



COMBINATIONS OF MULTIPLE GENES CONTROLLING ENDOSPERM CHARACTERS IN RELATION TO MAXIMUM EATING QUALITY OF VEGETABLE WAXY CORN

S. SIMLA¹, K. LERTRAT^{2,3} and B. SURIHARN^{2,3*}

¹Department of Agricultural Technology, Faculty of Technology, Mahasarakham University, Maha Sarakham 44150, Thailand

²Department of Plant Science and Agricultural Resources, Faculty of Agriculture, Khon Kaen University, Khon Kaen 40002, Thailand

³Plant Breeding Research Center for Sustainable Agriculture, Khon Kaen University, Khon Kaen 40002, Thailand

*Corresponding author's email: bsuriharn@gmail.com

Co-authors' email addresses: sakunkan.s@msu.ac.th, kamol9@gmail.com

SUMMARY

Incorporation of sweetness into waxy background is a means to diversify waxy corn products. The best gene combinations and the consumer preference have not been well documented for waxy corn. The objectives of this study were to incorporate sweetness into waxy background and identify gene combinations that are suitable for commercial production using sensory blind test of evaluation panel. The genes *su*, *sh₂* and *bt* were incorporated into waxy background as both a single gene and combination of 2 sweet genes. The segregation ratio of one sweet gene would be 3:1 of waxy kernels to sweet kernels, and the segregation ratio of two-gene combination would be 9:7 of waxy kernels to sweet kernels. Four of 6 crosses with 2 gene combinations were sweeter than all crosses with single gene combination. Two crosses with 2 gene combinations and 101*bt* as a female parent were not sweeter than those with one sweet gene. The parents with *su* and *sh₂* can be used as male and female parents, but the parent with *bt* (101*bt*) can be used as male parent only. Waxy trait was negatively associated with sweetness and crispiness, whereas sweetness was positively associated with crispiness and overall liking, indicating that increases sweetness in waxy corn can increase consumer preference. Sensory blind test can be used for evaluation of eating quality in waxy corn. Implication for breeding is discussed.

Key words: Overall preference, sweetness, tenderness, crispiness and gene combination

Key findings: The parents with *su* and *sh₂* can be used as male and female parents, but the parent with *bt* (101*bt*) can be used as male parent only. Waxy trait was negatively related with sweetness and crispiness but sweetness was positively associated with crispiness and overall liking.

Manuscript received: December 24, 2015; Decision on manuscript: April 5, 2016; Manuscript accepted: May 17, 2016.

© Society for the Advancement of Breeding Research in Asia and Oceania (SABRAO) 2016

Communicating Editor: Bertrand Collard

INTRODUCTION

Immature waxy corn (*Zea mays* var. *ceratina*) is consumed as vegetable in Asia such as in Thailand, China, Vietnam, Laos, Korea and Taiwan. Thailand has exported waxy corn hybrid seeds and frozen waxy corn. It has a promising trend for market expansion for both local and international market. Incorporation of genes controlling sweetness into waxy

background is a strategy to improve waxy corn quality and diversify waxy corn products. The attempt to incorporate sweetness into waxy corn in single kernels has not been succeeded, because of the epistatic effect of genes controlling sweetness over stickiness (Creech, 1965). However, it is possible to incorporate sweetness in one ear of waxy corn by segregation of genes in F₂ seeds.

In most breeding practices, 1 or 2 sweet genes have been used and the sweetness is much lower than normal sweet corn. By incorporating *su*, *sh₂* and *bt* genes into waxy background, it is possible to produce waxy corn hybrids with increased sweetness and other eating qualities such as creaminess texture in one ear (Lertrat and Thongnarin, 2008). Which gene combinations are most suitable is not well understood in waxy corn and breeding strategies to achieve maximum eating quality of waxy corn have not also been well documented.

The breeding program aiming to incorporate these genes into waxy background is undergoing at Khon Kaen University (Thongnarin *et al.*, 2005; Lertrat and Thongnarin, 2008; Thongnarin *et al.*, 2008). However, most previous study has been related to 1 or 2 sweet genes with simple inheritance. Genetic variation for sugar content in sweet corn has been reported. Corn genotypes with common gene combination for sweetness also differed in sugar content (Holder *et al.*, 1974; Ferguson *et al.*, 1979).

