
	<u>Husk</u>		
NS1	8.0ab	7.6b	7.8
Pioneer	9.6a	6.7bcd	8.2
CM	7.1bcd	7.5bc	7.3
GC	7.5bc	5.7cde	6.6
NS72	8.1ab	8.0ab	8.0
SC	2.6f	2.4f	2.5
WSC	4.6e	5.3de	4.9
mean	6.8	6.2	
F-test	B <sup>NS</sup>	G <sup>NS</sup>	BxG*
LSD <sub>0.05</sub>	-	-	1.82
	<u>Shoot: root ratio</u>		
NS1	6.0	6.8	6.4A
Pioneer	3.8	4.0	3.9E
CM	4.5	4.2	4.4D
GC	4.9	5.3	5.1C
NS72	5.0	5.5	5.2BC
SC	5.1	5.1	5.1C
WSC	4.6	6.4	5.5B
mean	4.8	5.3	
F-test	B <sup>NS</sup>	G***	BxG <sup>NS</sup>
LSD <sub>0.05</sub>	-	0.35	-

NS = not significant \*, \*\* and \*\*\* significant at  $p < 0.05$ ,  $0.01$  and  $0.001$ , respectively.

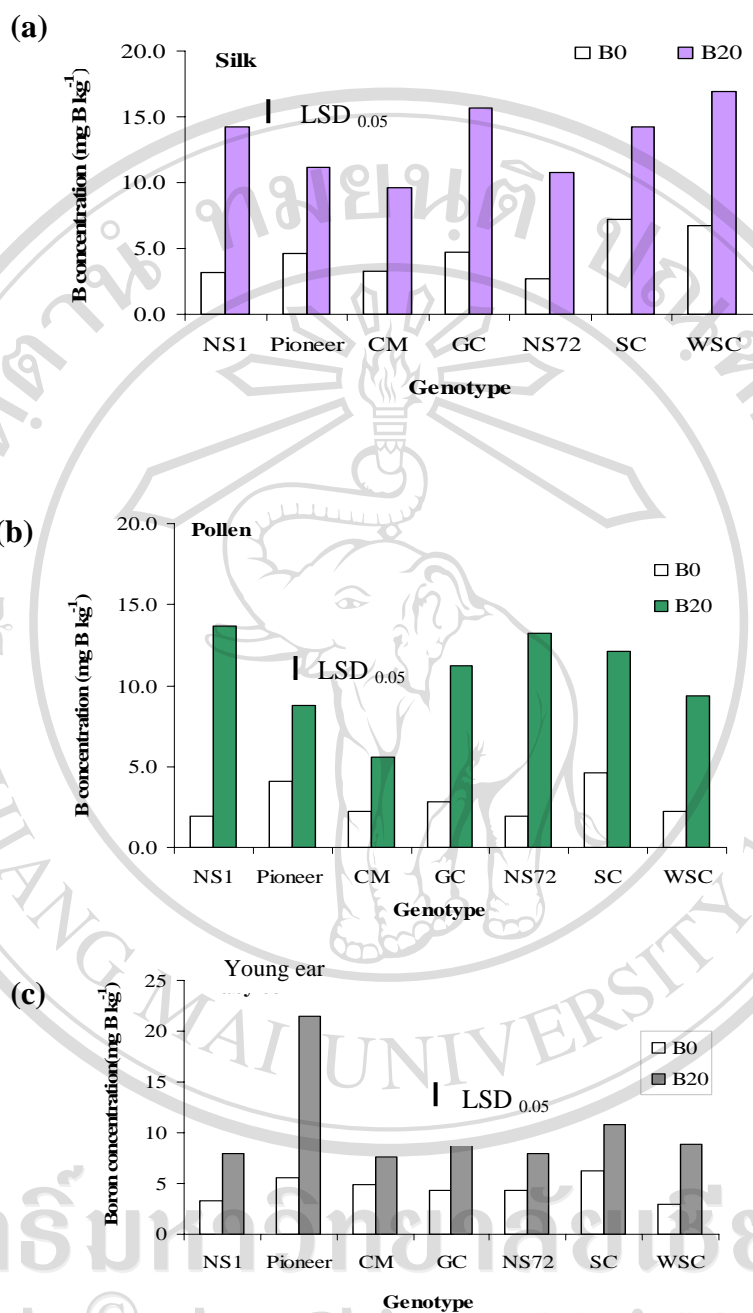
The difference between G in the same column and difference between B in the same row are indicated by upper case letters. The difference between G x B interaction is indicated by lower case letters.



**Figure 4.5** B concentration (mg B kg<sup>-1</sup> DW) in tassel, flag leaf and ear leaf of seven genotypes of maize at two levels of B supply at anthesis. The vertical bars represents the LSD<sub>0.05</sub> for G x B interaction.

Analysis of variance

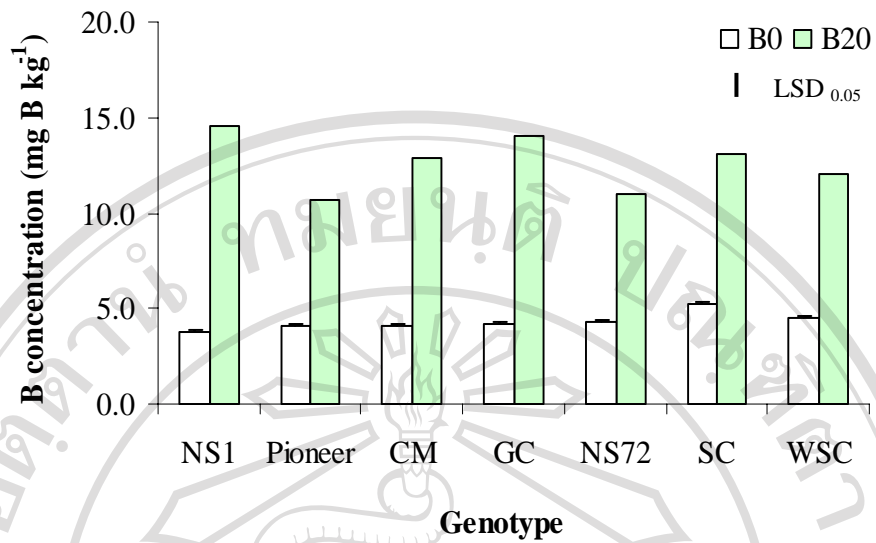
Effects	B	G	BxG
F-test (a) Tassel	***	***	***
(b) Flag leaf	***	***	***
(c) Ear leaf	***	***	***



**Figure 4.6** B concentration ( $\text{mg B kg}^{-1}$  DW) in silks, pollen grains and young ear of seven genotypes of maize at two levels of B supply at anthesis. The vertical bars represents the  $\text{LSD}_{0.05}$  for  $G \times B$  interaction.

Analysis of variance

Effects	B	G	BxG
F-test (a) Silk	***	***	***
(b) Pollen	***	***	***
(c) Young ear	***	***	***



**Figure 4.7** B concentration (mg B kg<sup>-1</sup> DW) in shoot of the seven genotypes of maize at two levels of B supply at anthesis. The vertical bars represents the LSD<sub>0.05</sub> for the G x B interaction.

