

The heterosis, heterobeltiosis, and the yielding ability of hybrids of three parental crossings of soybean (*Glycine max*)

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Abstract. Hanafiah DS, Siregar LAM, Lubis K, Haryati, Rahmadana HF. 2023. The heterosis, heterobeltiosis, and the yielding ability of hybrids of three parental crossings of soybean (*Glycine max* Merrill). *Biodiversitas* 24: 1032-1038. One of the ways to increase productivity is by assembling new superior cultivars. The base population of a new superior cultivar is first provided as a population with morphological traits correlated with yield. One method for enhancing a plant's characteristics is to cross between parents with various characteristics. This study aimed to examine the value of heterosis and heterobeltiosis of generation F1.1 from a cross of three soybean parents between the Grobogan, Anjasmoro and Dega 1 varieties and predict soybean yields using multiple linear regression models. The soybean varieties Grobogan, Anjasmoro, and Dega 1 were employed as parents in this study. Each variety used as parental material has specific characters. Population diversity of F1.1 with superior traits from the combination of the parents' traits is anticipated to result from the crosses of three soybean varieties as parents. Multiple linear regression models were used in the prediction procedure to determine the soybean yield. The results showed that the value of heterosis and heterobeltiosis on the character of the number of pods per plant and the weight of 100 seeds per plant had positive values, respectively, 26.856%; 5.676%; 16.889% and 5.689%. The result of the best coefficient of determination was 0.6460 in the F1.1 genotype.

Keywords: Heterobeltiosis, heterosis, soybean, three parents cross, yield prediction

INTRODUCTION

Genetic variety is an important aspect in the development of a plant variety. Establishing genetically diverse populations for traits in soybean selection and other plant breeding depends on parent selection (Friedrichs et al. 2016). One of the most popular grain crops in the world is the soybean (Gupta 2015). Soy possesses a variety of beneficial components for human health, including protein, saponins, phytoestrogens, and isoflavones (O'Keefe 2015).

Most soybean varieties were created through mutation, introduction, or selection of local soybean varieties, whereas the majority were created through crossing two or more parents (Krisnawaty and Adi 2022). Anjasmoro, Grobogan, and Dega 1 were the three soybean varieties employed in a crossing program in this study to generate offspring with a variety of genetic make-up. Generally, the more parents involved in the crossing, the more diverse a population will be for the desired traits (Makavu 2018).

Recently, the interaction between parents and crosses of plants has become an interesting topic of discussion (Fujimoto et al. 2018). One of agriculture's most significant genetic contributions is heterosis (Pandini et al. 2002). Heterosis, also called hybrid vigor, is a phenomenon in which heterozygous hybrids perform better than their parents. This phenomenon is very helpful for plants in reproduction and environmental adaptation. Additionally, hybrid breeding is one of the most effective strategies to

improve the grain yield of many crops, so it is very important for agricultural output (Liu et al. 2020).

There are three types of heterosis: best parent, regular variety (check parent), and mid parent (heterobeltiosis). Positive heterosis, or hybrid vigor, is the term used to describe offspring from crossings with traits superior to those of their parents (Ghazy and Fouad 2021). Breeders can select the best cross-combination to use as the basis for selecting superior genotypes by using their knowledge of heterosis (Ahmad 2015). Additionally, understanding the magnitude of heterosis can assist breeders in designing strategies and determining how dominant genes affect the crosses (Pandini et al. 2002).

Most projects for improving soybean varieties attempt to boost crop yield. Therefore, for yield potential, positive heterosis is often preferred (number of pods). In contrast, early maturity (early flowering) traits are more likely to exhibit negative heterosis (Ghazy and Fouad 2021). Therefore, it's crucial to comprehend the diverse gene-based behaviors of each character.

Knowing crop yields before harvest is a helpful way to learn about plant characteristics earlier, allowing breeders to choose superior plants. This issue can be resolved by predicting crop yields. Based on knowledge from the past and the present, prediction is the systematic estimation of what will occur in the future (Matondang 2018). For instance, estimating the quantity of soybean pods can help anticipate the early production potential of soybean plants.

According to Rasyid (2013), soybean plants with a good plant height will increase the number of primary branches and nodes, which might increase the number of pods per plant and surely affect the yield component of the plant.

