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Genetic inheritance of leaf and pod traits in blackgram [*Vigna mungo* (L.) Hepper]

Surendhar Anbazhagan * and Jayamani Palaniyappan

Department of Pulses, Tamil Nadu Agricultural University, Coimbatore- 641003, India.

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Abstract

An investigation was carried out on blackgram to examine inheritance pattern of the traits *viz.,* terminal leaflet shape, leaf vein colour, immature pod colour and pod pubescence. Five crosses *viz.,* CO 5 × VBN 9, CO 5 × VBN 10, CO 6 × VBN 9, CO 6 × VBN 10 and CO 6 × VBN 11 were made for studying the genetics of the traits. The qualitative traits were examined in the F_1 plants, F_2 population and F_3 progenies, and a chi-square test was performed to assess their fit to the expected segregation ratio. In the crosses CO 5 × VBN 10 and CO 6 × VBN 10, the terminal leaflet shape exhibited a segregation ratio of 3:1 in F2 and 1:2:1 in F3, with lanceolate shape being dominant over ovate shape. Purple pigmentation in leaf veins and immature pods was dominant in CO 5 × VBN 9 and CO 6 × VBN 9 crosses and co-segregated as a monogenic trait. In the crosses CO 6 × VBN 9, CO 6 × VBN 10 and CO 6 × VBN 11, hairy pods were dominant over glabrous pods and controlled by a single dominant gene. Thus, these traits *viz.,* terminal leaflet shape, leaf vein colour, immature pod colour and pod pubescence, could serve as useful morphological markers for germplasm characterization and confirming true hybrids in recombination breeding programs.

Keywords: Blackgram; Leaflet shape; Purple pigmentation; Pod pubescence; Inheritance and pleiotropy

1. Introduction

Blackgram, scientifically known as *Vigna mungo* (L.) Hepper is a highly self-pollinated diploid crop (Arumuganathan and Earle, 1991), rich in protein (25 %) and other essential nutrients (USDA, 2018). It can fit well in various cropping systems, especially in rice and wheat fallows due to its short life cycle (60-90 days), ability to fix atmospheric nitrogen and relative drought tolerance (Gomathi *et al.*, 2023). Typically, blackgram is intercropped with maize, sorghum, cotton, millets and pigeonpea, or it is cultivated in rotation with cereal crops. India's production stands at 2.77 million tonnes from an area of 4.63 million hectares, with productivity of 599 kg/ha (Indiastat, 2023). Traditionally, 60 per cent of the crop area is dedicated to *kharif* season cultivation, but there is a growing trend towards *rabi* season cultivation. This shift is driven by the adoption of early maturing varieties, particularly in rice fallows, with a maturity period of 60-70 days. The qualitative traits are governed by one or few genes, polymorphic, easily measurable and less accountable to environmental influences compared to quantitative traits (Ghafoor *et al.*, 2003). These traits serve as morphological markers for identifying varieties and characterizing germplasm in plant breeding. Additionally, these traits play a role in verifying hybrids resulting from crosses between parents with distinct traits. For qualitative traits to serve as effective morphological markers in hybridization programs, their inheritance pattern need to be clearly understood (Arshad *et al.*, 2005). In this study, the inheritance pattern of qualitative traits *viz.,* terminal leaflet shape, leaf vein colour, immature pod colour and pod pubescence were examined in various inter-varietal crosses of blackgram.

* Corresponding author: Surendhar Anbazhagan

2. Material and methods

In blackgram, five crosses *viz.,* CO 5 × VBN 9, CO 5 × VBN 10, CO 6 × VBN 9, CO 6 × VBN 10 and CO 6 × VBN 11 were made by hand emasculation and crossing following the method of Ghafoor *et al.* (1999) during summer, 2022. These crosses involved varieties with distinct morphological characters, as mentioned in Table 1. The resulting F_1 plants were evaluated during *kharif,* 2022. Subsequently, the true F_1 plants were advanced to the F_2 generation during *rabi,* 2022-23 and the plants were harvested individually. The seeds obtained from each F_2 plant were sown as progeny rows (F_3) of 4 m length during summer, 2023. In all the generations, the recommended spacing of 30 × 10 cm and good agronomic and plant protection measures were followed to grow a healthy crop. Morphological traits *viz.,* terminal leaflet shape, leaf vein colour, immature pod colour and pod pubescence, were observed following the guidelines of Descriptor (2007) across all generations of the crosses. Terminal leaflet shape and leaf vein colour were visually assessed at the 50 per cent flowering stage, while immature pod colour was recorded at the immature pod stage and pod pubescence was noted from fully developed green pods. Data from parents and hybrids were collected by visual observation of 10 plants (Descriptor, 2007). All plants in the F_2 generation were assessed for qualitative traits. In the F_3 generation, progenies were noted as either segregating or non-segregating for the respective traits. The number of F_2 plants and F_3 progenies in each cross are detailed in Table 1. Data from the F_2 population and F_3 progenies underwent chi-square testing to assess goodness of fit with the expected segregation ratio (Pearson, 1900).