Analysis of variance

Effects	B	G	BxG
F-test	***	*	**

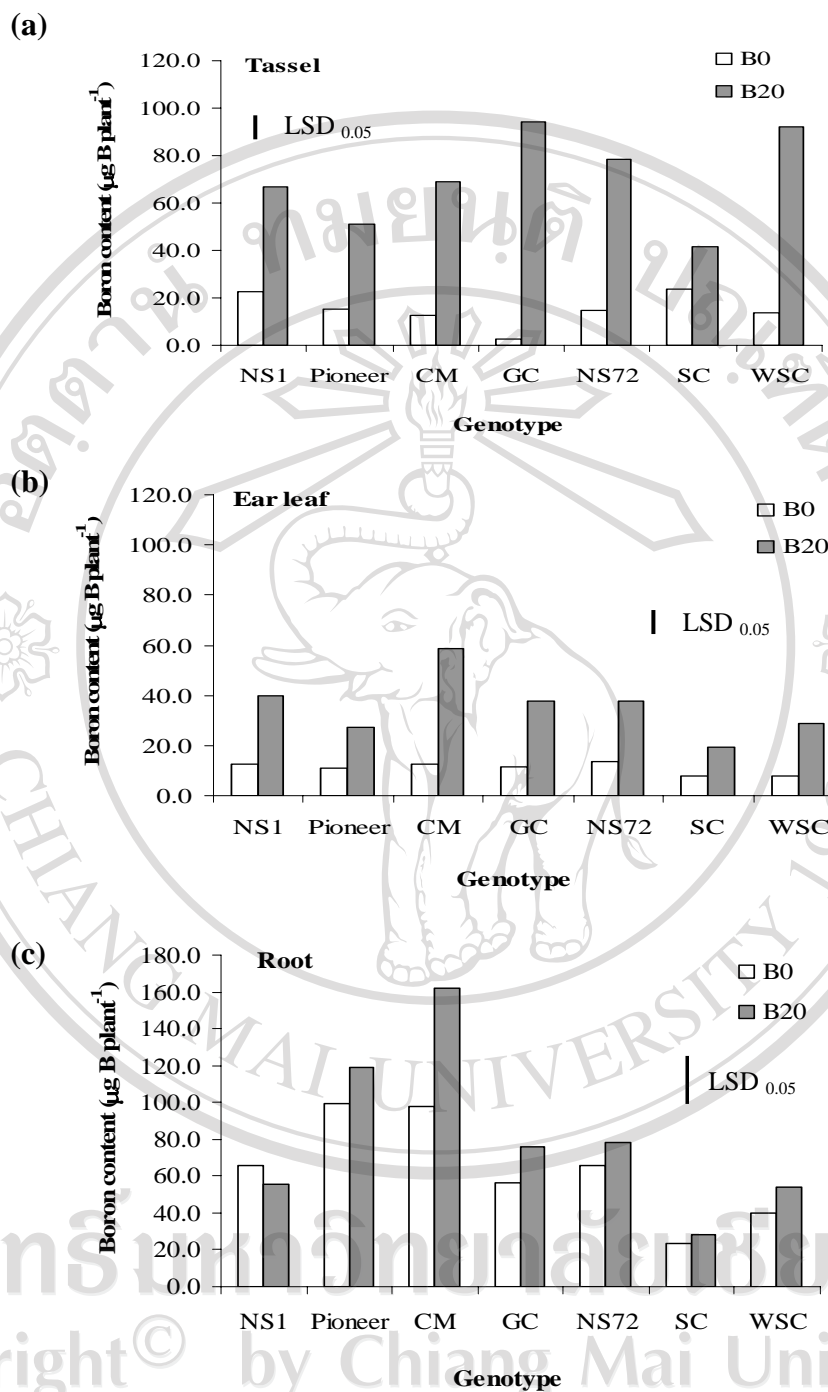
**Table 4.6** B concentration (mg B kg<sup>-1</sup> DW) in young ear and husk of seven genotypes of maize at two levels of B supply at anthesis.

Genotype	Added B (μM)		mean
	0	20	
	<u>Husk</u>		
NS1	3.1gh	5.2ef	4.1
Pioneer	2.8gh	20.8a	11.8
CM	3.9fg	7.5bc	5.7
GC	1.3i	6.9cd	4.1
NS72	2.8ghi	5.5de	4.1
SC	4.8ef	8.8b	6.8
WSC	2.4hi	7.9bc	5.2
mean	3.0B	9.0A	
F-test	B***	G***	BxG***
LSD <sub>0.05</sub>	0.5	1	1.5
	<u>Root</u>		
NS1	4.3	4.3	4.3B
Pioneer	5.2	5.8	5.5A
CM	4.5	5.7	5.1AB
GC	4.0	5.4	4.7AB
NS72	4.0	4.5	4.2B
SC	5.0	6.2	5.6A
WSC	3.9	5.0	4.4B
mean	4.4B	5.2A	
F-test	B***	G*	BxG <sup>NS</sup>
LSD <sub>0.05</sub>	0.5	0.9	-

NS = not significant \* and \*\*\* significant at  $P < 0.05$  and  $0.001$  respectively.

The difference between G in the same column and difference between B in the same row are indicated by upper case letters. The difference between GxB interaction is indicated by lower case letters.

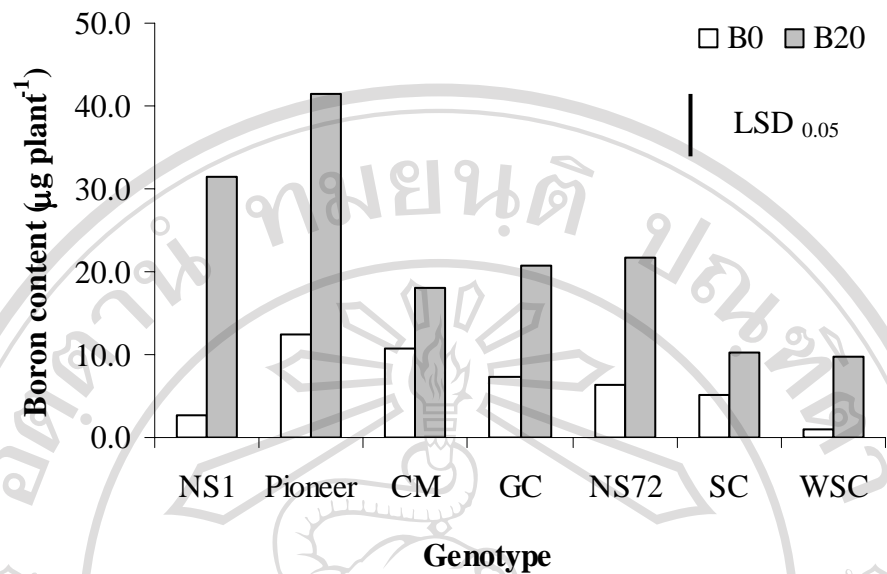
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**Figure 4.8** B content ( $\mu\text{g B plant}^{-1}$ ) in tassel, ear leaf and root of the seven genotypes of maize at two levels of B supply at anthesis. The vertical bars represents the LSD<sub>0.05</sub> for G x B interaction.

Analysis of variance

Effects	B	G	BxG
F-test (a) Tassel	***	***	***
(b) Ear leaf	***	***	***
(c) Root	***	***	**



**Figure 4.9** B content ( $\mu\text{g B plant}^{-1}$ ) in young ear of the seven genotypes of maize at two levels of B supply at anthesis. The vertical bars represents the  $\text{LSD}_{0.05}$  for G x B interaction.

Analysis of variance

Effects	B	G	BxG
F-test	***	***	***

**Table 4.7** Boron content ( $\mu\text{g B plant}^{-1}$ ) in plant parts of the seven genotypes of maize at two levels of B supply at anthesis.

