

Yield, biomass and phenology of the cross between wild and domesticated beans

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Abstract

Hybridization makes it possible to improve beans (*Phaseolus vulgaris* L.). Lines with a broad genetic base are an advantage for obtaining new varieties. *P. vulgaris* in Mexico, like corn, is one of the staple crops that fulfills various food functions and is subject to constant improvement. The present study aimed to evaluate the progeny or lines obtained from the cross between a wild and a domesticated parent, grown in greenhouses and hydroponics to determine seed yield, biomass production, and their phenology. The study was carried out in 2019 at the Montecillo Campus, College of Postgraduates. The assessment included wild beans (S13), domesticated beans, registered as Negro Tacaná, and five lines (53b, 51b, 118b, 111, and 3.3) product of the cross of S13 with Negro Tacaná. Each parent and each line was considered as a treatment and were distributed in a completely randomized design. Lines 118b and 3.3 produced the highest seed yields, 225.3 g plant⁻¹ on average; the wild parent, the domesticated one, and lines 11.1 and 51b intermediate yields (150.5 g plant⁻¹), and line 53b the lowest yield 98.6 g plant⁻¹. The S13 parent had the highest number of seeds per pod and weight of one hundred seeds, whereas this was the opposite in Negro Tacaná. The yield and time to occurrence of the phenological stages was a response to the environment and the management of the crop, which allowed the maximum expression of its genetic potential.

Keywords:

Phaseolus vulgaris L., genetic potential, hybridization, hydroponics.

Introduction

Beans (*Phaseolus vulgaris* L.) are the most important edible grain legume for human consumption as they are a source of protein and a relatively high amount of B vitamins, thiamin, riboflavin and minerals (Guzmán-Maldonado *et al.*, 2000). In addition, they are rich in fiber, prebiotics, and various micronutrients (Câmara *et al.*, 2013), which is why they are considered a functional food because they prevent or remedy diseases. In the Americas, the largest producers of beans are Brazil, the United States of America, and Mexico (Beebe *et al.*, 2013; FAO, 2023). In particular, Latin America contributes 32% of the production of this grain (FAO, 2023).

In Mexico, they are grown in almost all regions and soil and climate conditions; Zacatecas and Durango are the main producing states, with an average yield of 0.67 and 0.57 Mg ha⁻¹, respectively; in contrast, in Yucatan, the yield is 0.31 Mg ha⁻¹ (SIACON, 2021). The low yield is due to the fact that most of them are grown in areas with erratic rainfall and frequent periods of drought, soils with low moisture retention capacity, and poor in organic matter and nutrients (Herrera-Flores and Acosta-Gallegos, 2008; Assefa *et al.*, 2013).

To face the low yield of this crop, in addition to improving agronomic management, there are different methods of improvement; hybridization is one of them, which allows obtaining progeny or lines derived from the crossing of parents with outstanding characteristics; for example, higher yield, resistance to drought and diseases, among others. There are insufficient studies on wild bean populations and their parents in aspects of yield components, biomass, and phenology of progeny from wild and domesticated beans (García *et al.*, 1999; Aguirre *et al.*, 2003; Lanna *et al.*, 2018).

Outstanding characteristics (resistant to drought, high temperature, and pathogens) could be found in these populations, which could be used in genetic improvement programs and avoid low yields (Lanna *et al.*, 2018). Plant breeders prefer to use germplasm from lines of the same commercial class to obtain grains with similar attributes, promote mechanization and better yields (Herrera-Flores and Acosta-Gallegos, 2008) and therefore, wild populations or their lines are not used because they are more heterogeneous regarding their agronomic characteristics and are less productive (López *et al.*, 2005).