Compared to other prediction techniques, regression-based models have been utilized in agricultural projects more frequently (Abdipour et al. 2016). Multiple linear regression, which seeks to model the link between two or more explanatory variables and a dependent variable by assuming a linear relationship, is based on the linear and additive associations of the explanatory variables (Darlington and Hayes 2016). According to the research findings by Herwanto et al. (2019), multiple linear regression is a predictive or forecasting model in which more than one predictor can be used to predict rice yields. The multiple linear regression model has a 94.51% fit based on the quantity of available data (Herwanto et al. 2019). This study aimed to examine the value of heterosis and heterobeltiosis of F1.1 generation from a three-way crosses involving three soybean parents, Grobogan, Anjasmoro and Dega varieties, and predict soybean yields (Grobogan, Anjasmoro, Dega 1 and F1.1) using multiple linear regression models.

MATERIALS AND METHODS

The study site and time

This research was carried out in the farmer's field in Emplasmen Kwala Mencirim Village, Sei Binjai District, Langkat Regency, from May to - August 2022.

Research materials and tools

The materials used in this study were seeds of three soybean varieties (Grobogan, Anjasmoro, Dega 1) and the F1.1 [(Grobogan x Anjasmoro) x Dega 1], urea, TSP and KCl fertilizers, fungicides (Dithane M-45 active ingredient mancozeb) and insecticides (Decis active ingredient deltamethrin).

The tools used in this study were a hoe, cord, tape measure, tray, scissors, analytical scale, sprayer, laptop, camera, and stationery to make observations.

Observation

Agronomic character data were obtained by observing plant height (cm), number of primary branches, age of flowering (days), age of harvest (days), number of pods per plant, number of filled pods per plant, one-seeded pods, two-seeded pods, three-seeded pods, four-seeded pods, number of empty pods per plant, number of seeds per plant, seed weight per plant (gram) and 100-seeds weight (gram).

Procedures

This research was conducted by planting and observing each plant generation F1.1 and its three parents in the same planting environment. The Grobogan, Anjasmoro and Dega varieties were each planted with 32 plants, and F1.1 was as many as 24 plants (based on the availability of existing seeds). Soybean seeds were planted in beds measuring 100 cm x 140 cm, and the distance between the beds was 50

cm. Planting was done by making a planting hole as deep as ± 2 cm and a spacing of 20 cm x 40 cm; one seed was planted per planting hole.

The performance of the F1.1 offspring to the parents was heterosis.

$$\text{Heterosis (\%)} = [(H - MP) / MP] \times 100$$

Where:

H : population performance F1.1 (cross three parents)

MP : average performance of the parents.

The MP for a cross of three parents is measured as $MP = \{[(T1 + T2)/2] + T3\}/2$.

The best parental performance was measured as heterobeltiosis

$$\text{Heterobeltiosis (\%)} = [(H1 - HP) / HP] \times 100$$

Where:

H1 : the performance of the derivative of F1.1 and

HP : performance of the best parent

Analysis and Design in Predicting Growth

The multiple linear regression method was implemented to predict the number of pods per plant using Microsoft Excel. The data needed for the prediction process are plant height at flowering, number of primary branches and flowering age. After all the data has been collected, the next step is to predict the number of soybean pods by applying multiple linear regression. Multiple linear regression looks for the relationship between one dependent variable (Y) and two or more independent variables (X) (Babatunde and Abbasoglu 2019). Therefore, before entering the number of pods per plant prediction stage, what needs to be done is *modeling* or forming a multiple linear regression equation model first by searching for the values of a (constant), b1 (coefficient of plant height at flowering), and b2 (coefficient of the number of primary branches) and b3 (coefficient of flowering age).

Prediction of crop prediction was calculated using the formula:

$$Y = a + b1X1 + b2X2 + b3X3$$

Where:

Y : number of soybean pods

A : constant value

b1 : plant height coefficient

X1 : plant height

b2 : coefficient of the number of primary branches

X2 : number of primary branches

b3 : coefficient of flowering date

X3 : flowering date

The model performance to predict desired output was determined using three statistical quality parameters, i.e., mean absolute error (MAE), root mean square error (RMSE), and coefficient of determination (R²).