Genotype	Added B ( $\mu\text{M}$ )		mean
	0	20	
<u>Silk</u>			
NS1	2.1	19.5	11.1BC
Pioneer	12.1	23.2	17.6A
CM	4.0	12.2	8.1C
GC	5.7	21.2	13.4B
NS72	2.8	13.4	8.1C
SC	8.8	16.5	12.6B
WSC	2.4	14.8	8.6C
mean	5.5	17.2	
F-test	B <sup>NS</sup>	G <sup>***</sup>	BxG <sup>NS</sup>
LSD <sub>0.05</sub>	-	2.07	-
<u>Flag leaf</u>			
NS1	6.7	15.9	11.3AB
Pioneer	4.3	15.3	9.8BC
CM	4.2	24.6	14.4A
GC	1.5	9.7	5.6C
NS72	2.8	12.3	7.5BC
SC	2.7	11.3	7.0BC
WSC	0.6	16.3	8.5BC
mean	3.3B	15.0A	
F-test	B <sup>***</sup>	G <sup>**</sup>	BxG <sup>NS</sup>
LSD <sub>0.05</sub>	2.4	4.4	-
<u>Husk</u>			
NS1	24.1d	39.4c	31.7
Pioneer	27.2d	139.1a	83.1
CM	27.3d	55.7b	41.5
GC	8.7f	39.5c	24.1
NS72	22.1d	43.5c	32.8
SC	11.7ef	20.7de	16.2
WSC	9.5f	41.8c	25.7
mean	18.7B	54.2A	
F-test	B <sup>***</sup>	G <sup>***</sup>	BxG <sup>***</sup>
LSD <sub>0.05</sub>	3.9	7.3	10.3

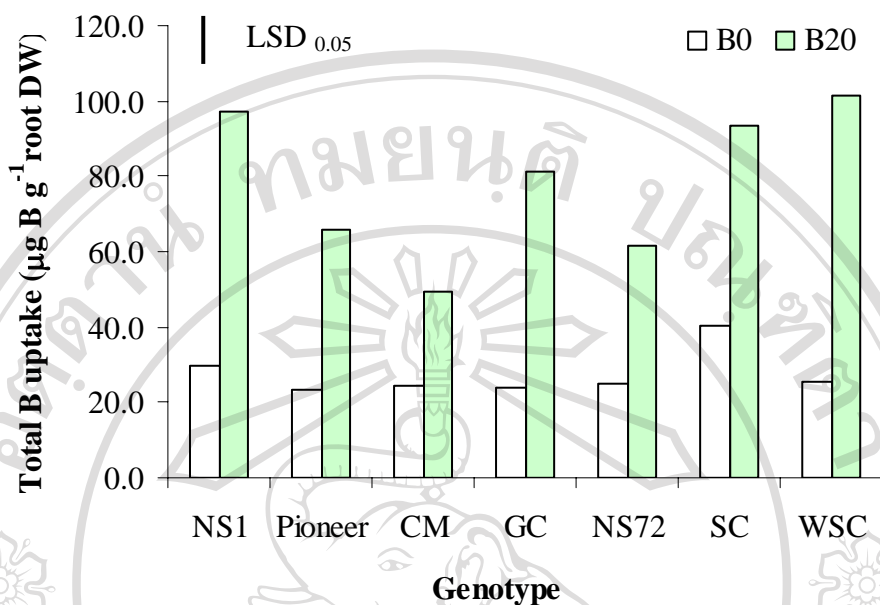


**Table 4.7** (continued).

Genotype	Added B ( $\mu\text{M}$ )		mean
	0	20	
<u>Total shoot</u>			
NS1	411.0	1217.0	814.1A
Pioneer	351.0	1257.0	804.0A
CM	435.6	1250.9	843.3A
GC	282.6	1072.5	677.6B
NS72	352.1	971.5	661.6B
SC	156.5	392.8	274.7C
WSC	213.8	1005.7	609.8B
mean	314.7B	1023.9A	
F-test	B***	G***	BxG <sup>NS</sup>
LSD <sub>0.05</sub>	55.2	103.2	-
<u>Whole plant (shoot+ root)</u>			
NS1	477.1de	1272.8ab	37.5
Pioneer	450.4de	1375.6a	32.7
CM	533.6d	1412.8a	37.8
GC	339.0ef	1148.2bc	29.5
NS72	417.7de	1049.3c	39.8
SC	179.7g	420.9de	42.7
WSC	253.4fg	1059.6c	23.9
mean	378.7B	1105.6A	34.3
F-test	B***	G***	BxG***
LSD <sub>0.05</sub>	58.1	108.8	154.0
<u>Shoot: root ratio</u>			
NS1	6.8de	22.1a	14.4
Pioneer	3.6e	10.7bc	7.1
CM	4.5de	7.9cd	6.2
GC	5.0de	14.2b	9.6
NS72	5.3de	13.1b	9.2
SC	7.1cde	14.3b	10.7
WSC	5.6de	19.4a	12.5
mean	5.4B	14.5A	
F-test	B***	G***	BxG**
LSD <sub>0.05</sub>	1.5	2.7	3.8

NS = not significant, \*\* and \*\*\* significant at  $P < 0.01$  and  $0.001$  respectively.

The difference between G in the same column and difference between B in the same row are indicated by upper case letters. The difference between GxB interaction is indicated by lower case letters.



**Figure 4.10** Total B uptake (shoot+root:  $\mu\text{g B g}^{-1}$  root dry weight) of the seven genotypes of maize at two levels of B supply at anthesis. The vertical bars represents the  $\text{LSD}_{0.05}$  for G x B interaction.

Analysis of variance

Effects	B	G	BxG
F-test	***	***	**

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#### 4.3.2.2 Reproductive development

There was interaction between genotype and B on their effects on in the day of tassel and silk emergence and tassel branching (Table 4.8). Boron deficiency had different effects on dates of tassel and silk emergence in different maize genotypes. B deficiency delayed tassel and silk emergence by 4-5 days in NS72 and Pioneer, hastened these development by a few days in WSC and having no effect in the rest of the genotypes. There was highly significant interaction between B and genotypes was found on tassel branching. WSC was the only genotype that showed effect of B tassel branching, it's tassel had 14 branches in B0 compared with 26 branches in B20. No significant response to B was found in tassel branching in the other six genotypes, which varied greatly in their tassel branching, from 7 branches tassel<sup>-1</sup> in Pioneer, 14 branches tassel<sup>-1</sup> in SC, 17-20 branches tassel<sup>-1</sup> in NS1, NS72 and CM, and the highest number of branches at 27 branches tassel<sup>-1</sup> in GC. B deficiency depressed the number of silk per ear only in NS72, NS1 and WSC, the effect of B on silk number was not significant in the other four genotypes (Figure 4.12). In NS72 the number of silk ear<sup>-1</sup> in B0 was less than one fifth of that in B20, the effect was more moderate in NS1 and WSC. CM, Pioneer and GC already had 400-500 silk ear<sup>-1</sup> in B0. SC generally had fewer silk, at about 300 ear<sup>-1</sup>, which did not respond to B level. The genotypes differed somewhat in the number of leaves and plant height, but no response to B was measured (Figure 4.13).

NS72 showed symptom of B-deficiency as multiple ears as already reported in Chapter 3). Similar symptom of multiple ears were seen in B0 plants of Pioneer, GC and NS1 this experiment (Figure 4.14). In case of SC, the leaf like structure at the tip

of each bract of the husk appeared to be a special characteristic of the genotype that were found in plants at both B treatments (Figure 4.14, a).



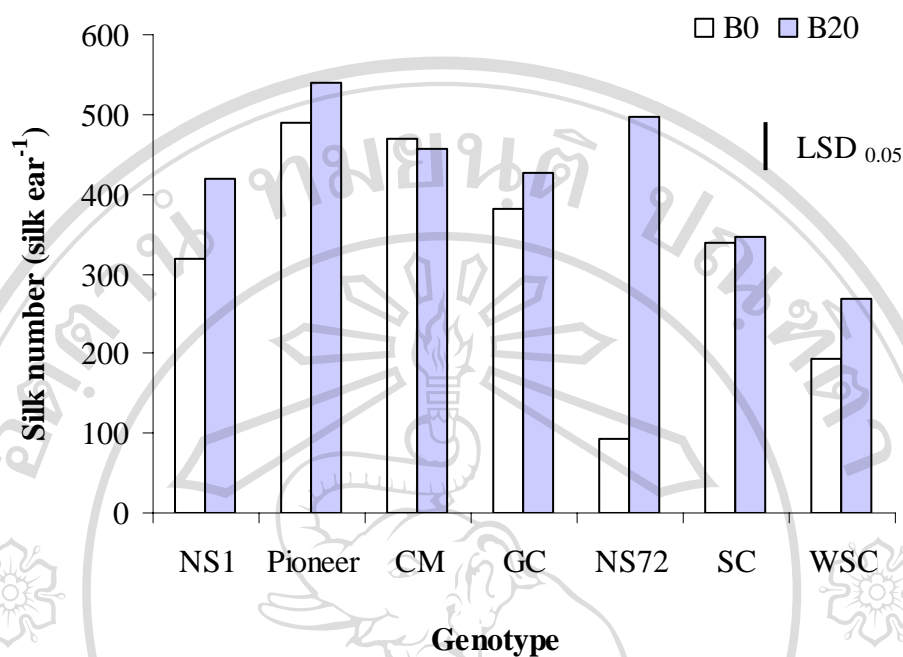
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**Table 4.8** Days of tassel emergence, days of silk emergence and the number branches in tassel of seven genotypes of maize at two levels of B supply at anthesis.