However, there is a great genetic diversity that has given them resistance to diseases and pests and they have been shown to be tolerant to abiotic stress and with higher nutritional quality (Peña-Valdivia *et al.*, 2011). The hypothesis raised in this study is that the wild beans provide favorable characteristics, such as a greater number of seeds and pods per plant with compared to the lines derived from the cross between them and domesticated beans. The research aimed to evaluate the progeny or lines obtained from the cross between a wild and a domesticated parent, grown in greenhouses and hydroponics to determine seed yield, biomass production and their phenology.

Materials and methods

Experiment location

This research was conducted in a greenhouse with a plastic cover and without temperature control in the spring-summer agricultural cycle of 2019 at the Montecillo Campus, College of Postgraduates, Texcoco, State of Mexico (19° 29' north latitude and 98° 53' west longitude, 2 250 masl). The temperature and relative humidity were recorded with a Hobo U12-012 digital device.

Plant material

The beans used were wild beans (S13) of indeterminate growth type IV, climber, with small seeds and dehiscent pods (Delgado *et al.*, 1988) and the domesticated beans Negro Tacaná (NT) of indeterminate growth type II, shrubby (Rosales *et al.*, 2004). Both materials were crossed and the lines: 11.1, 3.3, 53b, 51b, and 118b were obtained, all of indeterminate growth. The wild beans (S13) were registered at the International Center for Tropical Agriculture (CIAT), for its acronym in Spanish under number G23429, and were originally collected in Santa Isabel, municipality of Cholula, Puebla, Mexico. Cultivated beans (NT) were registered in the Journal of Crop Science (López-Salinas *et al.*, 1997). The seeds were provided by Dr. Jorge A. Acosta Gallegos from INIFAP.

Sowing and management of the parents and lines

The sowing of the parents and lines was carried out in a greenhouse in 2019 in polystyrene cups with a capacity of 250 ml, filled with tezontle of granulometry ≤ 0.5 cm as substrate. Irrigation was with water during the first five days, then it was irrigated with Steiner nutrient solution (Steiner, 1984) with osmotic potential of -0.036 MPa. The seedlings were transplanted when they presented the simple leaves unfolded [10 or 12 days after sowing (das)] in plastic pots of 28.5 cm of top diameter, 26 cm of base diameter, and 35 cm of height and 19 kg of tezontle with a granulometry ≤ 1 cm was added to each one.

After transplantation, it was irrigated every day with nutrient solution with osmotic potential of -0.072 MPa and every seven days with acidulated water with a pH of 5.5. Tap water was used to prepare the Steiner nutrient solution (Steiner, 1984). The pH was adjusted to 5.5 with commercial sulfuric acid, 1N.

Treatments and experimental design

Each of the lines derived from the cross, as well as each of the parents, was considered as a treatment. The experimental unit consisted of one plant per pot. The experimental design was completely randomized, eight replications per treatment were used.

Phenology and growing degree days

Phenology

The occurrence and length of each of the phenological stages in each of the parents and lines was recorded according to the description of the International Center for Tropical Agriculture (Fernández *et al.*, 1991). The days required for each of the phenological stages of the vegetative phase were: sowing (V0), emergence (V1), primary leaves (V2), first compound leaf (V3), third leaf (V4) and for the reproductive phases: pre-flowering (R5), flowering (R6), pod formation (R7), pod filling (R8) and ripening (R9). The vegetative phase runs from the beginning of germination to the moment of differentiation of the floral primordia and the reproductive phase from the appearance of flowers to the day on which the seed completes its development (Fernández *et al.*, 1991).

Growing degree days

To know the heat requirements of the parents and lines studied, the growing degree days (GDDs) (Snyder, 1985) were calculated, and the base temperature (T_b) was 10 °C (Bracho *et al.*, 2010).

Crop variables

Yield, its components, and biomass

When physiological maturity was reached, harvesting was carried out and seed yield (SY, seed weight at 10% moisture, g m^{-2}) and its components were evaluated: number of pods per plant (NPP), number of seeds per pod (NSP), number of seeds per plant (NSPI), and weight of one hundred seeds (WHS).