3. Results and discussion

3.1. Inheritance of terminal leaflet shape

The inheritance pattern of terminal leaflet shape was examined in two crosses, namely CO 5 × VBN 10 and CO 6 × VBN 10, where the female parents exhibited ovate shape, while the male parent displayed a lanceolate shape. The F_1 plants from both crosses produced lanceolate leaves (Table 2), indicating the dominance nature of the lanceolate shape over the ovate shape. Similarly, Jayashree *et al.* (2020) have reported the dominant nature of lanceolate shape in blackgram. In the F_2 generation of the above two crosses (comprising of 95 and 84 plants, respectively), a typical 3:1 segregation was observed for terminal leaflet shape (Lanceolate: Ovate) with probability level ranged from 75-80 per cent in CO 5 × VBN 10 and 80-85 per cent in CO 6 × VBN 10. This observation suggests that a monogenic dominant gene involved in the expression of the trait. In the F_3 progenies of both crosses, the terminal leaflet shape segregated in a 1:2:1 ratio (Lanceolate: Segregating: Ovate), with a probability ranged from 85 to 90 per cent in both the crosses. This observation provides further evidence for the monogenic dominant nature of the trait. The reports of Gupta *et al.* (2022) and Nair *et al.* (2024) supporting the monogenic control of terminal leaflet shape in blackgram. Consequently, the trait terminal leaflet shape was determined by a single gene, with the lanceolate shape being dominant over the ovate shape.

3.2. Inheritance of leaf vein and immature pod colour

Purple pigmentation was observed on the leaf veins and immature pods of the male parent (VBN 9) in two crosses *viz.,* CO 5 × VBN 9 and CO 6 × VBN 9, while the female parents exhibited green type. All F_1 plants from both crosses displayed purple pigmentation on leaf veins and immature pods (Table 3). A total of 95 and 64 F_2 plants were obtained from the crosses *viz.,* CO 5 × VBN 9 and CO 6 × VBN 9, respectively. In both crosses, three-quarters of the F_2 plants expressed purple pigmentation on leaf veins and immature pods, while the remaining quarter had green coloured leaf veins and immature pods. The phenotypic segregation followed a 3:1 ratio with a high probability level (P = 90-95 %). Further, co-segregation of purple pigmentation on leaf veins and immature pods was observed in both crosses. This purple pigmentation in more than one part is due to the pleiotropic effect of the single dominant gene '*Ppp1*' in blackgram (Dwivedi and Singh, 1985). In the present study, the monogenic control of this trait was observed with a pleiotropic effect purple pigmentation on leaf veins and immature pods. In the F_3 progenies derived from purple type plants (3/4), no segregation was observed in one quarter (purple type), while segregation was observed in the other two quarters. F_3 progenies from green type plants (1/4) produced only green coloured leaf veins and immature pods. A typical 1:2:1 ratio was observed for this segregation type in the crosses *viz.,* CO 5 × VBN 9 (P = 85-90 %) and CO 6 × VBN 9 (P = 90-95 %). Nair *et al.* (2024) summarized the genetics of purple pigmentation that controlled by single gene with pleiotropic effect. Thus, this purple pigmentation could be used as a morphological marker to fix the hybrids in the breeding program.