Genotype	Added B ( $\mu\text{M}$ )		mean
	0	20	
<u>Day of tassel emergence</u>			
NS1	53bc	50cd	52
Pioneer	51cd	48e	50
CM	52bc	55ab	54
GC	42e	45e	44
NS72	57a	53bc	55
SC	32f	31f	31
WSC	32f	42e	37
mean	46	46	
F-test	B <sup>NS</sup>	G <sup>***</sup>	BxG <sup>***</sup>
LSD <sub>0.05</sub>	-	2.3	3.3
<u>Day of silk emergence</u>			
NS1	58bc	56c	57
Pioneer	52d	48ef	50
CM	60ab	61a	60
GC	46f	49e	47
NS72	62a	57bc	59
SC	39g	39g	39
WSC	39g	48ef	44
mean	51	51	
F-test	B <sup>NS</sup>	G <sup>***</sup>	BxG <sup>***</sup>
LSD <sub>0.05</sub>		1.8	2.5
<u>Number of branch-tassel plant<sup>-1</sup></u>			
NS1	19bc	20b	20
Pioneer	5e	8e	7
CM	18bcd	16bcd	17
GC	26a	28a	27
NS72	15cd	19bc	17
SC	14d	14d	14
WSC	14d	26a	20
mean	16	19	
F-test	B <sup>***</sup>	G <sup>***</sup>	BxG <sup>**</sup>
LSD <sub>0.05</sub>	1.7	3.1	4.4

NS = not significant \*\*and \*\*\* significant at  $P < 0.01$  and  $0.001$  respectively.

The difference between G in the same column and difference between B in the same row are indicated by upper case letters. The difference between G x B interaction is indicated by lower case letters.

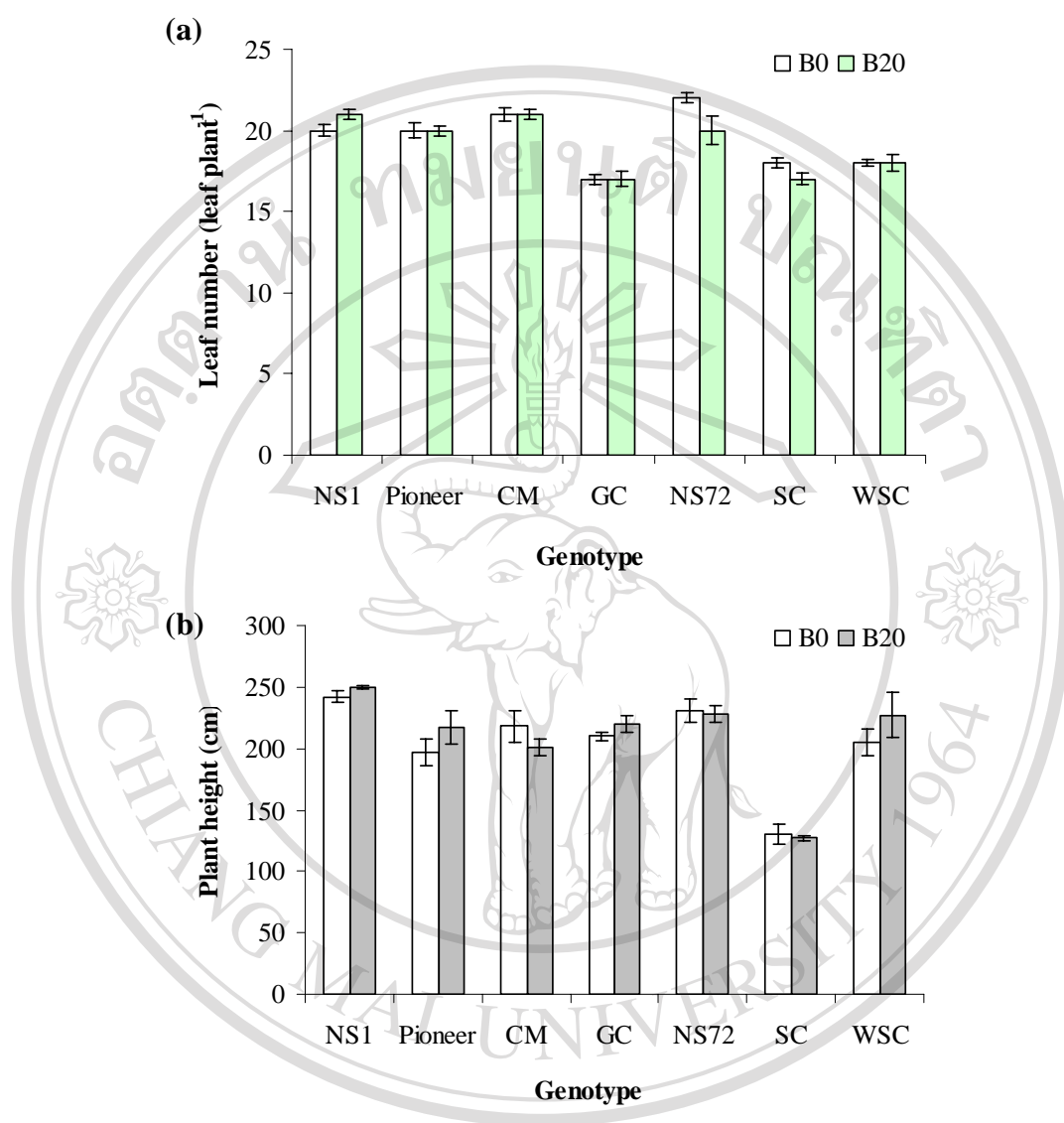


**Figure 4.11** Silk number (silk ear<sup>-1</sup>) of the seven genotypes of maize at two levels of B supply at anthesis.

The vertical bars represents the  $LSD_{0.05}$  for G x B interaction.

Analysis of variance

Effects	B	G	BxG
F-test	***	***	***



**Figure 4.12** Plant height (cm) and leaf number (leaf plant<sup>-1</sup>) of the seven genotypes of maize at two levels of B supply at anthesis. Bars represent mean of four replicates ( $\pm$ SE).

Analysis of variance

Effects	B	G	BxG
F-test (a) Leaf number	NS	***	NS
(b) Plant height	NS	***	NS



**Figure 4.13** Boron deficiency symptom in ear of three maize genotypes as multiple ears: Pioneer (b), GC (c) and NS1 (d). The ear of SC (a) produced more leaves on husk of both the B-deficient and the normal plant.