When multiple genes are involved, we raise the assumption that multiple genes controlling endosperm characters are related to eating quality of vegetable waxy corn. Thus, the objectives of this study were to incorporate sweetness into waxy background using different combinations of genes controlling

sweetness and to identify the gene combinations that are suitable for commercial production using sensory blind test of evaluation panel. The information obtained will be useful for planning suitable strategies for improving eating quality in waxy corn.

MATERIALS AND METHODS

Plant materials and field management

Five inbred lines of vegetable waxy corn with common waxy background but differing in genes controlling endosperm characters were crossed in all combinations to generate 20 F₁ hybrids (Table 1), and the resulting F₁ hybrids, their parents and 5 check varieties were evaluated for eating quality. The parental genotypes and phenotypes were shown in Figure 1. The check varieties included field maize (CPDK 919), waxy corn (Big White 852), small ear waxy corn (TNS), sweet corn (Accord) and super sweet corn (Sugar 75).

The 20 hybrids, their 5 parents and other 5 check varieties were planted in a randomized complete block design with 3 replications on the raised beds with 20 plants for each row plot, and the plots in the same row were separated by an alley of 25 cm wide. Therefore, there were 30 entries and 90 plots altogether.

Table 1. Inbred lines of vegetable corn used as parents in generating 20 F₁ hybrids.

Inbred Line	Phenotype	Genotype
101 wx	White kernel waxy corn	BtBt Sh ₂ Sh ₂ SuSu $wxwx$
101 bt	White kernel super sweet corn	$btbt$ Sh ₂ Sh ₂ SuSu $wxwx$
101 su	White kernel sweet corn	BtBt Sh ₂ Sh ₂ $susu$ $wxwx$
216 wx	Yellow kernel waxy corn	BtBt Sh ₂ Sh ₂ SuSu $wxwx$
216 sh_2	Yellow kernel super sweet corn	BtBt sh_2sh_2 SuSu $wxwx$

Sample preparation and chemical analysis

The immature ears of waxy corn from each plot were evaluated for sucrose, glucose, fructose, total sugar, phytoglycogen, starch content and amylopectin content at 24 days after pollination. The methods of sample preparation and the details for chemical analysis have been reported previously (Simla *et al.*, 2010). Briefly, the samples were trowed and homogenized in a blender. Pericarp and embryo were discarded. Each sample in a

quantity of 1 g was loaded in a micro-tube for extraction of sugars, phytoglycogen and starch, using different extraction solvents. Sugars were analyzed using high performance liquid chromatography (HPLC) method. Quantification of phytoglycogen was carried out using phenol-sulfuric colorimetric method. Quantification of amylopectin and total starch was carried out using the method similar to that used for the quantification of phytoglycogen except for the absorbed wavelength being 600 nm instead of 490 nm.

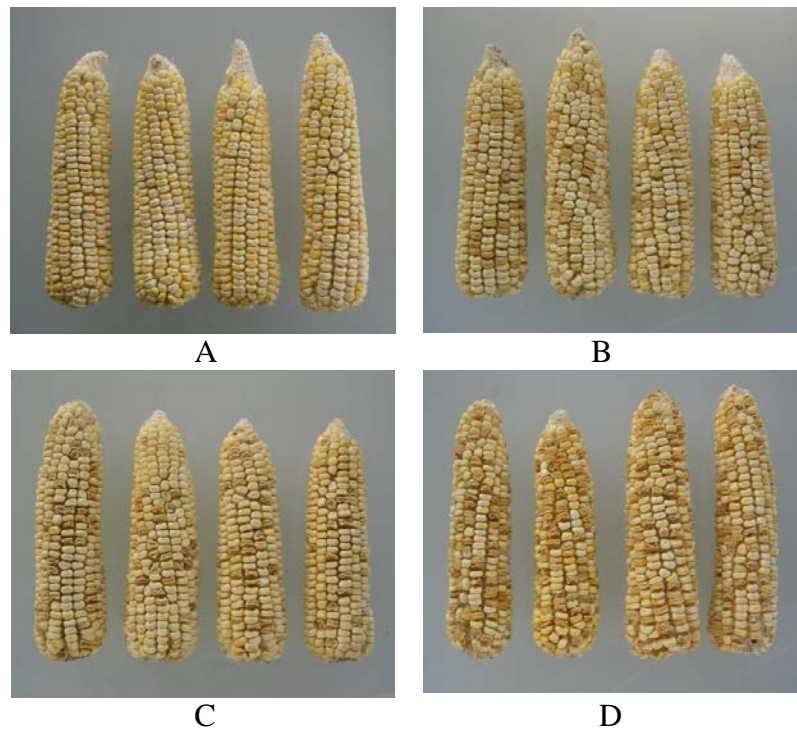


Figure 1. The appearance of dry ear waxy corn with different gene combinations A: waxy phenotype (*wx*) B: waxy and sugary phenotype (*wx* and *su*) C: waxy and brittle phenotype (*wx* and *bt*) (left) and waxy and shrunken phenotype (*wx* and *bt* or *sh₂*) (right) and D: waxy sugary and brittle phenotype (*wx*, *su* and *bt*) (left) and waxy sugary and shrunken phenotype (*wx*, *su* and *sh₂*) (right).

