Colegio de Postgraduados

Heterosis in the germination process and seed characteristics of the maize hybrid (*Zea mays* L.) HAZUL 10E

Gutiérrez-Hernández, Germán F.[1*]; **Arellano-Vázquez, José L.**[2]; **Ceja-Torres, Luis F.**[3]; **García-Ramírez, Elpidio**[4]; **Quiroz-Figueroa, Francisco R.**[5]

[1] Instituto Politécnico Nacional. Unidad Profesional Interdisciplinaria de Biotecnología. Av. Acueducto s/n. La Laguna Ticomán. C. P. 07340. Ciudad de México, México.
[2] Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias. Campo Experimental Valle de México. C. P. 56250. Coatlinchán, Estado de México, México.
[3] Instituto Politécnico Nacional. Centro Interdisciplinario de Investigación para el Desarrollo Integral Regional Unidad Michoacán. Justo Sierra 28. C. P. 59510. Jiquilpan, Michoacán, México.
[4] Universidad Nacional Autónoma de México. Facultad de Química. Av. Universidad y Copilco. C. P. 04510. Ciudad de México, México.
[5] Instituto Politécnico Nacional. Centro Interdisciplinario de Investigación Para el Desarrollo Integral Regional, Unidad Sinaloa. Blvd. Juan de Dios Bátiz P. 250. Col. San Joachín. C. P. 81101 Guasave, Sinaloa. México.
* Correspondence: gfgutierrez@ipn.mx

ABSTRACT
Objective: To quantify the heterosis in physical and physiological characteristics in seeds of simple and trilinear crosses of the HAZUL 10E corn hybrid.
Methodology: Seeds of the genotypes conforming to the HAZUL 10E corn hybrid (endogamic lines and simple and trilinear crosses) were used. The experimental design was completely randomized with three repetitions. Physical (weight, width, thickness, length, volume, density, and the width/length and thickness/length ratios) and physiological (normal and abnormal seedlings, death seeds, and lengths and dry matter of plumule, radicle, and total) characteristics of the seeds were evaluated. Heterosis and heterobeltiosis were determined, and the differences between both crosses were statistically tested with Student's t-test.
Results: Heterosis and heterobeltiosis were higher in the single cross than in the trilinear one. In the single cross, the highest values of both heterosis corresponded to the plumule, radicle, and total dry matter; followed by weight and volume seed. In the trilinear, the highest values corresponded to normal seedlings, radicle length, and biomass total. The variables corresponding to shape seed, normal seedlings formation, and radicle elongation, responded better to hybridization in the trilinear cross.
Study limitations: None presented.
Conclusions: In the single cross, heterosis increased the seed size and accumulated biomass in seedlings; while in the trilinear cross, it affected the shape of the seeds and the produced seedlings.

Keywords: Hybrid vigor; pigmented corn; seed germination; seed quality.

Citation: Gutiérrez-Hernández, G. F., Arellano-Vázquez, J. L., Ceja-Torres, L. F., García-Ramírez, E. & Quiroz-Figueroa, F. R. (2023). Heterosis in the germination process and seed characteristics of the maize hybrid (Zea mays L.) HAZUL 10E. *Agro Productividad*. https://doi.org/10.32854/agrop.v16i11.2730

Academic Editors: Jorge Cadena Iñiguez and Lucero del Mar Ruiz Posadas

Received: May 21, 2023.
Accepted: October 25, 2023.
Published on-line: December 27, 2023.

Agro Productividad, 16(11). November. 2023. pp: 119-125.

INTRODUCTION

Corn (*Zea mays* L.) is the most important crop in the world by planted area and production volume [1]. In 2021, 7.4 million hectares were planted in Mexico and 27.5 million tons of grain were harvested [2]. Blue corn is relevant for its wide use in traditional

Mexican cuisine (tortillas, tlacoyos, sopes, etc.) [3], due to its exotic color and its attributes of flavor, texture, and nutraceutical benefits [4].

In Mexico's central states (Estado de Mexico, Puebla, Tlaxcala, and Mexico City) 150 thousand hectares of blue corn are planted with native varieties, traditional technology, and rainfed conditions, and 300 thousand tons of grain are harvested, which do not satisfy the demand for this food.

The current grain yield of blue corn is 1.9 t ha^{-1} [2]. It is feasible to significantly increase it through the utilization of hybrids [5] developed on the genetic variability available in the region of interest [6].