RESULTS AND DISCUSSION

Table 1 shows that F1.1 offsprings of a cross between three parents had desirable traits such as a short flowering age (26.58 days), which can accelerate the time of harvest, higher seed weight per plant (17.62 g) and the higher 100 seeds weight (22 g) compared to the parents. At the same time, the plant height of F1.1 (31.58 cm) was undesirable because it was shorter than the optimal/desirable height. Soybean plants are expected to have a height that allows them to produce productive primary branches so that the number of pods can increase.

The mean agronomic performances of the observed character of soybean varieties Grobogan, Anjasmoro, Dega and the F1.1 are presented in Table 1.

The number of primary branches per plant of the F1.1 genotype was similar to that of the Grobogan variety, i.e., 2.72, while those of the Anjasmoro and Dega 1 varieties were 3.22 and 2.00 branches. The number of branches contributes to seed yield (Kuswanto et al. 2014). Furthermore, the formation of branches in soybean plants depends on the plant growth type: determinate or indeterminate. When the flowering phase begins, the branches of an indeterminate growth type will continue to expand, but the branches of a determinate growth type will stop growing when the flowers appear (Agudamu et al. 2016). The age of the flowering character in the F1.1 genotype was shorter than that of the three parent varieties (Grobogan, Anjasmoro and Dega 1). The flowering date of the F1.1 genotype was 26.58 days. The date of flowering is closely related to the soybean harvesting date. Therefore, breeders usually select a shorter harvesting date as a desirable trait for superior varieties.

The seed weight per plant of F1.1 was higher than the three parents. The seed weight of the F1.1 genotype was 17.62 g, while the Grobogan, Anjasmoro and Dega 1 varieties were 15.37 g, 16.67 g and 11.75. The same is true of the 100-seed weight; the F1.1 genotype had a higher value (22.0) than its three parents, Grobogan, Anjasmoro

and Dega 1, respectively, 20.35 g; 13.31 g and 20.82 g. The 100-seed weight measures the size of the seeds. The large seed size in the cross of three parents was most likely derived from the large seed of the Dega 1 variety, with a 100-seed weight of 20.82 g. 100-seed weight was also positively correlated with seed yield per plant and seed weight per plant (Faot et al. 2019). The performance of F1.1 plants derived from a cross of three parents, the values of heterosis and heterobeltiosis are shown in Table 2.

Table 2 shows that the highest F1.1 heterosis value was found in the four-seeded pods (522.222%), seed weight per plant (26.856%) and one-seeded pods 1 (17.012%), the weight of 100 seeds (16.889%) and the number of primary branches per plant (9.457%), and the lowest was in the plant height trait (-10.430). The highest value of heterobeltiosis F1.1 was found in the four-seeded pods (133.333%), 100-seed weight (5.689%) and seed weight per plant (5.676%) and the lowest was on the number of seeds per plant (-39.833%). The heterosis value will explain the best F1.1 generation offspring compared to the best parent in the attribute you want to observe. The heterobeltiosis value will explain the best offspring compared to the best parent overall (Khuwaja et al. 2022).

In the number of primary branches trait, heterosis was positive, and heterobeltiosis was negative, with a value of 9.457% and -15.528%. Yamgar et al. (2021) found that the highest values of heterosis and heterobeltiosis in the F1.1 generation were 32.20% and 6.85%, respectively. A high heterosis value indicates that hybridization will effectively produce superior varieties. Meanwhile, the characters of the age of flowering and age of harvest have negative heterosis and heterobeltiosis values of -8.011%, -1.315, and -4.418, except heterobeltiosis values on the age of harvest character. This indicates that the F1.1 generation is not better than its parents in the flowering age. But the heterobeltiosis value on the age of harvesting character is positive (1.385%), which means that the age harvest of the F1.1 generation is better than the best parents.