Sensory blind test

Frozen ears were trowled in hot water, cut at 5 cm from the ends of the ears and then cut into small pieces of 2 cm and placed in sealable plastic bags. Seven persons of a volunteer panel were asked to test the corn for sensory evaluation. They were well-informed about how to evaluate sensory parameters such as sweetness, tenderness, stickiness, crispness and overall liking.

Five rating scales were used for all parameters evaluated. For sweetness, drinking water, water with 5% sugar and water with 10% sugar representing rating scales of 0, 3 and 5, respectively, were prepared for comparison. For tenderness, the volunteers were asked to chew the samples 4 to 5 times per sample and rated the samples as 0 for no creamy texture to 5 for highly creamy texture. For stickiness, the volunteers were asked to chew the 4 to 5 times per sample and rated the samples as 0 to 5. For crispness, the volunteers were asked to evaluate pericarp crispness at first bite. The rating scale will be 0 if they feel no crispness and 5 if the pericarp is very crispy. Overall liking was also evaluated in the

same way. 0 represents most unfavorable and 5 represents most favorable.

Statistical analysis

Analysis of variance was carried out for all parameters according to a randomized complete block design (Gomez and Gomez, 1984) using MStat-C software program. Least significant difference (LSD) was used to compare means at 0.05 probability levels.

RESULTS

Carbohydrate characters

Significant differences among tested corn genotypes were observed for sucrose, glucose, fructose, total sugar, phytyglycogen, starch content and amylopectin content (Table 2). The waxy genotypes (101*wx* and 216*wx*) were characterized by low total sugar, low phytyglycogen, high starch content and high amylopectin content. The sugary genotype (101*su*) was characterized by intermediate total sugar, high phytyglycogen, low starch

content and low amylopectin content. Brittle and shrunken genotypes (101*bt* and 216*sh*₂) were characterized by high total sugar, low phytyglycogen, intermediate starch content and intermediate amylopectin content. In parental lines, sugary genotype (*susu*), brittle genotype (*btbt*) and shrunken genotype (*sh*₂*sh*₂) have waxy background (*wxwx*) but

waxy genotype is masked by epistatic effects.

In general for all types of gene combinations, sucrose constituted the largest proportion of total sugar, whereas glucose and fructose contributed smaller proportion to total sugar. The exception for high glucose was also observed in some genotypes such as in 216*wx*, 216*wx*/101*su*, 216*sh*₂/216*wx*, TNS and Accord.

Table 2. Carbohydrate compositions (mg/g fresh weight) of 5 inbred lines, their 20 hybrids and other 5 check varieties harvested at 24 days after pollination.