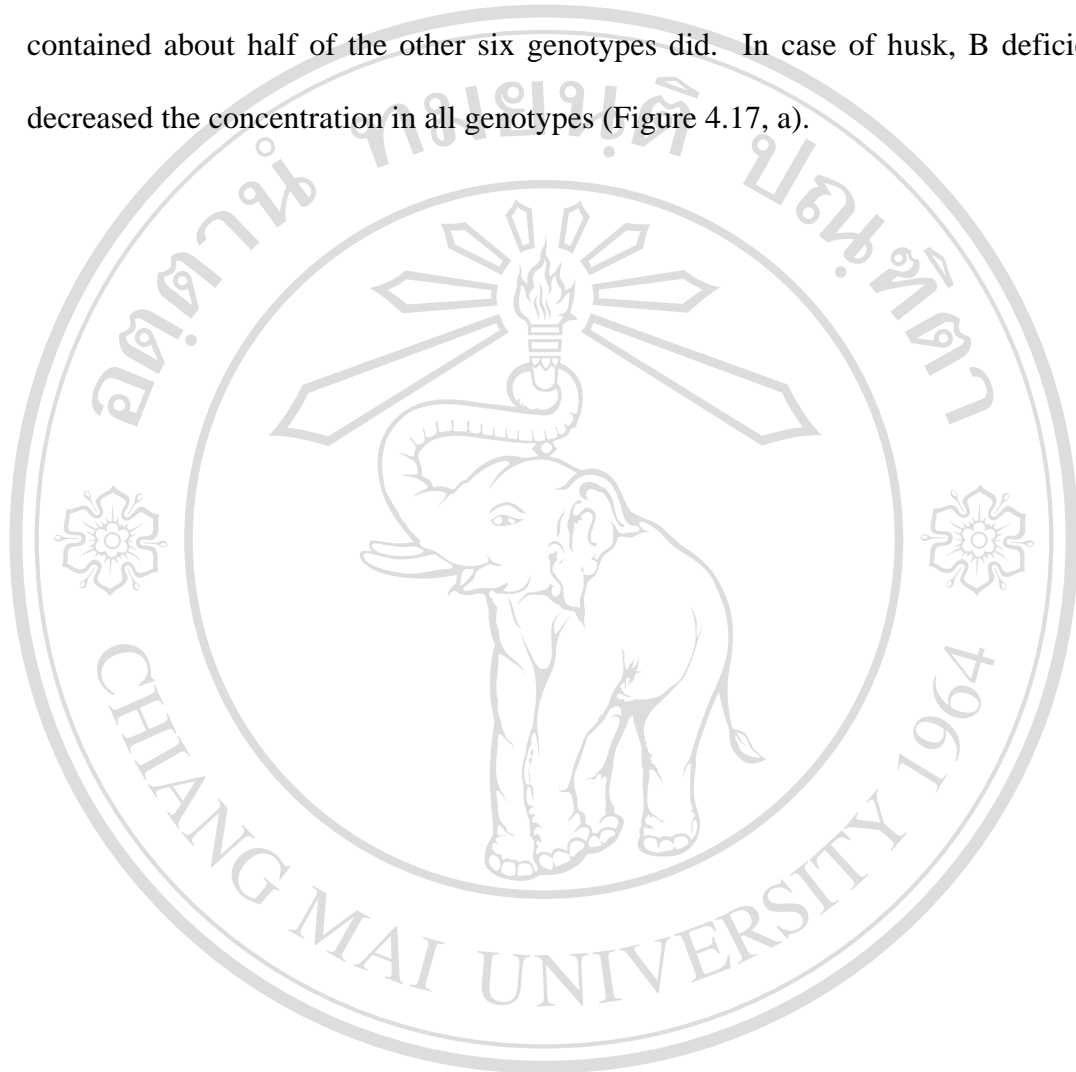


### 4.3.3 Yield and yield components

There were highly significant interaction between the effect of B and genotypes on grain yield and grain number (Table 4, 4.10 and 4.11). The effect of B-deficiency on grain dry weight and the number of grain per ear varied markedly between genotypes. In B0 NS1 and NS72 set almost no grain and CM set a few more, increasing B to B20 increase the number of grain in these genotypes to 400 grains ear<sup>-1</sup> or more. Grain yield of Pioneer and GC were less affected by B deficiency than NS1, NS72 and CM, but their grain yield in B0 were still only 23% and 46% of the yield in B20, with grain number in B0 20% and 50% of those in B20. Most tolerant to B deficiency was SC with grain yield and grain number in B0 almost the same as those in B20, followed by WSC which had grain yield and grain number in B0 about three quarters of those in B20. Boron x genotype interaction effect was not significant in 100 grain weight but varied between genotypes (Table 4.11). High 100 grain weight was found in CM and Pioneer (28-31 g) whereas small size was SC only about 9 g. Similarly, B had no effect on the straw yield, although genotypes differed in the amount of straw they produced. High straw yield was in NS1, CM and NS72 whereas SC was lowest in straw yield (Figure 4.15). For root, there was interaction highly significant interaction between the effect of B and genotypes on dry weight (Figure 4.16). NS1 was the only genotype which saw its root dry weight significant increased from 22 g plant<sup>-1</sup> at B0 to about 30 g plant<sup>-1</sup> in B20. The other six genotypes showed no effect of B on their roots dry weight at maturity.

Boron concentration in straw and root decreased without added B (B0) At B0, there was no significant difference in the concentration of B in straw (Figure 4.17, b). All genotypes had about 5 mg B kg<sup>-1</sup> compared with about 23 mg B kg<sup>-1</sup> at B20. For

root, there were only two (NS1 and WSC) of all genotypes that affected by B that in which B concentration decreased significantly at B0, particular in WSC root contained about half of the other six genotypes did. In case of husk, B deficiency decreased the concentration in all genotypes (Figure 4.17, a).



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**Table 4.9** Grain yield of maize (g ear<sup>-1</sup>) of the seven genotypes of maize at two levels of B supply.

Genotype	Added B ( $\mu\text{M}$ )		Relative yield (%) (B0/B20)
	0	20	
NS1	0.9g	79.5b	1
Pioneer	28.6ed	123.4a	23
CM	4.8fg	97.9b	5
GC	22.3ef	49.0c	46
NS72	0.9g	89.8b	1
SC	17.9efg	1.3ef	84
WSC	31.6cde	42.9cd	74
mean	17.9	72.0	
F-test	B***	G***	BxG***
LSD <sub>0.05</sub>	7.1	13.3	18.8

\*\*\* significant at  $P < 0.001$

The difference between GxB interaction (by LSD  $P < 0.05$ ) is indicated by different letters.

**Table 4.10** Grain number (grain number ear<sup>-1</sup>) of the seven genotypes of maize at two levels of B supply.

Genotypes	Added B ( $\mu\text{M}$ )		Relative grain number
	0	20	% (B0/B20)
NS1	5h	453a	1.1
Pioneer	87fg	426a	20.4
CM	19gh	398b	4.8
GC	115efg	227c	50.7
NS72	4h	433a	0.9
SC	215cd	222c	96.8
WSC	142def	187cde	75.9
mean	84	335	
F-test	B <sup>***</sup>	G <sup>**</sup>	BxG <sup>***</sup>
LSD <sub>0.05</sub>	28	52	74

\*\* and \*\*\* significant at  $P < 0.01$  and  $0.001$  respectively.

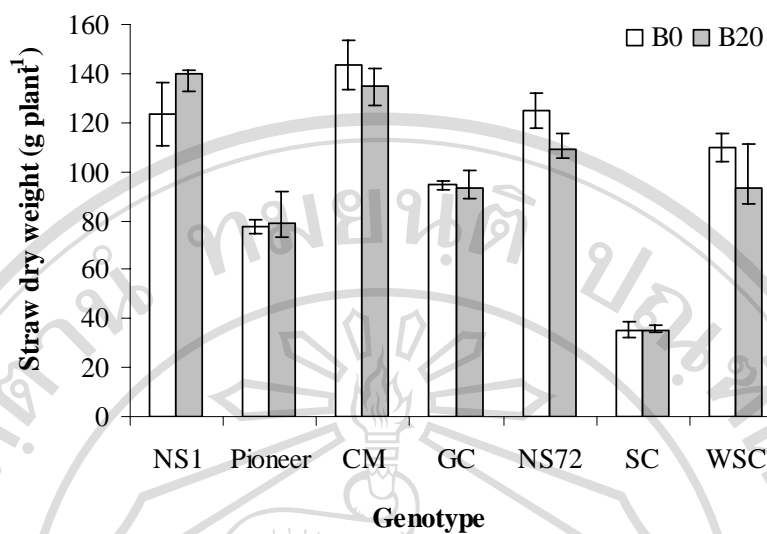
The difference between GxB interaction (by LSD  $P < 0.05$ ) is indicated by different letters.

