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Improving Kernel Weight and Number in Some Maize Subspecies Crosses

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ABSTRACT

Four maize subspecies inbreds were crossed to each other in the fall planting season in 2018. This was on the x- farm of the college of Agric. Engineering Sciences /University of Baghdad. F1 ear seeds were harvested, dried and shelled. The maize subspecies inbreds were *Zea mays* L., saccharata (S), *Zea mays* L. indentata, D and J, and *Zea mays* L., indurata (P) of popcorn. In fall season of 2019, the F1 crosses seeds were grown on the same farm for comparison. Data were recorded on all crosses plants and their four inbreds. The objective of this trial was to determine the inheritance of kernel colour, kernel weight, kernel shape and their relationships with plant grain yield. The results showed that plant height of crosses was not coincided with plant leaf area. Number of ear kernels and kernel weight were the direct components of grain yield. Number of ear kernels was a result of number of kernels per row and number of ear rows. Two crosses gave significantly higher grain yields. They were D×S, and J×D, which gave 13.3 t/ ha and 11.9 t/ha, respectively. The genetic diversity of D line was thought to be due to its late flowering. Shape of kernel was thought to be controlled by many genes, and so, kernel colour. Kernel dent was thought to be dominant or incomplete dominant. It was concluded that hybrid vigour is controlled by many plant attributes, such as SNP, transposons, methylation, epigenome, and parental imprinting of crossed inbreds. We recommend identification of some diverged phenotypic plant traits of crossed inbreds to have elite hybrids.

Keywords: maternal and paternal expressed genes, eigenomic, embryos.

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INTRODUCTION

Maize (*Zea mays* L.) hybrids are widely used all over the world for its high yield and tolerance to biotic and abiotic stresses. Hybrid vigour became very common in most of fruit trees and vegetables (Barber, *et al* 2012; Hanna and Kelsey, 2017; Tollenaar and Lee, 2011). There is no one proved theory explains hybrid vigour (Elsahookie, 1990; Elsahookie, 2006; Lauss *et al* 2018). Maize productivity and production are increasing in the world. Tollinaar and Lee (2011) stated that maize productivity in USA have increased by about 118 kg /ha/yr since 1930s, and with about 90 kg / ha /yr in Canada. Chen *et al* (2005) reported that new maize hybrids are higher in productivity than old hybrids by about 1.3 – 3.1 t/ ha, depending on hybrid, environment and growth variables. Chen *et al* (2017) stated that new maize hybrids kernels are 15% - 23% heavier weight, and by about 150 kernels per squarred meter more than old hybrids. Although there are many traits promote grain yield, such as stay – green leaves, erect leaves, higher parents grain yield … etc, but kernel number by unit of area and kernel weight still the most prominent components of grain yield. Liu *et al* (2017) reported that five pairs of genes control kernel weight in maize. However, kernel weight and number of kernels per unit of area are affected by hybrid vigour. This trial is dealing with maize subspecies crosses and their F1 reciprocals in term of some agronomic traits, plant kernel number, kernel weight, and their relationships with final grain yields of crosses.

MATERIALS AND METHODS

On the x- farm of the College of Agric. Engineering Sci./ University of Baghdad, a piece of land was ploughed, and divided into furrows of 90 cm wide beds. The planting was on both sides of bed, male and female to be crossed reciprocally. This was in mid-July 2018, a fall planting season in Iraq. There were four maize (*Zea mays* L.) subspecies inbred lines; sweet corn (S) sacchara, J and D indentata (dented) and popcorn flint (P) indurata. Weed control, irrigation and fertilization were done (Elsahookie, 1990). There were 48 furrows for the four inbreds to be crossed diallel. Four extra furrows were left for inbred propagation by selfing. Before silking, all ears were bagged by shoot bags. When silks appear with about 5 cm long, they were cut – off and rebagged to be crossed after 2 days from bagged tassels of required parent. At time of full maturity, ears were harvested and husked. Each group of crosses was kept in potato sacs and left to air dry on benches in wide room under fanning. After 5 weeks, ears were threshed by a hand sheller, cleaned and kept into envelops for planting in next year. In July 2019, at the same location, land was prepared and divided into plots of 3.0× 2.8 m. Every plot contained four rows 70 cm apart and 25 cm between hills. Two seeds were planted in each hill then thinned to one seedling 3 weeks after emergence. Weed control, fertilization, and irrigation were done as recommended (Elsahookie, 1990). Planting population density was about 57,000 plant / ha. There were twelve diallel crosses and the four parents were planted to cheek their agronomic traits. The design used was RCBD with 4 replicates. At time of full tasseling, one leaf dimension was measured of the leaf below ear leaf on five well-guarded plants of each experimental unit. The formula used to estimate plant leaf area was plant leaf area = L^2 b× 0.75, Lb = leaf below ear leaf (Elsahookie, 1985). After measuring leaves of plants of the experiment, we referred to ready tables give us plant leaf area (Elsahookie and Cheyad, 2013). The five plants taken for leaf area estimation were tagged to record all agronomic traits and mean plant grain yield for all crosses and their parents. At the time of full maturity, ears of the five plant of each experimental unit were harvested, husked, and left on benches in a wide room under fanning. They were left to five weeks since the grain moisture reached 14% -15%. Samples of 100 kernels of each experimental unit were taken to record kernel traits and their percentage in shape, color, and kernel dent. Embryos were taken off endosperms by a special knife. Endosperms and embryos were weighed, and percentages determined. Data were tabulated and statistically analyzed.