Table 1. Mean agronomic performance of the observed characters

Character	Mean			
	Grobogan	Anjasmoro	Dega	F1.1
Plant height (cm)	32.77	49.89	29.17	31.58
Number of primary branches	2.72	3.22	2.00	2.72
Age of flowering (days)	27.03	34.69	26.94	26.58
Age of harvesting (days)	79.19	94.56	77.47	78.54
Number of pods per plant	40.00	63.56	30.31	41.71
Number of filled pods per plant	38.47	60.72	29.66	39.92
One-seeded pods	9.50	9.19	7.25	9.71
Two-seeded pods	20.81	32.28	17.28	20.75
Three-seeded pods	8.03	19.19	5.16	9.17
Four-seeded pods	0.13	0.06	0.00	0.29
Number of empty pods per plant	1.53	2.84	0.63	1.79
Number of seeds per plant	76.22	130.13	56.19	78.29
Seed weight per plant (gram)	15.37	16.67	11.75	17.62
100-seed weight (gram)	20.35	13.31	20.82	22.00

Note: one-seeded pod means only one seed chamber in each pod, two-seeded pod means two seed chambers per pod, three-seeded pod means three seed chambers in each pod, and four-seeded pod means only four seed chambers in each pod

Table 2. Value of heterosis and heterobeltiosis of F1.1 derived from three-way crosses involving three parents (Grobogan, Anjasmoro and Dega 1)

Character	Heterosis	Heterobeltiosis
Plant height (cm)	-10.430	-36.712
Number of primary branches	9.457	-15.528
Age of flowering (days)	-8.011	-1.315
Age of harvest (days)	-4.418	1.385
Number of pods per plant	1.611	-34.382
Number of filled pods per plant	0.736	-34.260
One-seeded pods	17.012	2.193
Two-seeded pods	-5.312	-35.721
Three-seeded pods	-2.304	-52.226
Four-seeded pods	522.222	133.333
Number of empty pods per plant	27.407	-36.9963
Number of seeds per plant	-1.742	-39.833
Seed weight per plant (gram)	26.856	5.676
100-seed weight (gram)	16.889	5.689

Note: one-seeded pod means only one seed chamber in each pod, two-seeded pod means two seed chambers per pod, three-seeded pod means three seed chambers in each pod, and four-seeded pod means only four seed chambers in each pod

The quality and capacity of the pollen to fertilize the ovaries significantly impact the number of pods in soybean crosses. The amount of pollen that settles on the stigma affects the outcome of a cross and the development of pods (Jumrani et al. 2018). The effects of heterosis and heterobeltiosis on the characteristics of the number of pods per plant in this study were 1.611% and -34.382%. According to Sen (2020), heterosis and heterobeltiosis in several soybean crosses ranged from 2.59% to 18.13%, a very substantial proportion.

The values of heterosis and heterobeltiosis in F1.1 were positive for seed weight per plant and 100-seed weight, respectively, 26.856%, 5.676%, 16.889% and 5.689%. This shows that the offspring of this cross produces better in weight per plant and 100-seed weight than either of its three parents or the best parent. Due to the high levels of heterosis and heterobeltiosis in the observed features, the tested genotypes had a greater value than the average of the two parents and their best parents (Meena et al. 2016). Heterosis is brought about by the superior, dominant genes accumulated in one F1.1 genotype produced from a cross between the parents (Kong et al. 2020). Heterozygosity, maternal relationships, non-allelic interaction or epistasis, allelic interaction, and dominance or overdominance are some factors that impact the manifestation of heterosis (Kaur et al. 2017). The basic requisite in hybrid variety development is the identification of crosses with a high degree of exploitable heterosis (Rohini and Lakshmanan 2016).

The estimated value of heterosis and heterobeltiosis generally differs between individuals and parental pair crosses (Ibrahim et al. 2021; Ismail et al. 2020). Gene complementation and the number of dominant alleles were hypothesized as potential genetic underpinnings for heterosis in soybean by Burton and Brownie (2006).

In agriculture, regression models are frequently used for various objectives, including crop production prediction based on cultivation technology, soil characteristics, and water use efficiency (Williams 2017). It should be noted that linear regression produces adequately precise predictions (Lykhovyd 2018). The following are the search results for the constants and coefficients of each variable to predict the number of pods per plant.

After getting the values of a (constant), b1 (coefficient of plant height at flowering), b2 (coefficient of the number of primary branches), and b3 (coefficient of flowering date) from the implementation of the formation of multiple linear regression models, the next step is to calculate the predicted value using the formula provided on research methods (Table 3). The following are the search results for the predicted value of the number of pods per plant compared with data on the number of pods planted in the field (observed) (Table 4).