The total dry biomass (g plant^{-1}) was recorded by drying the plant material to a constant dry weight in an oven (Blue M, Illinois, USA) at 70 °C for 72 h. A Scientech® Series 12000 analytical balance was used for recording biomass. The modified harvest index (HIm, expressed as a percentage) considers the dry matter of the organs that suffered abscission during the crop cycle (Kohashi-Shibata *et al.*, 1980). The total biomass produced per plant was recorded and expressed in dry

weight; this data and seed production were used to determine the modified harvest index (HIm), which represents the proportion of dry matter corresponding to the organ of economic interest (grain) in relation to the total of the plant structures including the fallen organs (Kohashi Shibata *et al.*, 1980) and is considered as an indicator of the efficiency of a variety from the point of view of yield (Escalante and Kohashi, 2015).

Statistical analysis

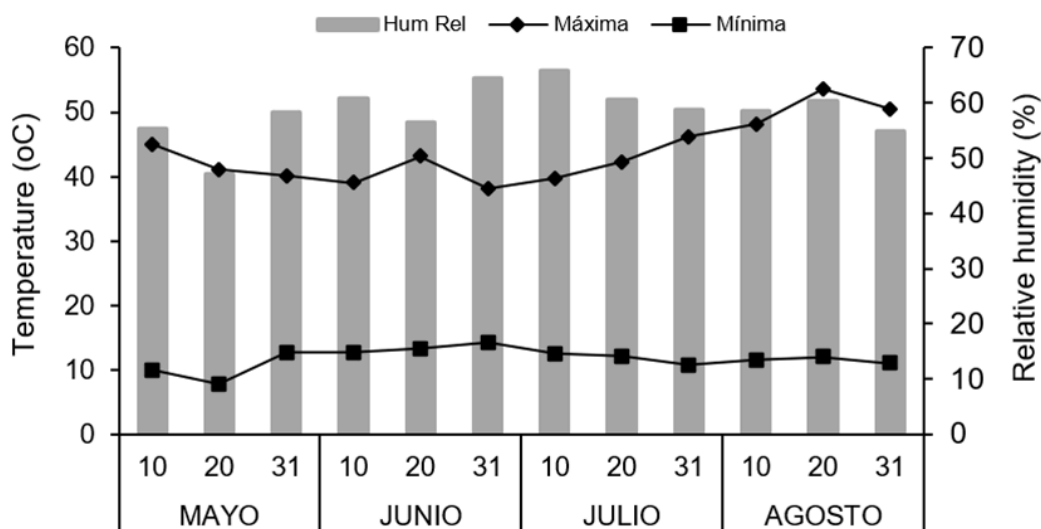
The data of each variable was used to perform the analysis of variance by using the SAS statistical package (SAS Institute Inc., 2012 version 9.3) and the multiple comparison of means was performed using Tukey's test ($p \leq 0.05$).

Results and discussion

Temperature and relative humidity

During the development of the plants in the greenhouse (May-August), the average relative humidity was 58% and the highest was recorded in the first ten days of July (66%), which prevented the development of diseases (Huertas, 2008). The maximum temperature (Tmax) increased significantly from the first ten days of July (40 °C) to the second tend days of August, when the highest temperature (54 °C) was recorded. The lowest temperature occurred in the second tend days of May (8 °C) (Figure 1), the average Tmax during the crop cycle was 43.9 °C and the Tmin 11.7 °C, the minimum temperatures did not damage the leaf blades. The mean temperature was 24 (± 2) °C.