No.	Treatments	Sucrose	Glucose	Fructose	Total sugar	Phytyglycogen	Starch content	Amylopectin content
1	101 <i>wx</i>	27.1	11.9	8.4	47.5	3.7	306.5	259.5
2	216 <i>wx</i>	13.1	23.6	17.1	53.8	2.1	198.5	167.6
3	101 <i>su</i>	47.8	17.8	7.4	73	116.7	40.5	34.1
4	101 <i>bt</i>	82	20.3	13.2	112.9	1.3	84.5	76.5
5	216 <i>sh</i> ₂	64.9	31.4	18.7	115	2.6	50.5	47.7
6	101 <i>wx</i> /216 <i>wx</i>	17.6	16.3	11.3	45.1	2.4	191.5	152.2
7	216 <i>wx</i> /101 <i>wx</i>	21.6	15.2	11	47.9	2.9	308.8	269
8	101 <i>wx</i> /101 <i>su</i>	43.5	20.6	9.6	73.7	26.8	162	134.5
9	216 <i>wx</i> /101 <i>su</i>	28.7	30.3	18.4	77.4	14.7	125.5	123
10	101 <i>su</i> /101 <i>wx</i>	37.1	19.3	10.2	66.6	20.9	148.8	122.2
11	101 <i>su</i> /216 <i>wx</i>	29.7	24.2	15	68.9	27.2	156.8	128.3
12	101 <i>wx</i> /101 <i>bt</i>	46.7	12.3	8.9	67.9	1.8	164.5	127.3
13	216 <i>wx</i> /101 <i>bt</i>	28.5	14.4	11.2	54.1	1	122.5	94.3
14	101 <i>bt</i> /101 <i>wx</i>	43.9	13.9	10	67.7	1.7	152.2	110.1
15	101 <i>bt</i> /216 <i>wx</i>	31.2	26.7	18	75.9	1.9	182.8	150.4
16	101 <i>wx</i> /216 <i>sh</i> ₂	41.4	12.6	9.3	63.3	1.6	158.8	122.2
17	216 <i>wx</i> /216 <i>sh</i> ₂	31.9	25	16.9	73.9	1.6	120.2	101.1
18	216 <i>sh</i> ₂ /101 <i>wx</i>	37	22.5	15.3	74.8	1.5	125.5	106.7
19	216 <i>sh</i> ₂ /216 <i>wx</i>	30	30.1	20.1	80.2	2.1	133	97.5
20	101 <i>su</i> /101 <i>bt</i>	55.8	20.2	10.5	86.5	8	113.5	97.1
21	101 <i>bt</i> /101 <i>su</i>	48.6	16.2	10.2	75	18.6	128.5	113.1
22	101 <i>su</i> /216 <i>sh</i> ₂	48.6	22.8	12.7	84.2	7.2	116.8	101.7
23	216 <i>sh</i> ₂ /101 <i>su</i>	46.8	25.5	15.8	88.2	8.1	136.5	114
24	101 <i>bt</i> /216 <i>sh</i> ₂	51.6	13.6	9.5	74.7	1.7	130.7	102.8
25	216 <i>sh</i> ₂ /101 <i>bt</i>	57.7	17.1	11.3	86.1	1.6	105.8	84.4
26	CPDK 919 (Ck)	22.7	14.8	9.7	47.2	1.4	209.8	149.7
27	Big White 852 (Ck)	24	15.5	11.1	50.5	2	340.8	326.1
28	TNS (Ck)	11.6	16.7	12.1	40.4	3.9	499.8	432.9
29	Accord (Ck)	26.9	40.5	27.3	94.7	125.1	52.2	35.7
30	Sugar 75 (Ck)	78.7	17.8	10.9	107.4	1.1	37.5	24.1
	LSD _{0.05}	2.31	1.44	0.64	4.21	1.37	22.53	22.04

All hybrids had lower total sugar than their parents with *bt* or *sh*₂ gene, and some hybrids had total sugar similar to that of their parent with *su* gene. The crosses of parents with similar waxy genotype (101*wx*/216*wx* and reciprocal) had similar carbohydrate pattern to those of their parents. The crosses of waxy genotypes (101*wx* and 216*wx*) with sugary genotype (101*su*) and reciprocals had

higher sucrose, glucose fructose and phytyglycogen than their waxy parents but lower than their sugary parent. However, hybrid lines had lower starch content and low amylopectin content than those of their waxy parents.

The crosses and reciprocals between waxy genotypes (101*wx* and 216*wx*) with brittle genotype (101*bt*) and shrunken

genotype (216sh₂) also followed this carbohydrate pattern. These crosses increased mostly sucrose and total sugar but reduced starch content and amylopectin content, whereas phytyglycogen seemed to be not affected (Table 2).

Again, similar carbohydrate patterns were observed in the crosses and reciprocals between sugary genotype (101su) with brittle genotype (101bt) and shrunken genotype (216sh₂). These crosses clearly increased sucrose, total sugar and phytyglycogen but reduced starch content and amylopectin content.

The last type of crosses was between brittle genotype (101bt) and shrunken genotype (216sh₂) for both direct cross and reciprocal. The crosses considerably increased sucrose and total sugar, but reduced starch

content and amylopectin content, whereas phytyglycogen was not affected. Sucrose and total sugar in the hybrid lines were higher than in waxy corn parents (101wx and 216wx) and check varieties (Big White 852 and TNS).

Sensory blind test

The lowest sweetness was found in waxy genotypes (101wx and 216wx), and the highest sweetness was observed in genotypes with gene controlling sweetness (101su, 101bt, 216sh₂, Accord and Sugar 75), whereas the hybrids in general were intermediate (Table 3). However, some hybrids were as sweet as the sugary genotype (101su) although they were not as sweet as brittle and shrunken genotypes (101bt, 216sh₂ and Sugar 75).