After the predicted number of pods per plant was obtained, the next step was to test the model's accuracy. The accuracy of this model is tested by calculating the mean square error (MSE), root means square error (RMSE), and coefficient of determination (R^2), as presented in Table 5.

Table 5 shows that the coefficient of determination values differed among the genotypes. The highest coefficient of determination (R^2) was in the F1.1 genotype (0.6460), meaning that the level of fit for the multiple linear regression model has a reliability level of 64.60%. In comparison, the lowest coefficient of determination was in Dega 1 genotype (0.2766), which means that the level of compatibility of the multiple linear regression model has a reliability level of 26.66%. The coefficient of determination values ranges from 0 to 1. The value of $R^2 = 0$ indicates that there is no influence between the independent variable and the dependent variable. If the R^2 is close to 1, the influence of the independent variable is stronger on the dependent variable (Herwanto et al. 2019). In this case, the variables of plant height at flowering, the number of primary branches per plant and age of flowering in the F1.1 genotype had an effect of 0.6460 or 64.60% in influencing the number of pods per plant. The value is close to 1 compared to the others. A modeling study by Abdipour et al. (2019) concluded that the seed yield of safflower has an R^2 value of 0.887 or 88.7%.

Table 3. The results of the calculated values a; b1; b2; and b3

Genotype	Constant value or intercept (a)	Coefficient of plant height at flowering (b1)	Coefficient of the number of primary branches (b2)	Coefficient of flowering date (b3)
Grobogan	-117.3021	0.7126	10.9261	4.1718
Anjasmoro	-54.3738	2.0856	2.0248	0.9511
Dega 1	-3.6950	1.6595	1.7811	-0.1188
F1.1	2.9330	0.2074	15.1736	-0.3973

Table 4. The correlation between the results of predicted and observed value in the field for the number of pods per plant

Sample	Grobogan		Anjasmoro		Dega 1		F1.1	
	Observed	Predicted	Observed	Predicted	Observed	Predicted	Observed	Predicted
1	23	43.65	44	44.58	34	31.25	50	50.06
2	49	44.15	76	61.26	37	37.06	15	34.68
3	45	48.21	25	40.94	36	38.72	15	34.71
4	56	41.40	48	45.91	39	33.91	13	19.29
5	37	38.15	55	58.76	14	18.78	46	51.41
6	28	46.90	79	83.10	39	35.66	58	50.89
7	48	49.33	79	47.71	40	37.35	38	50.48
8	34	44.19	41	48.75	41	30.97	55	50.60
9	24	37.48	53	46.06	32	29.97	46	51.41
10	31	38.11	26	38.32	27	29.90	52	65.83
11	34	41.51	93	90.40	30	34.24	71	66.31
12	46	44.08	62	81.02	33	34.12	45	33.99
13	10	20.66	74	69.40	31	28.69	56	65.28
14	8	13.30	47	44.58	13	19.98	78	81.65
15	43	44.58	45	43.29	48	36.44	34	49.02
16	55	54.01	78	81.02	19	25.78	61	49.09
17	40	54.25	62	34.30	38	33.53	33	49.89
18	21	29.73	52	47.05	17	28.19	27	34.37
19	31	42.87	103	90.76	31	35.49	26	34.92
20	38	34.58	100	78.99	19	26.82	34	34.06
21	67	47.39	21	41.18	32	32.22	25	35.23
22	49	39.69	53	49.53	35	30.47	43	65.40
23	49	30.44	73	75.80	31	32.29	50	50.25
24	47	50.62	122	93.41	30	41.33	30	34.96
25	61	36.58	44	62.60	26	22.14		
26	65	53.04	72	72.20	42	27.94		
27	56	38.91	91	86.23	20	30.80		
28	37	31.16	40	76.64	46	24.04		
29	64	51.83	32	38.26	18	26.28		
30	30	38.41	82	86.44	21	26.98		
31	15	12.37	77	90.76	30	25.58		
32	39	38.41	85	84.65	21	23.10		

Table 5. Accuracy of the test results for each genotype in the number of pods per plant