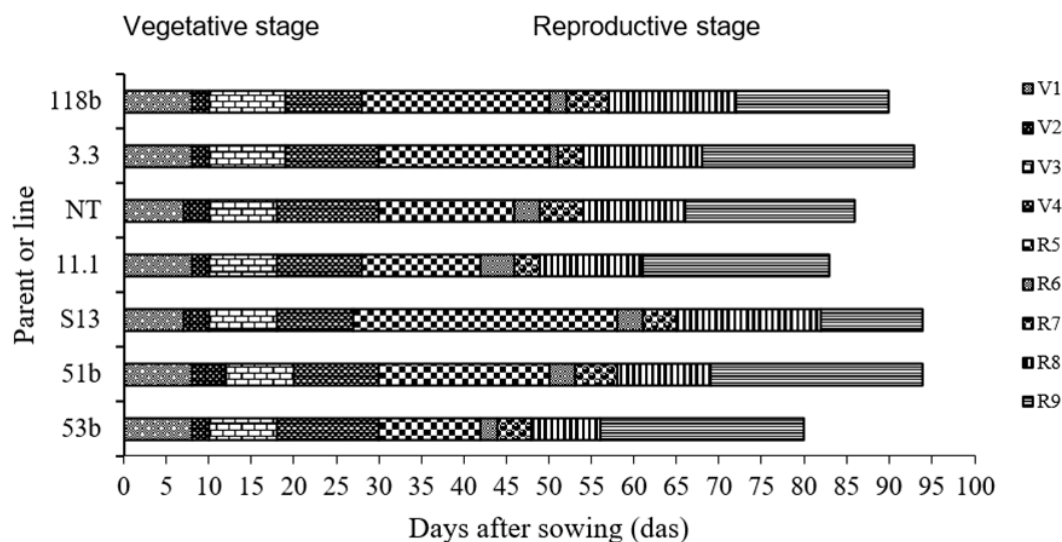
Figure 1. Average minimum and maximum temperature and relative humidity of 10-day periods recorded from May to August 2019.



Phenology and growing degree days

The duration of the biological cycle was different between parents and lines, so the vegetative and reproductive phases were different (Figure 2).

Figure 2. Length of the phenological phases of wild beans (S13), domesticated beans (NT), and the lines derived from the cross between the two, grown in a greenhouse under hydroponics. V1= emergence; V2= primary leaves; V3= first compound leaf; V4= third compound leaf; R5= pre-flowering; R6= flowering; R7= pod formation; R8= pod filling; R9= ripening.



Authors such as Salinas-Ramírez *et al.* (2012) documented similar trends in crops of green beans with different growth habits. In general, seedling emergence occurred between seven and eight das; S13 wild beans and NT domesticated beans presented a faster emergence (seven days) because their imbibition rate was higher than the rest of the lines (García-Urióstegui *et al.*, 2015).

A higher imbibition rate causes rapid changes in enzyme and metabolic activity, and consequently the rupture of the seminal coats (Chong *et al.*, 2002; Doria, 2010). Flores de la Cruz *et al.* (2018) indicated similar values of eight and six das for wild and domesticated beans, respectively. Genetics are inherent to each hybrid as there is a differential behavior in the emergence of each material.

The beginning of the flowering stage (R6) occurred first in 53b and 11.1. In S13, this stage occurred at 61 das. The S13, 118b, 51b, and 3.3 were the latest. Physiological ripening (R9) occurred at 93 das on average. NT, 53b, and 11.1 were earlier, reaching maturity at 82 das on average, whereas 118b, 3.3, S13, and 51b had it at 93 das (Figure 2).

The wild parent (S13) was the latest at R8 (82 das) and also at R9 (94 das), showing a slower ripening compared to the domesticated beans and the lines, which agrees with what was reported by Flores de la Cruz *et al.* (2018), who also cultivated them in a greenhouse, where, for wild beans, the R8 and R9 stages were extended by 12 and 19 das compared to the R8 and R9 stages of the domesticated beans.

This result is similar to that reported by Meza-Vázquez *et al.* (2015) in wild bean populations, and it could be an evolutionary adaptation that allows these plants to behave as biennials or perennials. S13 and 51b required 1 845 GDD, domesticated beans NT 1 668 GDD, whereas 53b needed 1 556 GDD to reach physiological maturity. The other lines required 1 755 (118b), 1 824 (3.3), 1 619 (11.1), 1 556 (53b). These results differ from those obtained by Bracho *et al.* (2010) since 887.9 GDD are needed for optimal growth.