Table 3. Sensory characteristics of 5 inbred lines, their 20 hybrids and other 5 check varieties harvested at 24 days after pollination.

No.	Treatments	Sweetness	Tenderness	Stickiness	Crispiness	Overall Liking
1.	101wx	1.06	2.80	3.89	1.88	2.76
2.	216wx	0.94	3.07	2.73	1.52	1.96
3.	101su	1.61	2.75	1.86	1.88	1.40
4.	101bt	3.93	1.85	1.06	3.06	3.24
5.	216sh ₂	3.91	1.38	1.40	2.93	2.69
6.	101wx/216wx	1.30	3.59	3.65	2.12	2.99
7.	216wx/101wx	1.20	3.30	3.46	1.82	2.63
8.	101wx/101su	1.33	2.94	3.59	1.76	2.46
9.	216wx/101su	1.45	3.67	2.85	1.95	2.26
10.	101su/101wx	1.24	3.22	3.24	1.83	2.16
11.	101su/216wx	1.41	3.84	2.97	1.89	2.63
12.	101wx/101bt	2.51	3.19	2.93	2.23	2.88
13.	216wx/101bt	2.02	3.32	2.89	2.13	2.96
14.	101bt/101wx	2.57	2.89	2.83	2.13	2.83
15.	101bt/216wx	3.19	3.33	2.89	2.34	3.17
16.	101wx/216sh ₂	2.72	3.01	2.63	1.93	3.02
17.	216wx/216sh ₂	1.83	2.38	1.90	1.98	1.97
18.	216sh ₂ /101wx	2.99	3.20	2.79	2.28	3.38
19.	216sh ₂ /216wx	2.26	2.71	2.53	2.21	2.45
20.	101su/101bt	2.58	3.31	2.38	2.11	2.37
21.	101bt/101su	2.99	3.07	2.55	2.64	3.03
22.	101su/216sh ₂	2.75	2.98	2.24	2.60	2.75
23.	216sh ₂ /101su	3.08	3.13	2.20	2.50	2.80
24.	101bt/216sh ₂	3.05	2.76	2.28	2.49	2.61
25.	216sh ₂ /101bt	3.19	2.82	2.73	2.33	3.30
26.	CPDK 919 (Ck)	0.38	1.07	1.43	1.35	0.58
27.	Big White 852 (Ck)	1.33	3.46	4.25	1.96	3.94
28.	TNS (Ck)	1.18	2.76	3.89	1.98	3.27
29.	Accord (Ck)	3.06	4.17	1.01	2.72	2.87
30.	Sugar 75 (Ck)	4.75	1.67	0.93	3.74	3.61
	LSD _{0.05}	0.45	0.40	0.27	0.18	0.21

High tenderness was observed in waxy genotypes (101wx, 216wx, Big White 852 and TNS) and sugary genotypes (101su and Accord), and low tenderness was recorded in shrunken and brittle genotypes (101bt, 216sh₂ and Sugar 75). The hybrids generally had high tenderness similar to that of their high parents but the tenderness did not exceed that of the highest genotype (Accord; 4.17).

Similar to tenderness, high stickiness was observed in waxy genotypes (101wx, 216wx, Big White 852 and TNS). Low tenderness was observed in brittle genotype (101bt), shrunken genotypes (216sh₂ and Sugar 75) and sugary genotype (Accord). However, 101su was intermediate. The hybrids were generally intermediate although some hybrids were similar to their high parents.

Similar to sweetness, high crispiness was found in brittle (101bt) and shrunken (216sh₂ and Sugar 75) genotypes, and low crispiness was found in waxy (101wx, 216wx, Big White 852 and TNS) and sugary (101su) genotypes. However, a commercial variety with sugary genotype (Accord) seemed to be intermediate. The hybrids were low to intermediate and neither of them exceeded high parents.

Liking ratings were dependent on varieties rather than types of genotypes. For waxy genotypes, high liking rates were found in Big White 852 and TNS. 101bt and Sugar 75 (shrunken genotype) also had high liking rates, whereas 216sh₂ had low liking rate. Waxy and sugary genotypes generally had low

liking rates. Liking rates of hybrids were low to high depending on hybrids. The poorest liking rate was observed in field maize (0.58).