At the Instituto Nacional de Investigaciones Forestales Agrícolas y Pecuarias (National Institute of Agricultural and Livestock Forestry Research, INIFAP), blue corn experimental hybrids were developed for high valleys (2400-2800 meters above sea level), with yields of 6.4 to 8.2 t ha^{-1}, from which HAZUL10 stood out for producing 8.2 t ha^{-1}, on average, in high, medium and low productivity environments [7].

Hybridization was described at the beginning of the 20th century [8]. Since then, it has successfully improved corn and is currently still applied for obtaining hybrids that help to satisfy the global demand for this grain [9]. With hybridization, the F1 expresses larger morphological dimensions and better grain yield than unrelated endogamic parents [10], to a magnitude dependent on its genetic background and its production environment. The above responses are caused by heterosis, a complex, and poorly understood phenomenon, but widely used in the corn and other basic crops improvement.

The objective of the present research was to quantify heterosis and heterobeltiosis in the physical and germinative characteristics of seeds of simple and trilinear crosses of the HAZUL 10E corn hybrid.

MATERIALS AND METHODS

Genetic material

Seeds from the parental genotypes of the experimental blue corn hybrid HAZUL 10E were used: L11 (BXCC-54-11-1-1-1), L12 (BXCC-5-9-6), L10 (NXOAX-19-5-1-1-2), Simple Cross A (BXCC-54-11-1-1-1) × (BXCC-5-9-6) and the Trilinear Cross B [(BXCC-54-11-1-1-1) × (BXCC-5-9-6)] × (NXO-AX-19-5-1-1-2). In this document, the data of the simple cross (treatment A) and the trilinear cross (treatment B) were presented. The hybrid was developed in the Maize Program of the Valle de México Experimental Field from the Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias (CEVAMEX, INIFAP).

Physical characteristics

Seed weight (SW). 100 seeds (mg) were weighed on an analytical balance (AE Adam P W 184, precision 0.1 mg). The width (SWID), thickness (ST), and length (SL) of the seeds were measured (mm) with a digital vernier (Mitutoyo CD-6 CSX). The volume (SV, mm^3), relative density (SRD, g/cm^3), and the SWID/SL and ST/SL ratios of the seeds were calculated according to the corn varietal descriptors [11-13].

Germinative characteristics

Normal germination

The standard germination test was used [14]. The seeds were placed in paper towels saturated with humidity in a germination chamber at 25 °C and 100% r. h. After seven days of incubation, the seedlings with complete morphology and free of pathogens (NS), abnormal seedlings (AS), and dead seeds, *i.e.*, lacking metabolic signs, (DS) were counted (%). NS was considered equivalent to seed germination.

Seedlings development

In normal seedlings, the length of plumule (PL), radicle (RL), and total length (TL) were measured (mm); these structures were then, dried in an oven (80 °C, 4 d) and their dry weight (PDW, RDW, and TDW, respectively) was determined (mg).

Heterosis estimation

Heterosis (%) was calculated in relation to the parent's average of the cross

$$H=\left[\left(F1-AP\right)/AP\right]*100$$

and also to the best parent (heterobeltiosis)

$$HB=\left[\left(F1-MP\right)/BP\right]*100;$$

where $F1$=Value of the cross, AP=Average parents of the cross, and BP=Best parent of the cross [15].

Statistical analysis

Student's paired data test was used to test the significance ($P\leq0.05$ and $P\leq0.01$) of the differences in H and HB within each cross, as well as that of the differences in H or HB between the mentioned crosses. The value of this difference was multiplied by (−1) to indicate the variation in the variables [16].

The statistical processing of the results was done with the SAS statistical software (ver. 9.2) [17].

RESULTS AND DISCUSSION

In response to the hybridization of the crossings involved in the formation of the trilinear corn hybrid HAZUL 10E, heterosis (H) exceeded heterobeltiosis (HB) in all measurements, confirmed by the negative values of the difference between both (Table 1). The above is explained because the H was calculated in relation to the average number of parents of the cross and the HB regarding the best parent, *i.e.*, it was more rigorous [15].

Both types of heterosis were higher in cross A than in B, because the genetic recombination occurred between two inbred lines in the first case, and between a simple cross and an endogamic line, in the second. This situation coincides with the argument

Table 1. Values (%) of heterosis (H) and heterobeltiosis (HB) of the simple (A) and trilinear (B) crosses of the experimental blue corn hybrid H10 E (UPIBI, IPN. Mexico City, Mexico, 2020).