**Table 4.11** Dry weight of 100 grains (g) of the seven genotypes of maize at two levels of B supply.

Genotypes	Added B ( $\mu\text{M}$ )		mean
	0	20	
NS1	4.8	18.0	18.7DE
Pioneer	31.0	30.2	30.6A
CM	27.1	29.1	28.1A
GC	22.0	23.1	22.6B
NS72	11.0	21.0	21.5CD
SC	8.7	9.2	9.0E
WSC	19.4	22.7	21.1BC
mean	17.7B	21.9A	
F-test	B**	G***	BxG <sup>NS</sup>
LSD <sub>0.05</sub>	2.93	5.5	-

NS = not significant, \*\* and \*\*\* significant at  $P < 0.01$  and  $0.001$  respectively.

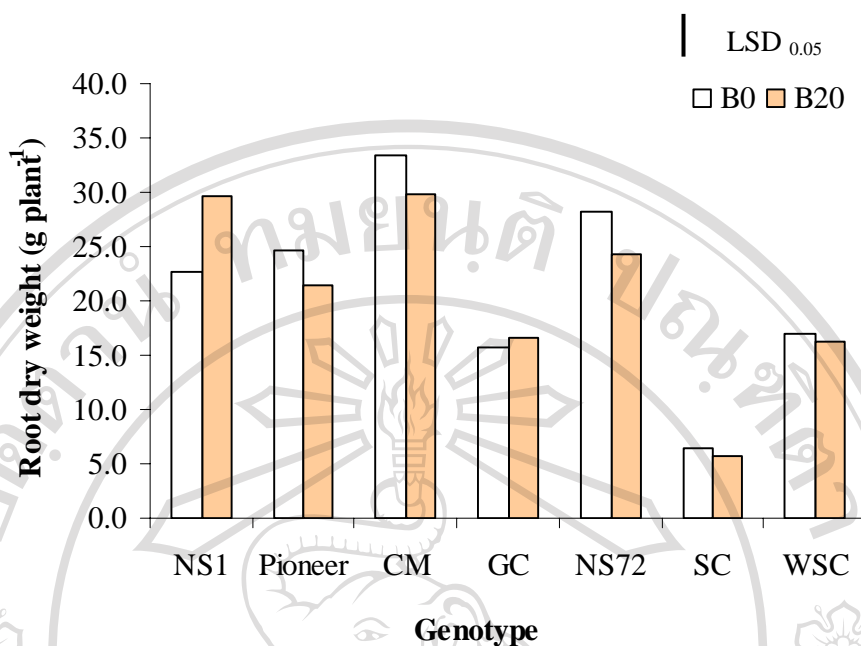
Mean value followed with the different letters indicated significantly different by LSD ( $P < 0.05$ ).



**Figure 4.14** Straw dry weight ( $\text{g plant}^{-1}$ ) of the seven genotypes of maize at two levels of B supply at anthesis. Bars represent means of four replicates ( $\pm\text{SE}$ ).

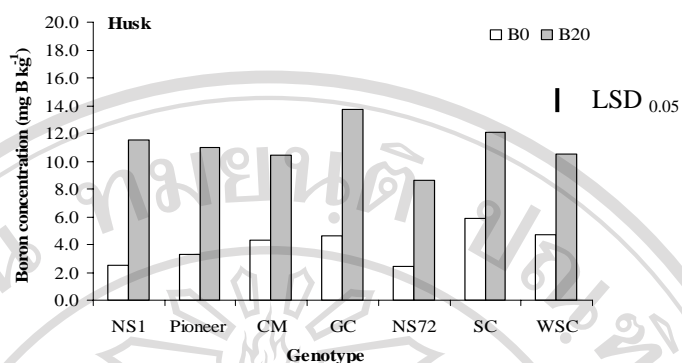
Analysis of variance

Effects	B	G	BxG
F-test	NS	***	NS

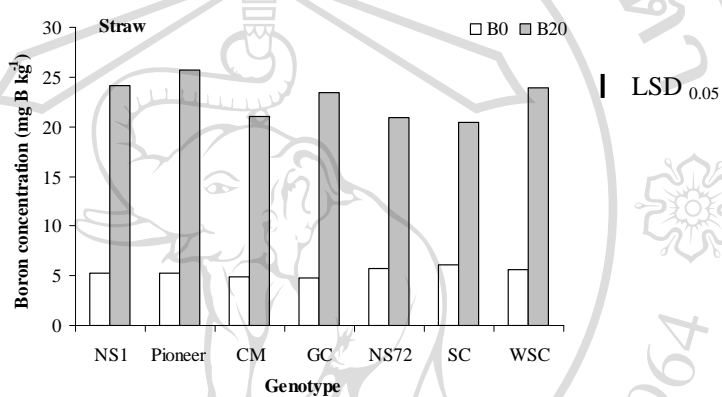


**Figure 4.15** Root dry weight (g plant<sup>-1</sup>) of the seven genotypes of maize at two levels of B supply at anthesis. The vertical bars represents the LSD<sub>0.05</sub> for G x B interaction.

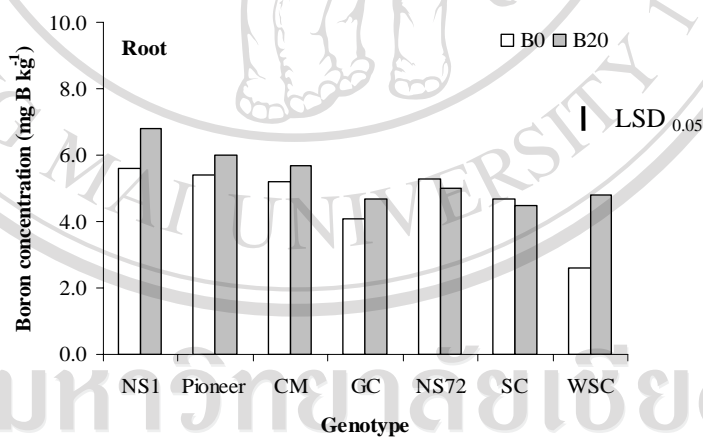
(a)



(b)



(c)



**Figure 4.16** Boron concentration ( $\text{mg B kg}^{-1}$ ) in husk, straw and root of the seven genotypes of maize at two levels of B supply at anthesis.

The vertical bars represents the  $\text{LSD}_{0.05}$  for G x B interaction.