Correlation

Table 4 showed correlation coefficients between carbohydrate characters and sensory blind test values. The correlation coefficients associated with sucrose, glucose fructose and total sugar were similar in pattern but different in magnitude. The highest positive correlations were observed between total sugar with sweetness ($r = 0.84, P \leq 0.01$) and crispiness ($r = 0.79, P \leq 0.01$). Total sugar was not correlated with general liking ($r = 0.19, P \geq 0.05$). The correlation between total sugar and tenderness was also not significant ($r = -0.30, P \geq 0.05$), but total sugar was negatively associated with stickiness ($r = -0.74, P \leq 0.01$). Phytoglycogen was not significantly associated with tenderness ($r = 0.30, P \geq 0.05$), stickiness ($r = -0.30, P \geq 0.05$) and general liking ($r = -0.26, P \geq 0.05$). For starch and amylopectin, these characters were strongly associated with stickiness ($r = 0.73, P \leq 0.01$ and $r = 0.74, P \leq 0.01$), and they were not correlated with tenderness ($r = 0.14, P \geq 0.05$ and $r = 0.16, P \geq 0.05$) and overall liking ($r = 0.15, P \geq 0.05$ and $r = 0.20, P \geq 0.05$). The negative correlations were found between starch and amylopectin with sweetness ($r = -0.60, P \leq 0.01$ and $r = -0.57, P \leq 0.01$). There were negative correlations between starch and amylopectin with crispiness ($r = -0.49, P \leq 0.01$ and $-0.46, P \leq 0.01$).

Table 4. Correlation coefficients between carbohydrate characters and sensory blind test values (with 29 degree of freedom).

Traits	Sweetness	Tenderness	Stickiness	Crispiness	Over All Liking
Sucrose	0.80**	-0.46**	-0.59**	0.74**	0.23
Glucose	0.23	0.17	-0.41*	0.24	-0.08
Fructose	0.21	0.22	-0.34	0.22	0.05
Total sugar	0.84**	-0.30	-0.74**	0.79**	0.19
Phytoglycogen	-0.04	0.30	-0.30	0.01	-0.26
Starch Content	-0.60**	0.14	0.73**	-0.49**	0.15
Amylopectin Content	-0.57**	0.16	0.74**	-0.46**	0.20

*, ** significant at 0.05 and 0.01 probability levels, respectively

Table 5 showed correlation coefficients among sweetness, tenderness, stickiness crispiness and overall liking. The tenderness was associated with stickiness ($r = 0.51, P \leq 0.01$) but negative correlation was found between stickiness with sweetness and crispiness ($r = -0.60, P \leq 0.01$ and $r = -0.57, P$

≤ 0.01). The relationship between crispiness and sweetness ($r = 0.52, P \leq 0.01$) was strong and positive, and the evaluation panel preferred sweetness ($r = 0.52, P \leq 0.01$) and crispiness ($r = 0.56, P \leq 0.01$) to tenderness and stickiness.

Table 5. Correlation coefficients among sweetness, tenderness, stickiness crispiness and overall liking (with 29 degree of freedom).

	Sweetness	Tenderness	Stickiness	Crispiness
Tenderness	-0.24			
Stickiness	-0.60**	0.51**		
Crispiness	0.91**	-0.25	-0.57**	
Overall liking	0.52**	0.28	0.25	0.56**

** significant at 0.01 probability level

DISCUSSION

Carbohydrate characters

Combining waxy and sweetness characters in a single kernel of corn is not possible because epistatic effect of gene controlling sweetness over waxy gene (Simla *et al.*, 2009) and the possible way to combine these traits is to mix the segregating kernels in the same ears of

several types of hybrids (Figure 1). Therefore, sweetness can be combined into waxy background to improve eating quality. However, types of gene combinations suitable for commercial production have not been well researched. In this study, sugary gene (*su*), brittle gene (*bt*) and shrunken gene (*sh₂*) were incorporated into waxy background of waxy corn for both single gene and combinations of 2 genes (Figure 2).