Genotype	RMSE	MSE	R ²
Grobogan	11.3564	128.9692	0.3900
Anjasmoro	14.5137	210.6488	0.6038
Dega 1	7.4262	55.1493	0.2766
F1.1	11.0889	122.9645	0.6460

Note: RMSE: root mean square error; MSE: mean square error; R²: coefficient of determination

Table 6. The correlation between the results of predicted and observed value in the field for yield potential

Genotype	Observed	Predicted	RMSE	MSE	R ²
Grobogan	1.921	1.938	0.0197	0.0004	1.000
Anjasmoro	2.084	2.112			
Dega 1	1.469	1.484			
F1.1	2.202	2.218			

Note: RMSE: root mean square error; MSE: mean square error; R²: coefficient of determination

Root mean square error (RMSE) is a method for evaluating the results of forecasting techniques used to measure the level of accuracy of the forecast results of a model (Chai and Draxler 2014). RMSE and mean square error (MSE) are closely related. The RMSE value results from the calculation of the square root of the MSE value (Chicco 2021). The results of this study showed that Dega

1 has the lowest RMSE value (7.4262) while Anjasmoro has the highest RMSE value (14.5137). A low RMSE value indicates a good level of prediction accuracy (Snyder 1999). The prediction of the number of pods in Dega 1 was better compared to the other three genotypes.

The yield potential was a yield of a cultivar when cultivated in surroundings to which it is acclimated, with

nutrients and water not being limited, and with pests, diseases, weeds, lodging, and other stresses being well managed (Huang et al. 2017). Soybean yield potential is determined by the area of 1 hectare (ha) divided by the spacing and multiplied by the average seed weight per plant. The study results showed that the yield potential (Table 6) of the three soybean parents Grobogan, Anjasmoro and Dega 1 ($t\ ha^{-1}$), respectively, was 1.921, 2.084, and 1.469. These potential yields are below the average yield potential ($t\ ha^{-1}$) of Grobogan, Anjasmoro and Dega 1, respectively, was 3.4, 2.25 and 3.82 (Balitkabi 2016). Changes in the weather during the planting season may cause the decreased yield potential observed in this study. According to Veas (2022), high temperatures and water stress will decrease yield elements, including the quantity and weight of seeds. Reducing the weight of soybean seeds under conditions of high temperature and lack of water will lead to a decrease in CO_2 fixation sites which involves a reduction in leaf thickness and area (Carrera et al. 2021), along with damage to the photosynthetic machine (Ergo et al. 2018). At the same planting site and environmental conditions, the crosses of the three parents, known as F1.1, had a higher yield potential ($2.202\ t\ ha^{-1}$) than the three parents. This demonstrates that F1.1's outcomes are superior to those of their parents.

Predictions of yield potential have been carried out using independent variables, i.e., plant height, number of primary branches and flowering age. In addition, the sample genotypes were used to forecast yield potential because crossing three parents resulted in only a few number seeds. The results of potential yield predictions are shown in Table 6.

Table 6 shows that the results of the potential yield predictions that have been carried out are close to the actual values in the field. A low RMSE value indicates this, and it was 0.0197. A low RMSE value indicates a good level of prediction accuracy (Snyder 1999). A predictive model with lower RMSE values performs better (Moayedi et al. 2019). Besides that, output estimates in the first generation F1.1 crossbreeding study of three individuals in the population were dispersed and still unstable; predicting the yield potential is a little challenging. The segregation will occur in the first generations of crossed plants (Kuckuck 2020). Environmental conditions also influence yield potential. Since yield potentials are dependent on nearly perfect crop management (e.g., planting date, plant population, optimal nutrition for all essential nutrients, elimination of biotic and abiotic stresses), the only environmental factors that can limit productivity are solar radiation, temperature, and reduced vapor pressure (Lobell et al. 2009).

In conclusion, based on the analysis of heterosis and heterobeltiosis, F1.1 has a character superior to its parents in the character of seed weight per plant and 100 seeds weight per plant. The best prediction accuracy for the number of pods planted was found in the Dega 1 variety, which had the lowest RMSE value of 7.4262. The accuracy of the prediction results will be good if it has more samples; therefore, more samples are needed to make

predictions. In addition, further research is still needed on the F2 generation of a cross of three parents of soybean to obtain the desired traits and characters.

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