RESULTS AND DISCUSSION

Some agnomic traits of maize subspecies crosses and their parents are shown in Table 1. Some other traits, such as ear length, and ear diameter were found of less importance so, they were not shown. Plant height of crosses and parents were significantly different. Plant height of crosses ranged between 136.0 of cross S×J to 178.0 cm for J×P, while the tallest one among all was the popcorn parent. Plant leaf areas were also different, but plant height was not necessarily to be always positively correlated with leaf area. The cross S×P had 154.3 cm height with 0.490 m^2 leaf area compared to pop parent of 198.8 cm height and 0.485 m^2 plant leaf are. Leaf area, leaf duration, erect leaves, and stay-green leaves are effective if they are all available in the genotype, for leaf area is the main source of metabolites which nourish the sink, the main yield. Number of rows is important to give more kernels per ear if the row has high number of kernels. However, row number of subspecies crosses ranged between 12 to 18 rows. The cross D×P has 12 rows and gave 71g plant grain yield, but S×J had 12 rows too, but gave 134g plant grain yield. That difference goes back to number of kernels per row. At the same time, the higher row number (18 row) of the cross D×S gave 233g plant grain yield, that was due to high row kernel number, high ear kernels, and heavy kernel weight. The maximum row kernel number was 46 in two crosses; D×J and P×J while the lower was 25 with sweet corn parent. Ear kernel number counts on number of rows and kernel per row. The higher ear kernel number was in the cross D×S, which gave the higher plant grain yield, which is equivalent to 13.3 t/h if we consider planting population density as 57,000 plants per hectare.

The second rank grain yielder is the cross J×D which gave 208g/ plant which is equivalent to 11.9 t/ha, and they are significantly different. We need to note here that both crosses shared the paren (D), this is a long life line, it delays maturity about 14 days as compared to other parents, and this is an important trait that give a higher probability in genetic variation to other parents crossed with.

Table 1. Some agronomic traits of maize crosses, reciprocals and parents

Crosses and parents		Plant height cm	Leaf area m^2	Row/ ear	Kernel/ row	Kernel/ ear	Kernel Weight g	Embryo Weight g	Plant Yield g
1	**S×D**	159.3	0.490	16	42	647	0.185	0.013	124
2	**D×S**	160.8	0.480	18	39	702	0.333	0.016	233
3	**J×S**	141.3	0.454	14	29	400	0.323	0.014	129
4	**S×J**	136.0	0.469	12	33	396	0.338	0.016	134
5	**D×J**	154.0	0.471	12	46	552	0.310	0.016	171
6	**J×D**	158.3	0.484	14	42	585	0.353	0.018	208
7	**D×P**	147.3	0.463	12	29	351	0.200	0.014	71
8	**P×D**	138.8	0.466	16	46	460	0.200	0.011	92
9	**P×S**	163.5	0.475	14	33	462	0.373	0.014	172
10	**S×P**	144.5	0.483	14	25	350	0.250	0.012	88
11	**J×P**	178.0	0.480	16	35	560	0.210	0.013	118
12	**P×J**	160.0	0.469	12	46	549	0.220	0.012	121
13	**S**	148.8	0.498	14	25	354	0.170	0.013	60
14	**P**	198.8	0.485	14	39	539	0.250	0.010	135
15	**D**	178.0	0.466	14	42	579	0.260	0.028	151
16	**J**	165.0	0.450	14	38	532	0.225	0.021	120
L.S.D 5%		**007**	**0.062**	**01**	**04**	**052**	**0.011**	**0.002**	**015**