This difference can be explained by the genetic variability that exists between the different genotypes of beans and the interaction with temperature throughout the cycle, as a result of the greenhouse environment that is different from the natural one since temperature influences the

physiological and biochemical processes in the growth and development of plants (Barrios-Gómez and López-Castañeda, 2009). This is confirmed by Maqueira-López *et al.* (2021), who mention that temperature is one of the most important factors to achieve better yields in beans.

Knowledge of the requirements of growing degree days to achieve the different phenological stages allows us, with prior knowledge of temperature, to appropriately schedule agricultural work (Barrios-Gómez and López-Castañeda, 2009; Pichardo-Riego *et al.*, 2013).

Seed yield and its components

The variables of seed yield (SY), number of seeds per plant (NSPI), number of normal pods per plant (NPP), seeds per pod (NSP), and weight of one hundred seeds (WHS) showed highly significant differences ($p \leq 0.05$): S13 presented the highest production of NPP (817) and NSPI (3 513), characteristics that are related to wild plants (Table 1) (Zimdahl, 2007; Ross and Lembi, 2009).

Table 1. Seed yield and its components, dry weight, and modified harvest index in wild beans, domesticated beans, and their lines derived from the cross between both, grown in greenhouse and hydroponics.

Parent or line	SY (g plant ⁻¹)	Pods plant ⁻¹ (NPP)	Seeds plant ⁻¹ (NSPI)	Seeds pod ⁻¹ (NSP)	WHS (g)	† Dry weight (g plant ⁻¹)	HIm (%)
118b 3.3 NT	226.5 a 224.5	296 b 229 bc	1641 b 1526 cb	5.8 abc 6.6 a	13.8 c 14.7 b	489.5 ab 514.4	46.2 a 45.4 a
11.1 S13 51b	a 156.4 b 154.2	103 d 201 c	553 e 1108 cd	5.8 abc 5.5 bc	28.3 a 13.9	a 320.7 c 333.4	48.9 a 47.3 a
53b CV HSD _{0.05}	b 153.6 b 138.1	817 a 192 c	3513 a 974 de	4.4 d 4.9 cd	c 4.4 e 14.2	bc 493.3 ab 311	31.2 a 45.6 a
	bc 98.6 c 18 47	171 cd 19 87	1088 cd 20 466	6.5 ab 19 1	bc 9.1 d 1 1	c 288.4 c 18 168	39.6 a 24 24

Values with different letters within each column are statistically different ($p \leq 0.05$). SY= seed yield; WHS= weight of one hundred seeds; †= dry weight that includes all the aerial part and the fallen organs; HIm= modified harvest index.

Likewise, the indeterminate growth of S13 allows it to have a long reproductive period and at the same time to continue with vegetative growth, both of which are favorable characteristics for the survival of wild populations in the wild (Herrera-Flores and Acosta-Gallegos, 2008). Plants with indeterminate growth habits showed a greater number of nodes and therefore a greater number of potential sites to produce flowers, which increases the number of pods and seeds (Kelly, 2001). Nonetheless, S13 obtained the lowest WHS (4.4 g) and NSP (4.4) and therefore the lowest SY (153.6 g plant) compared to NT and lines, which shows that for NT domesticated beans, the WHS (28.3 g), NSP (5.8), and SY (156.4 g plant⁻¹) were higher (Table 1).

Among the changes that have been identified in domesticated bean genotypes is the increase in some values of anthropogenic importance, such as the size of various plant organs, especially pods and seeds (Peña *et al.*, 2012). In particular, under conditions of water stress, grain yield is closely related to photosynthate remobilization (Padilla-Chacón *et al.*, 2017), which affects the photosynthate partitioning efficiency from vegetative structures to pods and from the pod wall to grain (Sofi and Saba, 2016).

