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Heterosis in intra-specific hybrids of *Grewia optiva*: An important fodder tree of North Western Himalayas

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Abstract
The present study was undertaken to know heterotic effects of different genotypic combinations of *Grewia optiva* for nursery characters. Ten superior genotypes, six females (SO-1, SO-2, SO-4, SO-8, CH-2 and SI-15) and four male (SO-3, SI-6, BI-4 and HA-4) were chosen and were control crossed using Line X Tester (6X4 factorial) mating design. All the crosses produced viable seeds and heterosis was prevalent for the entire nursery characters. Maximum relative heterosis for plant height and collar diameter was displayed by cross SI-15 X BI-4 (49.06 and 44.14%), SO-8 X SI-6 and SO-8 X BI-4 (23.16 and 46.63%) exhibited maximum positive heterosis for number of leaves and internodal length. SO-8 X SI-6 has come up with highest positive mid parent value of 122.57% for leaf area (cm^2). For Leaf fresh and dry weight (g), significant positive heterosis was obtained in SO-8 X SI-6 (169.29 and 171.56%, respectively). On the basis of the results, SI-15 X BI-4, SO-8 X HA-4, SO-8 X BI-4 and SO-1 X BI-4 hybrids has been recommended for field level exploitation. Whereas, SI-15, SO-8 and SO-1 (female) and HA-4, BI-4 and SI- 6 (male) genotypes would be considered as stable parents for future breeding programmes.

Keywords: *Grewia optiva*, genotypes, breeding, heterosis, hybrid

Introduction
Grewia optiva, commonly known as 'Beul', having chromosome number 2n = 18 (Coleman, 1982) [5] is very popular agroforestry tree species for its utility as fodder, fuel and fiber It grows in low and mid-hills regions in the western and central Himalaya. It is naturally distributed in Bhutan, Nepal, Pakistan and in India it is found in areas of Himachal Pradesh, Uttarakhand, Jammu and Kashmir, Punjab, Sikkim, and parts of Uttar Pradesh (Orwa *et al* 2009) [15]. In western and central Himalayas it is mainly utilized for fodder purpose because livestock rearing is one of the major occupations in hilly regions that provides manure, draught power for agriculture and local transportation and forms important source of food and cash income to millions of households.

Closer scrutiny of the sector, however, reveals that the contribution to the GDP by livestock sector is far too low for such a large size of livestock population. This low productivity of the sector is as much attributable to underfeeding of the livestock and the poor livestock breeds. An idea of the meager fodder availability can be had from the fact that about 50 per cent of the cattle population depending largely upon free rangeland grazing in forests, pastures, village commons and the like end up getting only about 1.5 kg of dry fodder/day/ACU (Adult Cattle Unit) against the healthy fodder requirement norm of 3 per cent body weight. In absolute terms, the country by 2020 is facing an estimated shortage of 728 million tons of green fodder and 157 million tones-of dry fodder (Anonymous, 2013) [1]. Availability of nutritious fodder is the biggest constraint in animal husbandry. Except for rainy season (July to September), there is scarcity of fodder throughout the year. In hilly and mountain regions, the demand of feeds and fodder for livestock is much higher than their availability (Singh and Bimal, 2004) [17, 18] and the production of dry and green fodder was 2187,000 and 3230,000 tones respectively (Basic Animal Husbandry Statistics, 2004) [2]. Leaf fodder is a very useful resource of green fodder especially during winter months when all other fodder sources have been exhausted. The productivity of existing genotypes of *G. optiva* is comparatively low and not of much economic benefit to farmers. The wide gap in potential and actual yield is due to the use of locally available wild material. Superior quality planting material having high fodder quality and yield are being identified for further multiplication and production of quality planting material from the existing germplasm.

The development of high-yielding varieties through plant breeding has significantly increased the productivity, especially in the late half of the 20th century (Evenson and Gollin 2003) [6]. *G. optiva* is monoecious and highly cross-pollinated in nature that shows heterosis (2012) [20] and such a pollination mechanism can be exploited for the production of hybrid variety. Heterosis in tree growth is evident in many hybrids and is best illustrated in studies of *Eucalyptus* and *Populus* (Singh and Singh 2004) [17, 18]. Hybrid vigour and inheritance of economic traits have also been reported in plants from the Euphorbiaceae (Sridhar *et al.* 2009) [19]. Based on the earlier experience from other cross-fertilized crops, it appears that the application of heterosis breeding in *G. optiva* could justify hybrid variety production. Hybrid cultivars could be bred to use the heterosis effect. Growth and fodder quality can be genetically enhanced through development of hybrid varieties. Therefore, the present study was under taken to know heterotic effects of different genotypic combinations for nursery characters in *G. optiva*.