Analysis of variance

Effects	B	G	BxG
F-test (a) Husk	***	***	*
(b) Straw	***	*	**
(c) Root	**	***	*



#### 4.3.4 Relating grain yield with tissue B concentrations

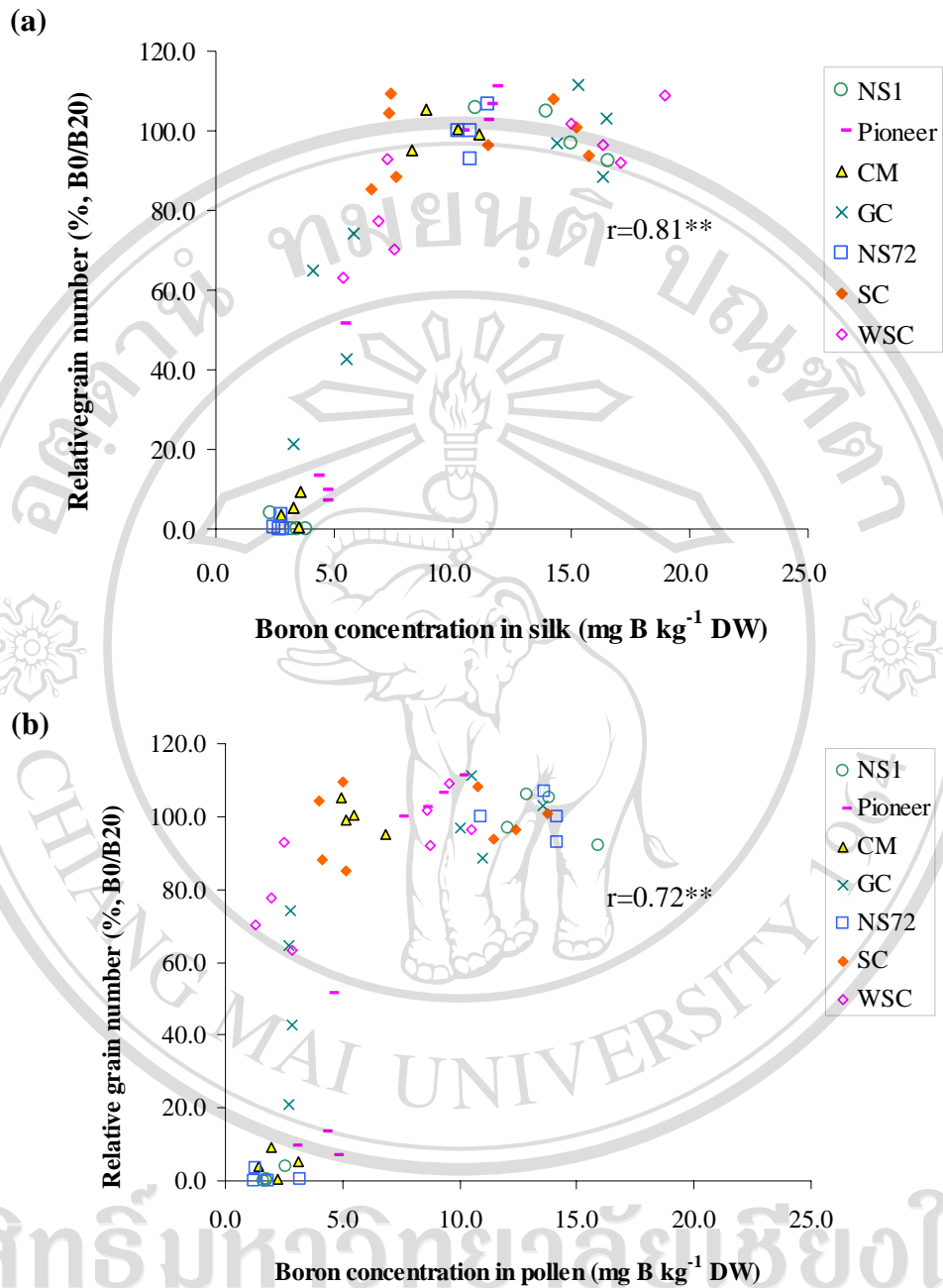
Relative grain yield (defined as grain yield in B0 relative to B20, %) was closely correlated between silk B ( $r=0.84^{**}$ ; Figure 4.18). Similar relationship was found in relative grain number (number of grain in B0/number of grain in B20) and silk B (Figure 4.19).

Moreover, in relative yield that defined as being 100% times the grain yield of B0 which provided adequate amount of all nutrient (without added B) divided by the yield of a treatment which is the same but added B (B20) showed that the relative grain yield of all genotypes were closely correlated between silk B and grain number ( $r = 0.84^{**}$ ; Figure 4.18 and 4.19).

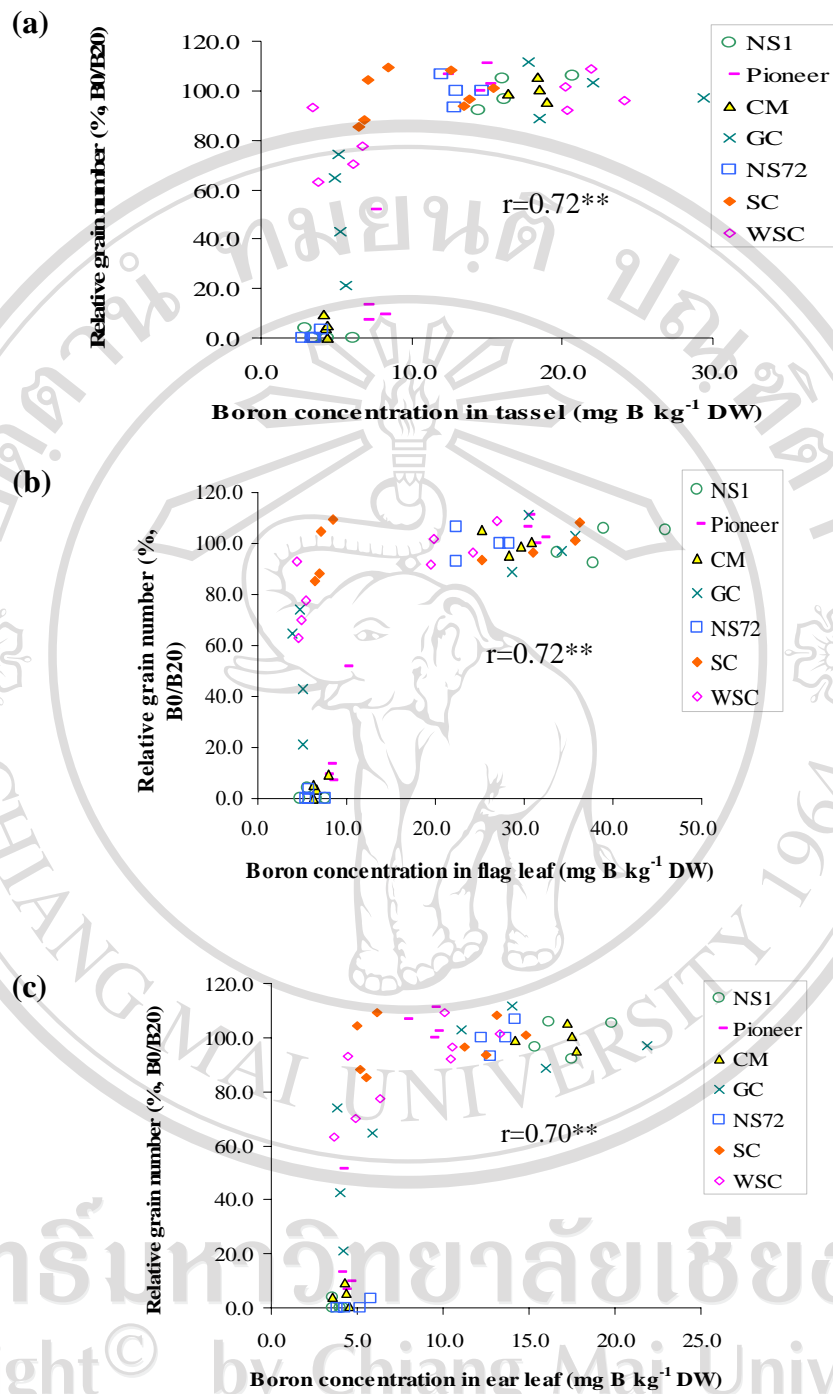
**Table 4.12** Correlation coefficients ( $r$ ) between grain yield and boron concentration ( $\text{mg B kg}^{-1}$  DW) in reproductive tissue.