The NT cultivated beans had the lowest NPP (103) and NSPI (553) because they had a short flowering period, together with the selection to which they were subjected when it was sought to reduce the NSPI, but the size of the beans increased and consequently, their yield was intermediate (Table 1); these results agree with what was reported by Flores de la Cruz *et al.* (2018), where the NPP (131) and NSPI (698) for domesticated beans were lower compared to S13 wild beans for NPP (1014) and NSPI (3 067).

However, these results differ from those obtained by García-Nava *et al.* (2014), who show a lower yield in this material, this difference is attributed to the fact that they used pots of a much smaller volume (4.5 kg) and two plants for each experimental unit. The WHS of the NT cultivated beans (28.3 g) was higher than that of S13 (4.4 g), the yields of the lines (13.14 g average) were higher than S13 (Table 1). This is because the cultivated beans were selected based on their weight and not the number of seeds (553) and the wild beans have the characteristic of their smaller seed size but produce a large number of seeds (3 513).

In particular, line 118b obtained high SY ($226.5 \text{ g plant}^{-1}$) while line 53b obtained the lowest value ($98.6 \text{ g plant}^{-1}$). This is due to a low productive potential due to a lower production of flowers, pods, and seeds per plant (Morales-Rosales *et al.*, 2008; Escalante and Kohashi, 2015), whereas these are the most dominant characteristics of the wild parent (Herrera-Flores and Acosta-Gallegos, 2008). Yield depends on genotype, environmental growing conditions, and their interaction (Maqueira-López *et al.*, 2021).

In this case, the experimental environment in which the parents and the lines of the bean plants developed was similar, the crops grew in greenhouses in combination with nutrition by hydroponics, which eliminates or decreases the factors that limit plant growth and production, since there is no competition for nutrients and efficient use of water because there are no losses due to filtration and evaporation (Cánovas, 2001). Therefore, the different responses in yield can be explained by the genotypic variability between parents and lines.

Biomass and modified harvest index

The HIm did not show significant differences between parents and lines, $p \geq 0.05$ (Table 1), presenting an average of 43.5%. The highest biomass corresponded to lines 3.3 and 118b and S13 wild beans, with 499 g plant^{-1} on average, due to the fact that they showed the highest dry weight production in leaf blades and branches, as described by García-Esteva *et al.* (2003). The lowest biomass was found in lines 53b and 51b and the NT parent and it was $306.7 \text{ g plant}^{-1}$ on average, this could be associated with the fact that they showed a shorter growth cycle (Pichardo-Riego *et al.*, 2013).

When considering the SY, biomass, and HIm, the parents and lines can be categorized into three groups according to their statistical differences ($p \leq 0.05$); the first group is made up of 118b and 3.3, with 225 g plant^{-1} on average and an HIm of 45.8%; the second consists of NT, 11.1, 51b, and S13, with $150.5 \text{ g plant}^{-1}$ and 42.5%; and the third includes 53b, with $98.6 \text{ g plant}^{-1}$ and 42.6% on average. This variation among cultivar patterns and the effect that environmental conditions have on them can be directed towards maximizing productivity and selecting the best cultivars for a particular purpose (Núñez *et al.*, 2009).

Conclusions

Lines 118b and 3.3 produced the highest seed yield and biomass and were also the latest. In contrast, the 11.1 had an intermediate yield, and the 53b the lowest, and they were the earliest. Lines 11.1 and 53b produced the lowest biomass. Selection can then be targeted to desirable characteristics in a specific way according to the objectives and interests of the producer.

The study of the morphological characteristics and stages of phenological development of the different genotypes of *Phaseolus* can support the knowledge to improve their yield, conservation, and genetic improvement and identify desirable attributes for agronomic quality to specify the potential of use and the needs of producers.

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