Hybrid vigour of ear kernel number was 21.2% and for kernel weight 21.9%, while it was 54.3% for plant grain yield, that for grain yield counts on many attributes, it is worthy to note that some crosses of heavy kernels were of heavy embryo such as D×S, S×J and J×D. These traits are good to have high grain yield crosses. The share magnitude of inbreds in the endosperm of their crosses are not the same in all cases of crossing, e.g, the paren D when used male with S line did not give high plant grain yield but it did when used as female with the same S parent. Whereas, when this parent D used as female with, J, it did not give high plant grain yield, but it did when used as male with the same parent J. Sabilli and Larkins (2009) reported that endosperms continue mitotic divisions until up to 25 days after fertilization. Xin *et al* (2013) stated that the share of embryo in the grains of F1will be 1 materna l:1 paternal, for it is a diploid, while endosperm as a triploid, its share is 2 materna l:1 paternal. At the same time, these researchers have analyzed the familiar U.S.A. maize hybrid; Mo17×B73 and its, reciprocal. When they analyzed genomes of these two parents, they found over 6 million SNPS, 157994 insertion, and 194549 deletions between the inbreds, besides hundreds base pairs short gaps, and millions of imprinted genes.

Grossmann *et al* (2011) tested parents and their hybrid in *Arabidopsis thaliana*, and found the big difference between parents and their hybrid was in percentage of sRNA (21 nt – 24 nt), and they believed that some of those sRNAs have epialleles which were related to hybrid vigour, i.e that gene imprinting and DNA methylation differential of MEGs and PEGs are important to hybrid vigour. One of these results was confirmed by Lin *et al* (2017). On the other hand, Zhang *et al* (2011) concluded that there is a complex regulation of gene imprinting in maize endosperm, for example, they found 179 genes of protein coding were expressed in the endosperm were imprinted along with 38 long noncoding RNAs.

Attributes related to hybrid vigour looks too many so, that will be no specific genes control this phenomenon in all hybrids. Epigenetics, gene imprinting, translation, transcription…etc all could be related to hybrid vigour. Bressmen and Zhu (2014) mentioned that in some living beings, transposable elements could share up to 90% of their genomes. Dong *et al* (2019) stated that genomic imprinting in flowering plants occurs primarily in the enelosperm. Grimanelli *et al* (2005) reported that the transcriptional activation of the two parental genomes might not occur at the same time in the zygote during early kernel development, while Nodine and Bartel (2012) reported that both paternal and maternal genomes might contribute equally in early stages of embryonic development, but as we have seen in results that the share of parent is not equal when it is male or female. Regal *et al* (2016) when studied *A. thaliana* hybrid found that when methylation in the hybrids goes down, many genic expressions appear, and they were not existing in their parents.

This trial has been done to explore the role of maize kernel in maize hybrids. A similar research was applied previously and showed that sweet corn kernel weight was increased by 50% when crossed with feed corn, and a negative hybrid vigour in grain yield was found among other crosses (Elsahookie 2005). Meanwhile Yousif and Elsahookie (2007) extended thier research by using another inbreds and found that one cross produced two ears per plant and gave 1575 kernels, and another cross had 61% hybrid vigour in grain yield. A similar result on number of rows per ear was not changed that much through crossing, and most crosses gave ear rows between 16-18 rows.

Another study on relationship between genetic distance between inbreds and hybrid vigour. Elsahookie and Al-Falahi (2013) found that genetic distance between parents was not the reason of hybrid vigour. This indicates that hybrid vigour counts on several traits between inbreds crossed parents. Elsahookie *et al* (2018) worked on selecting plants from inbred population and found that selecting plants of stay –green leaves, or very late or very early flowering or of long ears, and found that crosses of these newly selected inbreds from same inbred population gave an increase of 2 t/ha of grains over the hybrid yield of original inbreds. Epigenomics and diversity degree of crossed inbreds are important to have good hybrids (Elsahookie *et al* 2019). Finally, this article showed that the most direct and prominent grain yield components in maize hybrid grain yield are number of kernels per plant and kernel weight. Meanwhile, the percent of share in endosperm weight and / or plant kernel numbers differ when we have reciprocal crosses. This implies that male and female share in the endosperm is not necessarily to be equal. The maximum ear kernel obtained was 702 kernels, and the higher grain yield was 13.3 t / ha.

