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Study on Interspecific Hybridization between Daincha (*Sesbania aculeatea*) and Related Species

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

Wide hybridization can enrich crops' genetic background and diversity by comparing different species and integrating the different parental traits. In the present study, interspecific hybridization was performed between Daincha and two related sesbania species by hand emasculation and crossing technique. A total of 460 hybrids were obtained in the *Sesbania aculeata* X *Sesbania bispinosa cross*, but no hybrids were obtained in their reverse cross, indicating the same cross combination has a large difference in the reciprocal crosses and that the hybrid has certain unidirectionality. The hybrids looked more like their *Sesbania aculeata* parent in stem and leaf characteristics. A total of 570 hybrids were obtained in the cross *Sesbania aculeata* X *Sesbania rostrata*, while no hybrids were obtained by reverse cross combination. The hybrid showed intermediate morphological traits when compared to their parents. Developing hybrids and

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segregating populations are useful genetic resources and provide interspecific hybridization to enlarge the gene pool of Daincha species.

Keywords: Crossability; Sesbania aculaeata; Sesbania bispinosa; Sesbania rostrata.

1. INTRODUCTION

Significant constraints affecting rice production are high production costs and low crop productivity. Although direct seeding of rice in the lowlands is a cost-effective and labour-saving method of crop improvement. Excessive weed growth is a major constraint leading to high production costs and low nitrogen use efficiency. Scarcity and high cost of organic manures also constrain wetland rice productivity in India. *Insitu* green manuring offers an effective method of providing organic manure and has considerable potential for weed suppression. Although many green manure crops are available, the N_2 fixing leguminous crops are important. Green manure crops supply organic manure and enrich the nitrogen status of the soil [1,2,3,4]. Among leguminous green manuring crops, daincha (*Sesbania aculeata*) is one of the most important crops, and its incorporation in the soil adds about 60-80 kgs of nitrogen/ha. *Sesbania* green manure, after decomposition, increases humus and available nitrogen and lowers the C:N ratio of soil [5]. Though daincha is a potential green manure crop, the genetic variability of the species is very low [6,7,8,9,10]. It is necessary to create genetic variability through various breeding programme. Hence the present breeding programme was formulated to study the crossability of three *Sesbania* species.

2. MATERIALS AND METHODS

Three *Sesbania* species (*Sesbania aculeata*, *Sesbanica bispinosa*, and *Sesbania rostrata)* were grown during Jun-Sep 2020 in the Experimental Farm of the Department of Plant Breeding and Genetics, Agricultural College and Research Institute, Tiruchirappalli. The staggered sowing was done in all three species for the continuous availability of the flower buds for the hybridization program. The interspecific crosses and their reciprocals were attempted in *Sesbania* species by emasculation at 5-6 AM and pollination at 8-9 AM upon flowering. The data were recorded on the number of flower buds emasculated and pollinated, the number of mature pods obtained, and the number of mature seeds obtained. The F_1 hybrids were raised and evaluated during Rabi 2020-21 along with their parents and observations were recorded.

3. RESULTS AND DISCUSSION

In the present crossing program, the crossability of Sesbania species ranged from 0-3.26% (Table 1) and inferred that the crossing is possible among the three species (*S. aculeata, S. bispinosa*, and *S. rostrata*). The difference in pod setting percentage indicates the presence of reproductive barriers that render introgression difficult among the crosses. The *Sesbania aculeata* X *Sesbanica bispinosa* combination exhibited a higher pod setting percentage. It was recorded that the pod setting was observed only when *Sesbania aculeata* was used as a female parent as compared to the other two species. The crossed pods differ from selfed pods by showing their ill filled appearance. Moreover, the crossed pods developed normal and wrinkled seeds on maturity.

The F_1 showed a very low seed germination percentage, ranging from 10 to 45.2 percent. The germination percentage was higher in the *Sesbania aculeata x Sesbania bispinosa* cross when compared to the *Sesbania aculeata* X *Sesbanica rostrata* cross. No pod setting due to the abscission of crossed flowers 4 days after pollination was observed in the reciprocal crosses. These reciprocal differences in the crossability of *Sesbania* species were noticed when *Sesbania bispinosa* or *Sesbania rostrata* was used as a female parent. These results clearly exhibited the presence of unfavorable interaction between the genetic and cytoplasmic factors [11]. This unfavorable interaction may be due to cross incompatibility barriers, *viz.,* delayed pollen germination and penetration of stigma, style and ovule [12] and slow pollen tube growth [13] unable to release male gametes to the ovule, failure to unite with the egg, and embryo abortion reported in various pulse crops. No external factors are involved in hybridizing these species because they have the same anthesis time, pollen/ ovule maturity period, and pollen dispersal time.

Table 1. Crossability of *Sesbania* species

S.No.	Particulars	*Sesbania aculeata* X *Sesbania bispinosa*	*Sesbania aculeata* X *Sesbania rostrata*
1.	Number of flowers crossed	460	570
2.	Number of crossed pods obtained	15	15
3.	Pod setting percentage (%)	3.26	2.63
4.	Number of crossed seeds obtained	105	102
5.	Seed setting percentage	22.82	17.89
6.	No. of seeds germinated	40	24.5
7.	Number of hybrid plants	15	10

In interspecific crosses, the endosperm development is affected by the absence or delayed endosperm nuclear division, which leads to the abortion of endosperm and subsequent drying of young pods. The observation of shrivelled seeds was noticed in *Sesbania aculeata* x *Sesbania bispinosa* and *Sesbania aculeata* x *Sesbania rostrata* crosses, is probably related to the failure of endosperm to reach maturity; similar findings were reported by various authors in *Vigna* species [14]. In distant hybridization, the low frequency of the pod set indicates the presence of prefertilization barriers, and reduced germination percentage and sterility of F_1 progenies confirmed the presence of post-fertilization barriers. The low hybrid pod recovery and varying levels of sterility in F_1's, hybrid inviability, lethality, and hybrid sterility [15] are explained by various workers in leguminous crops [16,17].

4. CONCLUSION

In the present hybridization programme, the hybrid showed morphological variation in intermediate leaf let size observed in *Sesbania aculeata* X *Sesbania rostrata* cross. Whereas in the *Sesbania aculeata* X *Sesbania bispinosa* cross, the hybrids were morphologically similar to the female parent. Hence, further confirmation by molecular marker is required to fix the true hybrids in this cross combination. The present study creates variability of desirable genotypes through hybridization and further generation advancement. Using alternative methods like mutation breeding, embryo rescue ovule culture, and chromosome doubling offers the development of new cultivars with desirable genes from related species.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of this manuscript.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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