Yield or yield components	Tassel B	Silk B	Pollen B	Flag leaf B	Ear leaf B
Relative grain no (%)	0.77**	0.81**	0.72**	0.72**	0.70**
Relative grain DW (%)	0.81**	0.87**	0.76**	0.79**	0.79**
100 Grain weight (g)	0.32	0.14	0.08	0.14	0.18
Grain number ear <sup>-1</sup>	0.73*	0.64*	0.73**	0.79**	0.74**
Grain DW ( $\text{g plant}^{-1}$ )	0.62*	0.54*	0.60*	0.62*	0.66*

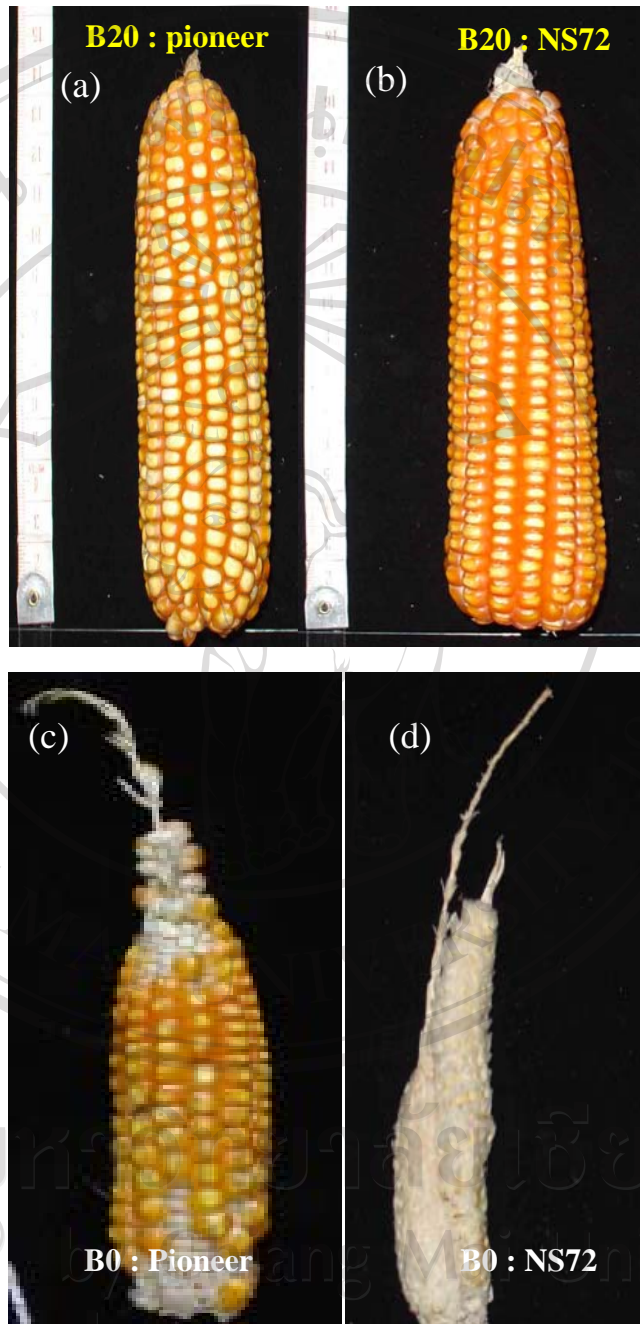
\* and \*\* significant at  $P < 0.05$  and  $0.01$  respectively.



**Figure 4.17** The relationship between B concentration (mg B kg<sup>-1</sup> DW) in silk and pollen and relative grain yield (% B0/B20 of grain dry weight) of all seven genotypes of maize at two levels of B supply at anthesis.



**Figure 4.18** The relationship between B concentration (mg B kg<sup>-1</sup> DW) in tassel, flag leaf and ear leaf and grain number (grain number ear<sup>-1</sup>) of seven genotypes of maize at two levels of B supply at anthesis.



**Figure 4.19** B deficiency symptom in ear of maize (c: Pioneer, d: NS72) and normal ear (a: Pioneer; b: NS72).

#### 4.5 Discussion

The interaction between B and genotypes was not affected on vegetative growth (dry weight of shoot), but varied between genotypes. SC had higher proportion shoot and root in dry weight and the content of B than other genotypes (NS1, Pioneer, CM, GC, NS72 and WSC). B treatment was affected on the concentration of B in various plant parts such as YEB in NS1, GC, CM, SC and WSC but not in Pioneer and NS72 (Table 4.2). However, the addition of B (B20) was significantly increased B concentration in shoot of all genotypes, but was no differed at B0.

Highly significant genotype x boron interaction was observed for grain yield. Considerable genotypic variation in B efficiency was base on relative grain yield (B0/B20). These results indicated that most efficiency genotypes were SC and WSC exceeded 70% relative grain yield. Moderate efficiency was GC had 46% whereas inefficiency genotypes were NS1 and NS72. These results can be ranked from SC and WSC > GC > pioneer > NS1 and NS72. The higher grain yield as SC genotype (B0/B20, 84%) was associated with higher the concentration of B in silk ( $7 \text{ mg B kg}^{-1}$ ) and pollen ( $5 \text{ mg B kg}^{-1}$ ) and also higher the number of grain per ear. For WSC was similar reacted of SC, but the concentration of B in pollen and relative grain yield was lower by  $2 \text{ mg kg}^{-1}$  and 74%, respectively. The others three of the most susceptible to low boron were NS1, GC and NS72 associated with low B concentration in both the silk was about  $3 \text{ mg B kg}^{-1}$  and the pollen was  $2 \text{ mg B kg}^{-1}$ , particularly in B-deficient NS72 was also markedly reduced the number of silk per ear. This concentration of B was approximately  $3 \text{ mg B kg}^{-1}$  of plant tissues that

agreement with results reported by Vaughan (1977) that of this minimum concentration necessary for growth and normal function in all parts. Under B deficiency, the four of all maize genotype produced more ears at the first node included NS72, Pioneer, GC and N1, however NS72 produced higher ear.

**Table 4.13** Effects of B on days to tassel emergence, days to silk emergence, the number of branches-tassel plant per plant, leaf number and plant height (cm) of the seven genotypes of maize at two levels of B supply at anthesis.

Parameters	B effects on parameters
1. Days to tassel emergence	NS72, Pioneer, WSC
2. Days to silk emergence	Pioneer, GC, NS72, WSC
3. Number of branches-tassel per plant	WSC
4. Silk number (silk ear <sup>-1</sup> )	Pioneer, NS72, WSC
5. Total leaf (leaf plant <sup>-1</sup> )	non
6. Plant height	non

These data indicated that genotypes differences in grain yield production allocated to the male (pollen) and the female (silk) development. Grain yield was closely correlation ( $r= 0.64^{**}$ ) between grain number and silk B. Moreover, these evidences showed that SC and WSC were able to produced grain yield at low boron in which to lower requirement at the functional sites. Two sweet corn genotypes (SC and WSC) had higher silk B than others. It may be related with sugar alcohol (B-polyol complex) that majority role of B in transportation and redistribution in some higher plant (Raven, 1980; Brown and Hu, 1996; Hu *et al.*, 1997). Generally, the results indicated that differences among genotypes in during reproductive

development (Table 4.14), by evaluation in days to tassel and silk emergence, number of branches-tassel plant<sup>-1</sup> and silk number. Only two genotypes of all were not affected by B treatment included CM and SC whereas WSC was affected by B in all of observed parameters.

In summary, a genotypic variation in vegetative and reproductive responses to low B, particularly the relative grain yield (both dry weight and grain number) was closely related between silk B. Since the concentration of B in flag leaf and ear leaf were not difference between genotypes under B deficiency, while difference in silk B among genotypes appeared to be greatly influenced through genetic effect at B<sub>0</sub>, thus, the concentration in silk was more associated with grain set reduction than other tissues. It is likely that B efficiency in some maize genotypes may available in this experiment (cv. SC and WSC). Further work, should be concentrated on translocation and distribution of B between genotype such as sweet corn that more important information can explain mechanism of B that may be associated efficient genotype. Moreover, consideration of B efficiency should be useful in maize breeding for selection to grow in low B soils.