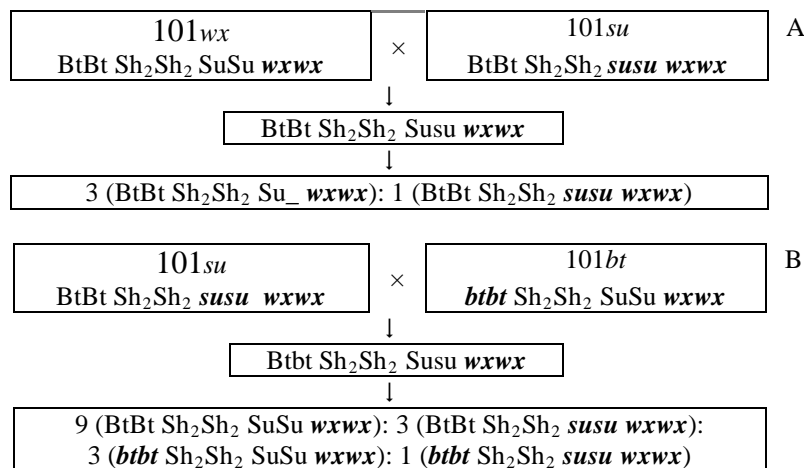


Figure 2. Schematic diagrams of cross combinations and segregation ratios of kernels in F₁ corn hybrids; A: combination of two recessive genes and B: combination of three recessive genes.

In the single gene combination, the segregation ratio in the F₁ hybrid would be 3:1 for waxy kernels to sweet kernels when either *su* or *bt* or *sh₂* is incorporated in to waxy background (Figure 1). In the 2 gene combination, the segregation ratio would be 9:3:3:1 for waxy kernels, sweet kernels for gene 1, sweet kernels for gene 2 and sweet kernels for combination of gene 1 and gene 2, respectively. Therefore, the segregation ratio would be 9:7 for waxy kernels to sweet kernels. Theoretically, the combinations of 2 genes must be sweeter than the combinations

of single sweet genes because they have higher number of sweet seeds in the ears.

The basic knowledge of genes controlling carbohydrate characters in corn has long been available. However, the information on gene combinations in practical breeding of waxy corn to increase sweetness is still lacking, and the authors are not able to compare the results with other investigations. In the ears of waxy corn with genes controlling sweetness, the ears contain both waxy kernels and sweet kernels, and the higher number of sweet kernels was, the sweeter of the ears was.

The increase in number of sweet kernels was in opposite direction of the reduction in number of waxy kernels, and, therefore, the relationship between waxy trait and sweetness was negative.

The results supported the theory in which 4 of 6 crosses with 2 gene combinations were sweeter than all crosses with single gene combination and these results was in agreement with previous studies of the sugary enhancer hybrids (*se₁se₁* genotype) have a higher quality for human consumption than the sugary hybrids (*Se₁Se₁* genotype) (Evensen and Boyer, 1986; Darrah *et al.*, 2003; Ordás *et al.*, 2008a; Ordás *et al.*, 2008b). Therefore, sugary enhancer hybrid has increased consumer demand very fast but the production area was limited due to poor plant stand establishment of hybrid (Evensen and Boyer, 1986; Marshall and Tracy, 2003). However, this is not the problems with double recessive gene of waxy corn because seed of waxy corn hybrid with double recessive is stand very well.

Moreover, 2 crosses that had *101bt* as female parent were as sweet as those with single gene combination. The reason for this exception is not understood and it is possibly due to maternal effect. This hypothesis must be further confirmed. For maximum sweetness, it has been concluded that the hybrids should be combined with 2 sweet genes and the *101bt* should not be used as female parent as it causes lower sweetness.

However, sweetness alone does not determine the preference of the consumers. This was agreement with sweet corn kernel that the quality is unique combination of flavor, texture and aroma (Lertrat and Pulam, 2007). The consumer preference is determined by several quality factors and the eating culture of the consumers. Waxy corn is popular as vegetable among Asian countries and the immigration of Asian people to other parts of the world makes opportunity for market expansion. Furthermore, incorporation of sweet genes into waxy background can diversify waxy corn products.

Sensory blind test and correlation

Tester panel could differentiate among corn genotypes for sweetness, tenderness, stickiness and crispiness, and there were high correlations between sugar characters and

sweetness especially for sucrose and total sugar. This indicated that sensory test can be used for evaluation of sweetness as a surrogate for expensive sugar analysis. This test could also be used to evaluate tenderness, stickiness and crispiness.

In general, brittle and shrunken genes contributed to sweetness and crispiness and sugary gene contributed to sweetness and tenderness, whereas waxy gene contributed to tenderness and stickiness. Their contributions were expressed in hybrids, and, therefore, the combinations of genes in hybrids for the best eating quality of waxy corn had to be determined.

The highest preference was found in a waxy genotype, Big White 852, and the preference for a shrunken genotype, Sugar 75, and a brittle genotype, *101bt*, was also high. Moreover, some hybrids with single gene combinations and double gene combinations for sweet traits were also received high preference. This indicated that preference for waxy corn in this study was dependent on individuals of the tester panel. This means that individuals of the test panel preferred differently and their preference was not in the same pattern. The results also indicated the need for further research on consumers' behavior for segmentation of the consumers who prefer different types of vegetable corn.