Variable	Genotypes							
	A				B			
	H	HB	H−HB	t	H	HB	H−HB	t
SW	63.93	45.07	−18.86	**	13.61	−3.21	−16.82	**
SV	35.01	22.85	−12.16	**	22.44	14.75	−7.69	*
SL	17.31	11.3	−6.02	*	4.79	2.20	−2.59	**
SWID	15.37	13.34	−2.03	ns	7.83	5.86	−1.97	ns
ST	−0.36	−6.07	−5.71	**	9.71	5.35	−4.36	*
SRD	12.95	0.42	−12.53	**	0.05	−10.72	−10.77	**
SWID/SL	−2.06	−8.63	−6.57	**	3.80	3.80	0	ns
ST/SL	−15.07	−18.19	−3.12	**	5.63	5.93	0.3	ns
NS	8.93	−4.58	−13.51	ns	62.61	35.62	−26.99	ns
AS	25	−25	−50	ns	−50.00	−50.00	0	ns
SD	0	−29.58	−29.58	ns	−55.71	−62.50	−6.79	*
PL	18.38	9.02	−9.36	ns	−4.21	−13.79	−9.58	*
RL	8.08	3.28	−4.80	ns	45.52	34.62	−10.9	*
TL	11.38	5.78	−5.60	ns	25.55	19.46	−6.09	ns
PDW	92.16	61.04	−31.12	*	−0.03	−15.45	−15.42	*
RDW	84.79	52	−32.79	*	78.86	38.52	−40.34	*
TDW	87.96	58.18	−29.78	ns	41.68	14.04	−27.64	*

*=Significant (P≤0.05), **=Highly significant (P≤0.01), ns=Not significant. SW=Seed weight, VS=Seed volume, LS=Seed length, WS=Seed width, TS=Seed thickness, RDS=Relative seed diameter, PN=Normal seedlings, PA=Abnormal seedlings, SI=Seeds inert, LP=Plumule length, LR=Radicle length, TL=Total length, PDW=Plumule dry weight, RDW=Radicle dry weight, TDW=Total dry weight, WS/LS=Seed width/Seed length, TS/LS=Seed thick−ness/Seed length.

that the heterotic response depends on the genetic background of the genotypes involved in the crossing [18].

In genotype A, the highest magnitudes of H and HB were noted in the characters corresponding to the dry matter production of the seedlings (PDW, RDW, and TDW), followed by weight (WS) and volume seed (SV); while, for B, the highest values corresponded to germination (NS), biomass (RDW and TDW) and radicle elongation (LR). In these variables, the effects of the metabolic process called heterosis or hybrid vigor were expressed, through which the offspring, in this case, the crosses, assume larger dimensions compared to the parents in certain variables [19, 20], in which the phenotypic ones predominate; However, in the present study heterosis also occurred in physiological ones, as was the case of NS, RDW, TDW, and RL.

Thus, the response to heterosis varied depending on the cross and the measured trait, although total and radicle biomass showed a high response in both crosses. The H and HB values of the NS in genotype B also stood out, this evidenced the favorable genetic recombination of the L10 line, which served as a pollinator for the L11×L12 simple cross, for seed germination, which combined with the heterosis manifested in RL, RDW, and TDW, conferred favorable characteristics for the establishment of the assessed hybrid in the crop field. This result can be attributed to L10 coming from the Central Valleys of Oaxaca state, Mexico, a different environment from that in the Valley of Mexico, in which L11 and L12 were collected; so the lines belong to dissymbol heterotic groups, whose genetic recombination had a positive effect on the physiological quality of HAZUL 10E [18].

In A, the differences between H and HB (Table 1) were significant in 53% of the variables, while in B in 64%; Both crosses concur in that these significances were shown by seed size and dry matter production (SW, SV, SL, ST, SRD, PDW and RDW); this data showed the wide heterogeneity of the L11 and L12 parental lines regard the aforementioned variables, which was balanced considering the average of parents (H) but not by including only the best of them (HB), therefore, the differences were significant.

In the same sense, comparing H or HB between the crosses (Table 2) confirmed that both occurred with a greater magnitude in A than in B. In A, there were significant

Table 2. Differences between heterosis and heterobeltiosis of the simple (A) and trilinear (B) crosses of the experimental blue corn hybrid H10 E (UPIBI, IPN. Mexico City, Mexico, 2020).