Materials and Methods

G. optiva genotypes from different districts of Himachal Pradesh are maintained in the seed orchard of Department of Tree Improvement and Genetic Resources, Dr Y. S. Parmar University of Horticulture and Forestry Nauni, Solan (H.P.) India. For carrying out present study, ten superior genotypes were chosen from the orchard for estimation of heterosis. The plant material of 6 females (SO-1, SO-2, SO-4, SO-8, CH-2 and SI-15) and 4 male (SO-3, SI-6, BI-4 and HA-4) genotypes were control crossed using Line X Tester (6X4 factorial) mating design (Table 1).

Table 1: Sources of parent material

Female Parents				Latitude	Longitude
S. no.	District	Village	Code		
1	Solan	Gaura	SO-1	30° 90'N	77° 09'E
2	Solan	Nauni	SO-2	30° 86'N	77° 16'E
3	Solan	Deog	SO-4	31° 10'N	77° 67'E
4	Solan	Kailar	SO-8	31° 19'N	76° 71'E
5	Chamba	Shahu	CH-2	32° 56'N	76° 10'E
6	Sirmour	Madhobag	SI-15	30° 71'N	77° 21'E
Male Parents					
1	Solan	Dharja	SO-3	30° 91'N	77° 03'E
2	Sirmour	Kalagat	SI-6	30° 51'N	77° 9'E
3	Bilaspur	Khutira	BI-4	31° 56'N	76° 48'E
4	Hamirpur	Hamirpur Kunal	HA-4	31° 67'N	76° 53'E

Observations were recorded on per cent successful crosses as per method used by Jan and Pfeffer (1999) [9]. The height of the shoots was recorded from the ground level to the apex of the leading shoot by using measuring scale. Collar diameter of the plant in the nursery was measured above the ground level at collar region with the help of digital calliper. The average number of leaves per plant was recorded on the basis of twelve randomly selected plants of each progeny. Internodel length (cm) was measured as distance between two nodes, at mid of the plant height. The full grown leaves were collected randomly from the seedlings and leaf area (cm^2) was measured with the help of leaf area meter. Leaf fresh weight was taken for a composite sample of 10 leaves randomly from F_1 hybrid plants and weighed and leaf dry weight by drying fresh leaves at 60 ^{0}C in hot oven till they lose moisture and attained a constant dry weight. Leaf dry matter content was obtained by the ratio of the weight of a leaf sample after drying at a temperature of (60± 2 ºC) to its fresh weight and expressed in per cent. Heterosis was calculated in terms of percentage increase or decrease of a hybrid against its mid parent value with respect to individual character, here after called relative heterosis or mid parent heterosis (Fehr 1987) [7].

$$\text{Relative heterosis} = \frac{F_1 - MP}{MP} \times 100$$

MP is mean value Mid parent

Results and Discussion

All the twenty-four crosses produced viable seeds. The analysis of variance for all the characters studied revealed highly significant differences among parents and their F_1 hybrids. This implies that there was significant genetic variability existing among parents and their F_1 hybrids (Table 2). The presence of positive significant heterosis over mid parent and better parent indicates significant increase and the negative heterosis over MP indicates significant decrease in the interested characters among the F_1 hybrids as compared to their parents. Significant differences were observed among crosses, the range for per cent successful crosses lied between 23.81 to 88.10 per cent for different crosses. Percent successful cross was highest in cross SO-2 X SO-3 (88.10%), followed by SO-2 X BI-4 (80.85%) and CH-2 X BI-4 (76.47%). The minimum was obtained for cross SO-8 X HA-4 (23.81%). (2012) [20] also reported that the per cent successful crosses in *Grewia optiva* genotypes lied between 20 to 70 per cent for different crosses. In an earlier study of Khanduri and Sharma (2002) [11] for intraspecific hybridization in *Pinus roxburghii* reported that controlled pollination resulted enormous fertilization success with no signs of incompatibility. Nagarajan *et al*. (1998) [13] revealed that fruit setting following controlled pollination was more than 75% in all clones of tamarind, in contrast only negligible fruit set was observed in self-pollination and have indicated that tamarind is a self-incompatible species. Similarly, Omokhafe *et al*., (2007) [14] reported significant variation in fruit set across the twelve crosses in *Hevea brasiliensis*. There was higher fruit set (7.14%) in the main crosses than in the reciprocals (6.42%). Chaudhary (2011) [12] reported the percent successful crosses in *Salix* varied between 69.80% for *S. tetrasperma* (LP) x *S. tetrasperma* (TFB) and 7.30 per cent for *S. tetrasperma* (LNF) x *S. alba* (SE63-007). Per cent successful cross was highest (70%) in cross HA-4 (Hamirpur Kanal) × SI-10 (Seenaghat) and lowest (20%) was obtained for cross SO-8 (Kailar) × KA-1 (Dharamshala).