According to shape of kernel, dent of kernel and colour are shown in Tables 2 and 3. As we note in Table 2, the shape of kernels was inherited in different percentages indicating that this trait is under several pairs of genes. These percentages differ when we compare reciprocal crosses. Meanwhile, dent of kernel was less affected by reciprocal. The flint kernel in general was dominant over dent kernel. Crossing sweet corn line with dent or flint (popcorn) produce dent kernels. Colour of kernels in crosses gave similar differences of that of kernel shape (Table 3). This is due to number of genes control aleurone color. YYY gives dark yellow kernel color, and yellow colour will be less when we have YYy and Yyy, and when we reach yyy the colour will be white (Elsahookie,1990), beside the reflection of endosperm colour on kernel colour. Hybrids produced by crossing white × white parents will hold white colour, and that crossing of yellow × yellow parents will give a yellow kernel hybrid.

Table 2. Shape and dent of kernels of F1 maize subspecies crosses and parents.

Crosses and parents		Shape of kernel	Dent of kernel
1	**S×D**	T40%, R 40%, SH20%	F 70%, sh 30%
2	**D×S**	tR75%, SH 10%, R15%	D100%
3	**J×S**	SH45%, T15%, R20% SQ20%	D 55%, sh 45%
4	**S×J**	T85% tR 5%, SH 10%	D 90%, sh 10%
5	**D×J**	tR100%	D100%
6	**J×D**	T100%	D100%
7	**D×P**	tR100%	F100%
8	**P×D**	tR100%	F100%
9	**P×S**	SH 30%, tR 70%	D 70%, sh 30%
10	**S×P**	SH 45%, tR 55%	D 55, sh 45%
11	**J×P**	tR100%	F100%
12	**P×J**	Tr85%, R 15%	F100%
13	**S**	SH 100%	sh100%
14	**P**	tR100%	F100%
15	**D**	tR100%	D100%
16	**J**	SQ 100%	D100%

Abbreviations: **SQ** = square, **SH** = shrunken, **tR** =tipped round, **R**= round, **T**= triangular, **t** = tipped, **D**= dent, **F** = flint.

Table 3. Kernel color of F1 subspecies crosses and parents.

Crosses and parents		Colure percentage of kernels
1	**S×D**	W 30%, LY 55%, T 15%
2	**D×S**	W 50%, LY 45%, T 5%
3	**J×S**	DY 80%, LY 20%
4	**S×J**	LY 90%, T 10%
5	**D×J**	LY 40%, W 25 %, DY 35%
6	**J×D**	W 45%, DY 15%, LY 40%
7	**D×P**	W 50%, LY 50 %
8	**P×D**	W 50%, LY 50 %
9	**P×S**	LY 70, T 30%
10	**S×P**	LY 55%, T 45%
11	**J×P**	LY 85%, DY 15 %
12	**P×J**	LY 60 %, W 35%, DY 5%
13	**S**	T 100%
14	**P**	DY 100
15	**D**	W 100%
16	**J**	DY 1005

Abbreviations: **W** = white, **DY**= dark yellow**, LY** =light yellow, **T**= transparent.

CONCLUSION

Hybrid vigour is a trait counts on several traits such as number of SNP, insertion, deletions, base pair shot gaps and imprinted genes. sRNAs (21 nt-24 nt), DNA methylation, and number of transposons are also thought to be related to hybrid vigour, and this will differ with different crossed inbreds. Number of kernels per ear and kernel weight were the most directly related traits to hybrid grain yield. The hybrid which gave 54.3% grain yield hybrid vigour had 21.2% ear kernel number hybrid vigour coincided with 21.9% kernel weight hybrid vigour. Some reciprocal crosses were significantly different in grain yield due to maternal effect of endosperm (2n) as compared to (n) paternal endosperm. This explains the different effect of inbred when it is female or male. Sweet corn inbred when crossed to dent inbred, it gave a 50% increase in the F1 kernel weight. Flint kernel was in general dominant over dent. The colour of kernels is highly affected by female parent than by male parent. When the female was white kernel and crossed to sweet or pop inbreds, it gave a 50% white kernels. Shape of kernel is thought to be controlled by several genes. There are many other traits should be known to understand the appearance of commercial elite hybrids.

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