Variable	Genotypes							
	A	B	(H−H)	t	A	B	(HB−HB)	t
	H	H			HB	HB		
SW	63.93	13.61	−50.32	**	45.06	−3.21	−48.27	**
SV	35.01	22.44	−12.57	ns	22.85	14.75	−8.10	ns
SL	17.31	4.79	−12.52	**	11.30	2.20	−9.10	**
SWID	15.37	7.83	−7.54	ns	13.34	5.86	−7.48	ns
ST	−0.36	9.71	10.07	ns	−6.07	5.36	11.43	ns
SRD	12.95	0.05	−12.9	ns	0.42	−10.72	−11.14	ns
SWID/SL	−2.06	3.80	5.86	ns	−8.63	3.80	12.44	*
ST/SL	−15.07	5.63	20.7	*	−18.19	5.93	24.12	*
NS	8.93	62.62	53.69	*	−4.58	35.62	40.21	ns
AS	25.00	−50.00	−75	ns	−25.00	−50.00	−25.00	ns
SD	0.00	−55.71	−55.71	*	−29.58	−62.50	−32.92	**
PL	18.38	−4.21	−22.59	ns	9.02	−13.79	−22.81	ns
RL	8.08	45.51	37.43	**	3.27	34.62	31.35	*
TL	11.37	25.56	14.19	*	5.78	19.46	13.68	ns
PDW	92.16	−0.03	−92.19	ns	61.04	−15.45	−76.48	ns
RDW	84.79	78.85	−5.94	ns	52.00	38.52	−13.48	ns
TDW	87.96	41.68	−46.28	ns	58.18	14.04	−44.14	ns

*=Significant ($P \leq 0.05$), **=Highly significant ($P \leq 0.01$), ns=Not significant. SW=Seed weight, VS=Seed volume, LS=Seed length, WS=Seed width, TS=Seed thickness, RDS=Relative seed diameter, PN=Normal seedlings, PA=Abnormal seedlings, SI=Seeds inert, LP=Plumule length, LR=Radicle length, LT=Total length, PDW=Plumule dry weight, RDW=Radicle dry weight, TDW=Total dry weight, WS/LS=Seed width/Seed length, TS/LS=Seed thick−ness/Seed length.

differences in the variables SW, SL, ST/SL, NS, SD, RL, and TL; while, in HB the significances were for SW, SL, SWID/SL, ST/SL, DS, and LR, *i. e*., both ways of quantifying heterosis coincided in the variables (except NS, SWID/SL, and TL) that revealed the disparity of the heterotic response caused by the already described dissimilar genetic constitution of A and B.

The analysis of the significances (Table 2) also showed that the ST/SL, NS, RL, and TL variables, in A and SWID/SL, ST/SL, and RL, in B; reached positive and significant values, in contrast to the rest of the variables. So these variables better responded to hybridization in the trilinear cross (B), both in H and HB, which had a positive impact on the shape (SWID/SL and ST/SL) and seed germination (NS), as well as in the length growth of the radicle of the produced seedlings.

CONCLUSIONS

In the simple cross, heterosis increased seed dimensions and accumulated biomass in seedlings; while in the trilinear cross, it affected the shape of the seed and the normal seedlings produced.