The relationships between overall liking with sweetness and crispiness were high and the relationships between overall liking with tenderness and stickiness were low. Sweetness and crispiness are characters of sweet corn, whereas tenderness and stickiness are characters of waxy corn. When these characters are combined in the same ears, it is true that consumers prefer sweetness and crispiness. The results indicated that consumers like waxy corn with sweet character and incorporation of genes controlling sweetness into waxy background will increase consumer's preference.

Three recessive genes controlling sweetness (*su*, *bt* and *sh₂*) were incorporated into waxy corn for single genes and two-gene combinations of these genes in order to improve eating quality of waxy corn. The combinations of 2 sweet genes provided maximum sweetness in waxy corn but *101bt* was not appropriate for use as female parent because it could lower sweetness.

ACKNOWLEDGEMENTS

We are grateful for the financial support provided by the University Staff Development Program, Mahasarakham University, the Commission on Higher Education, Thailand (Grant No. 028/2548), the National Center for Genetic Engineering and Biotechnology, Bangkok, Thailand. The authors would like to thank Mahasarakham University Development Fund and the Faculty of Agriculture KKU for providing financial support for research presentation and manuscript preparation activities, respectively. Mr. Thawan Kesmala is also acknowledged for his critical reading and valuable comments during manuscript preparation.

REFERENCES

- Creech RG (1965) Genetic control of carbohydrate synthesis in maize endosperm. *Genetics*. 52:1175–1186.
- Darrah LL, McMullen MD, Zuber MS (2003) In Corn: Chemistry and technology, *Breeding, genetics, and seed corn production*, eds., White PJ, Johnson LA (American Assn. Cereal Chemists, St. Paul, MN), 2nd ed, pp 35–68.
- Evensen KB, Boyer CD (1986) Carbohydrate composition and sensory quality of fresh and stored sweet corn. *J. Amer. Soc. Hort. Sci.* 111:734–738.
- Ferguson JE, Dickinson DB, Rhodes AM (1979) Analysis of endosperm sugar in sweet corn inbred (Illinois677a) which contains the sugary enhancer (se) gene and comparison of se with other corn genotypes. *Plant Physiol.* 63:416–420.
- Gomez KA, Gomez AA (1984) *Statistical procedures for agricultural research*. John Wiley and Sons, Inc. London, UK (2nd ed.) pp. 680.
- Holder DG, Glover DV, Shannon JC (1974) Interaction of shrunken-2 with five other carbohydrate genes in corn endosperm. *Crop Sci.* 14(5):643–646.
- Lertrat K, Pulam T (2007) Breeding for increased sweetness in sweet corn. *Int J Plant Breed.* 1(1):27-30.
- Lertrat K, Thongnarin N (2008) Novel approach to eating quality improvement in local waxy corn: Improvement of sweet taste in local waxy corn variety with mixed kernels from super sweet corn. *Acta Hoertic.* 769:145–150.
- Marshall SW, Tracy WF (2003) In Corn: Chemistry and technology, *Sweet corn*, eds., White PJ, Johnson LA (American Assn. Cereal Chemists, St. Paul, MN), 2nd ed, pp. 537–569.
- Ordás B, Malvar RA, Ordás A, Revilla P (2008a) Reciprocal differences in sugary × sugary enhancer sweet corn hybrids. *JASHS.* 133(6):777–782.
- Ordás B, Revilla P, Ordás A, Malvar, RA (2008b) Hybrids sugary × sugary enhancer of sweet corn: A valuable option for cool environments. *Scientia Hort.* 118:111–114.
- Simla S, Lertrat K, Suriharn B (2010) Carbohydrate characters of six waxy corn varieties as affected by harvest time and storage duration. *Asian J Plant Sci.* 9(8):463–470.
- Simla S, Lertrat K, Suriharn B (2009) Gene effects of sugar compositions in waxy corn. *Asian J Plant Sci.* 8:417–424.
- Thongnarin N, Lertrat K, Buddsarakul S (2005) Sumlee Esarn and KhonKaen Salubsee: Two new single cross waxy corn hybrids. *Agri Sci J.* 36:5–6.
- Thongnarin N, Lertrat K, Techawongstien S (2008) Combining ability study in waxy corn (*Zea mays* var. *ceritina*) inbred lines. *Acta Hoertic.* 769:151–156.