3.3. Inheritance of pod pubescence

The hairy pods were observed in the male parents of three crosses *viz.,* CO 6 × VBN 9, CO 6 × VBN 10 and CO 6 × VBN 11, whereas the female parent CO 6 produced glabrous pods (Table 4). All F_1 plants from the above crosses exhibited hairy pods, indicating the dominant nature of hairiness over the glabrous pods. A total of 85, 62 and 105 F_2 plants were documented for pod hairiness in the CO 6 × VBN 9, CO 6 × VBN 10 and CO 6 × VBN 11 crosses, respectively. Across all these crosses, three-quarters of the plants displayed hairy pods, while one-quarter produced glabrous pods, aligning with the expected 3:1 ratio typical of monogenic inheritance. The probability levels for this inheritance pattern were estimated at 85-90 per cent for CO 6 × VBN 9 and CO 6 × VBN 10 and 75-80 per cent for CO 6 × VBN 11. The earlier reports by Singh (1961), Arshad *et al.* (2005) and Sirohi and Singh (1998) also confirmed the monogenic dominant nature of hairiness on pods in blackgram. Upon analysing the F_3 progenies, it was observed that among the F_2 plants with hairy pods (three parts), one-third gave rise to non-segregating F_3 progenies with hairy pods, while two-third yielded segregating F_3 progenies concerning pod hairiness. Conversely, the remaining F_3 progenies (one part) with glabrous pods showed consistent non-segregation across all crosses. Segregation in F_3 for pod hairiness followed a 1:2:1 ratio in the crosses *viz.,* CO 6 × VBN 9 (P = 90-95 %), CO 6 × VBN 10 (P = 85-90 %) and CO 6 × VBN 11 (P = 85-90 %). Thus, the expression of pod pubescence was governed by a single gene, with hairy pods exerting dominant over glabrous ones.

Table 1 Details of crosses and qualitative traits used for the inheritance study

Cross	Qualitative traits	Female parent	Male parent	Number of F_2 and F_3 progenies
CO 5 × VBN 9	Leaf vein colour	Green	Purple	126
	Immature pod colour	Green	Purple	
CO 5 × VBN 10	Terminal leaflet shape	Ovate	Lanceolate	95
CO 6 × VBN 9	Leaf vein colour	Green	Purple	85
	Immature pod colour	Green	Purple	
	Pod pubescence	Glabrous	Hairy	
CO 6 × VBN 10	Terminal leaflet shape	Ovate	Lanceolate	84
	Pod pubescence	Glabrous	Hairy	
CO 6 × VBN 11	Pod pubescence	Glabrous	Hairy	105

Table 2 Inheritance pattern of terminal leaflet shape in various generations of two crosses

Generation	Phenotypic segregation for terminal leaflet shape			Total number of plants	Expected ratio	χ^2 value	Probability range (%)
	Lanceolate	Segregating	Ovate				
CO 5 × VBN 10							
F_1	35	-	-	35	-	-	-
F_2	70	-	25	95	3:1	0.087	75-80
F_3	25	45	25	95	1:2:1	0.263	85-90
CO 6 × VBN 10							
F_1	45	-	-	45	-	-	-
F_2	64	-	20	84	3:1	0.063	80-85
F_3	19	44	20	84	1:2:1	0.325	85-90

Table 3 Inheritance pattern of leaf vein and immature pod colour in various generations of two crosses

Generation	Phenotypic segregation for leaf vein and immature pod colour			Total number of plants	Expected ratio	χ^2 value	Probability range (%)
	Purple	Segregating	Green				
CO 5 × VBN 9							
F_1	48	-	-	48	-	-	-
F_2	95	-	31	126	3:1	0.010	90-95
F_3	34	61	31	126	1:2:1	0.269	85-90
CO 6 × VBN 9							
F_1	40	-	-	40	-	-	-
F_2	64	-	21	85	3:1	0.003	90-95
F_3	23	41	21	85	1:2:1	0.200	90-95

Table 4 Inheritance pattern of pod pubescence in various generations of three crosses

Generation	Phenotypic segregation for pod pubescence			Total number of plants	Expected ratio	χ^2 value	Probability range (%)
	Hairy	Segregating	Glabrous				
CO 6 × VBN 9							
F_1	40	-	-	40	-	-	-
F_2	63	-	22	85	3:1	0.035	85-90
F_3	20	43	22	85	1:2:1	0.104	90-95
CO 6 × VBN 10							
F_1	45	-	-	45	-	-	-
F_2	62	-	22	84	3:1	0.063	80-85
F_3	19	43	22	84	1:2:1	0.261	85-90
CO 6 × VBN 11							
F_1	38	-	-	38	-	-	-
F_2	80	-	25	105	3:1	0.079	75-80
F_3	25	55	25	105	1:2:1	0.238	85-90

4. Conclusion

Understanding the inheritance of the qualitative traits is essential for supporting the breeding efforts aimed at developing improved cultivars with desired agronomic traits. The investigation into the inheritance patterns of terminal leaflet shape, leaf vein colour, immature pod colour and pod pubescence in blackgram revealed monogenic control of these traits across the crosses and pleiotropic effect of purple pigmentation on leaf veins and immature pods. These findings emphasize the potential utility of these qualitative traits as morphological markers for germplasm characterization and hybrid confirmation in blackgram breeding programs.

Compliance with ethical standards

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Disclosure of conflict of interest

No conflict of interest to be disclosed.

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