REFERENCES

1. FAOSTAT (Organización de las Naciones Unidas para la Alimentación y la Agricultura). Available online: https://www.fao.org/faostat/es/#data/QCL (accessed on 3 August 2022).
2. SIAP (Servicio de Información Agroalimentaria y Pesquera). Anuario estadístico de la producción agrícola. Servicio de información agroalimentaria y pesquera, agricultura. Ciudad de México, México. Available online: https://nube.siap.gob.mx/cierreagricola (accessed on 3 August 2022).
3. Arellano-Vázquez, J.L.; Rojas-Martínez, I. 2015. Mayor rendimiento con los nuevos híbridos de maíz azul H-Azul-10, H-Azul-12 y la Variedad Sintética VS-Azul-07 en siembras de temporal del Altiplano Central de México. Desplegable para productores No. 69. Campo Experimental Valle de México CIRCE-INIFAP. México.
4. Arellano-Vázquez, J.L.; Rojas-Martínez, I.; Gutiérrez-Hernández, G.F. 2013. Híbridos y variedades sintéticas de maíz azul para el Altiplano Central de México: Potencial agronómico y estabilidad del rendimiento. 2013. *Rev Mex Cienc Agric* 4. 999-1011.
5. Lee, E.A.; Tollenaar, M. 2007. Physiological basis of successful breeding strategies for maize grain yield. *Crop Sci* 47. S202-S215.
6. Arellano-Vázquez, J.L.; Rojas-Martínez, I.; Gutiérrez-Hernández, G.F. 2014. Variedades de maíz azul Chalqueño seleccionadas por múltiples caracteres y estabilidad del rendimiento. *Rev Mex Cienc Agric* 5. 1469-1480. DOI: 10.29312/remexca.v5i8.828.
7. Arellano-Vázquez, J.L.; Herrera-Zamora, A.; Gutiérrez-Hernández, G.F.; Ceja-Torres, L.F.; Flores-Gómez, E. 2021. Color, contenido de antocianinas y dimensiones de semilla en líneas endogámicas de maíz azul y sus cruzas. *IDESIA* (Chile) 39. 75-82.
8. Shull G.H. 1909. A pure line method of corn breeding. *Am Breeders Assoc Rep* 5. 51-59.
9. MacRobert, J.F.; Sentimela, P.S.; Gethi, J.; Worku, M. 2014. Manual de producción de semilla de maíz híbrido. Centro Internacional de Mejoramiento de Maíz y Trigo. CIMMYT, México, D.F., 37 p. Available online: https://repository.cimmyt.org/bitstream/handle/10883/16849/57179.pdf (accessed on 9 December 2022).
10. Guerrero-Guerrero, C.; Gallegos-Robles, M.A.; Luna Ortega, J.G.; Orona-Castillo, I.; Vázquez-Vázquez, C.; García-Carrillo, M.; Moreno-Resendez, A.; González-Torres A. 2014. Combining ability and heterosis in corn breeding lines to forage and grain. *AJPS* 5. 845-856. DOI: 10.4236/ajps.2014.56098.
11. Gutiérrez-Hernández, G.F.; Ortiz-Hernández, Y.D.; Corzo-Ríos, L.J.; Aquino-Bolaños, T. 2020. Composición química y germinación de semillas de tobalá (*Agave potatorum*). *Interciencia* 45. 223-228.
12. Rocandio-Rodríguez, M.; Santacruz-Varela, A.; Córdova-Téllez, L.; López-Sánchez, H.; Castillo-González, F.; Lobato-Ortiz, R.; García-Zavala, J.J.; Ortega-Paczka, R. 2014. Caracterización morfológica y agronómica de siete razas de maíz de los Valles Altos de México. *Rev Fitotec Mex* 37. 351-361.

13. Carballo-Carballo, A.; Ramírez-Moreno, E.; Coronado-Hernández, R.; Alcázar-Andrade, J.; Simental-Sánchez, F.J.; Martínez-Valdés, G.; Estrada-Gómez, J.A.; Mejía-Andrade, H.; Virgen-Vargas, J.; Espinosa-Calderón, A.; Padilla-Ramírez, R.; Castillo-González, F.; López-Herrera, A.; Pérez-Jerónimo, G. 2014. Guía técnica para la descripción varietal. Secretaría de Agricultura, Ganadería, Desarrollo Rural, Pesca y Alimentación, Servicio Nacional de Inspección y Certificación de Semillas. México. Available online: https://www.gob.mx/cms/uploads/attachment/file/120832/Maiz.pdf (accessed on 3 November 2022).
14. ISTA (International Seed Testing Association). International Rules for Seed Testing. Rules. Available online: http://www.seedtest.org/en/international-rules-_content---1--1083.html (accessed on 19 October 2022).
15. Iqbal, M.; Khan, K.; Rahman, H.; Khalil, I.H.; Sher, H.; Bakht, J. 2010. Heterosis for morphological traits in subtropical maize (*Zea mays* L.). *Maydica* 55. 41-48.
16. Arellano-Vázquez, J.L.; Vázquez-Ramos, J.M.; García-Ramírez, E.; Gómez-y-Gómez, Y.M.; Gutiérrez-Hernández, G.F. 2017. Monitoreo de la calidad proteica de maíz *o2* (*Zea mays* L.) en líneas endogámicas y su progenie F1 y F2. *Agrociencia* 51, 4. 425-436.
17. SAS (Statistical Analysis System). 2002. SAS Inst. Inc. SAS/STAT. Ver. 9. Cary NC, USA.
18. Díaz-Chuquizuta, P.; Hidalgo-Meléndez, E.; Mendoza-Paredes, M.; Cieza-Ruiz, I.; Jara-Calvo, T.W.; Valdés-Rodríguez, O.A. 2023. Nuevo híbrido trilineal de maíz amarillo duro para el trópico peruano. *Agromeso* 34. 51177. DOI:10.15517/am.v34i1.51177
19. Nepir G.; Wegary, D.; Zeleke, H. Heterosis and combining ability of highland quality protein maize inbred lines. 2015. *Maydica* 60. 1-12.
20. Sanghera, G.S.; Wani, S.H.; Hussain, W.; Shafi, W.; Haribhushan, A.; Singh, N.B. 2011. The magic of heterosis: New tools and complexities. *Nat Sci* 9. 42-53