Outcome of the present investigations revealed that heterosis over mid parent value was prevalent for the entire nursery characters (plant height, collar diameter, number of leaves, intermodal length and leaf area) and for dry matter production. Results pertaining to plant height among F_1 hybrids showed that all the crosses expressed positive heterosis. Maximum positive heterosis was shown by cross SI-15 X BI-4, SO-8 X SI-6 and SO-8 X BI-4 (+49.06, 46.20 and 44.56%, respectively). The lowest positive mid parent heterosis was observed in SO-2 X SI-6 and SO-2 X SO-3 (+7.44 and 8.40%, respectively) (Fig 1.). With respect to collar diameter, maximum positive significant heterosis was shown by the crosses SO-8 X BI-4 and SO-8 X SI-6 (+98.63 and 86.37%). Maximum hybrids expressed positive heterosis over mid parent and two among them showed negative mid parent heterosis. Significant positive heterosis (+23.16 and 22.91%) was shown by the crosses SO-8 X SI-6 and SO-8 X BI-4 respectively. Highest negative mid parent heterosis (-

4.91 and -1.60%) was exhibited by SO-2 X SI-6 and CH-2 X SO-3 crosses. The observation on F_1 hybrids for internodel length (cm) revealed that there was an increase trend in internodel length over their mid parent. Out of 24 crosses, only one (SO-2 X SI-6) showed maximum negative relative heterosis (–0.94%). Cross SO-2 X SO-3 again maintained its consistence with respect to internodel length character without showing any percent increase or decrease over mid parent. Maximum significant positive heterosis of +46.63and 43.00% was shown by the crosses SO-8 X BI-4 and SO-8 X SI-6 respectively (Table 3.). The above results are in conformity with the findings of Castillo *et al*. (1998) [3], who have reported that F_1 hybrid between *Leucaena leucocephala* K636 × *L. pallida* K748 (KX2 hybrid) in particular has demonstrated outstanding characteristics as forage plant combined with high palatability and growth characters. Msangi *et al*. (2004) [12] in the evaluation of *Leucaena* species and its hybrid for fodder production reported significant differences among the species and its hybrid for fodder production, growth and adoptability. Other studies made by Smart *et al.*, (2005); Chaudhary, (2011) [12] have demonstrated the superior performance of intra-specific hybrids over their parents in willows.

Furthermore, 70.00% of the F_1 hybrids displayed positive heterosis for leaf area (cm^2), and 30.00% showed negative heterosis over their mid parents. Maximum positive mid parent (122.57%) was observed in SO-8 X SI-6 followed by SO-8 X BI-4 (64.20%), whereas maximum negative heterosis of-25.99 and -18.25% was expressed by the crosses CH-2 X BI-4 and SO-2 X SO-3 respectively. For leaf fresh and dry weight (g), 54.16% of the F_1 hybrids displayed negative heterosis and 45.83% showed positive heterosis for fresh leaf weight and 41.66% of crosses exhibited negative heterosis and 59.33% with positive heterosis for leaf dry weight over their mid parents. Maximum positive mid parent heterosis for fresh leaf weight (169.29 and 103.62%) was revealed in F_1 SO-8 X SI-6 and SO-8 X BI-4, whereas maximum negative heterosis (-41.32 and -37.95%) was expressed by the crosses SO-2 X HA-4 and SO-2 X SO-3 respectively. In case of leaf dry matter production, 66.66% of the F_1 hybrids exhibited positive heterosis and 33.33% showed negative heterosis over their mid parents (Table 3.).

Ghosh *et al.* (2009) [8] evaluated eight mulberry (*Morus* sp.) hybrids, *viz.*, C-2036, S-1908, C-2037, C-2038, C-2039, C-2040, C-2041 and C-2042 against S-1635. Heterosis for leaf yield and its attributing characters in 8 hybrids were significant. Positive heterosis was ranging from 5.95 to 159.31% over better parent, 13.92 to 159.31% over mid parent and 1.50 to 27.79% over standard variety (S-1635) among the crosses. Similar results were also found by Kadam (2002) [10], Singh and Singh (2004) [17, 18] and Ozel *et al.* (2010) [16] in Poplars.

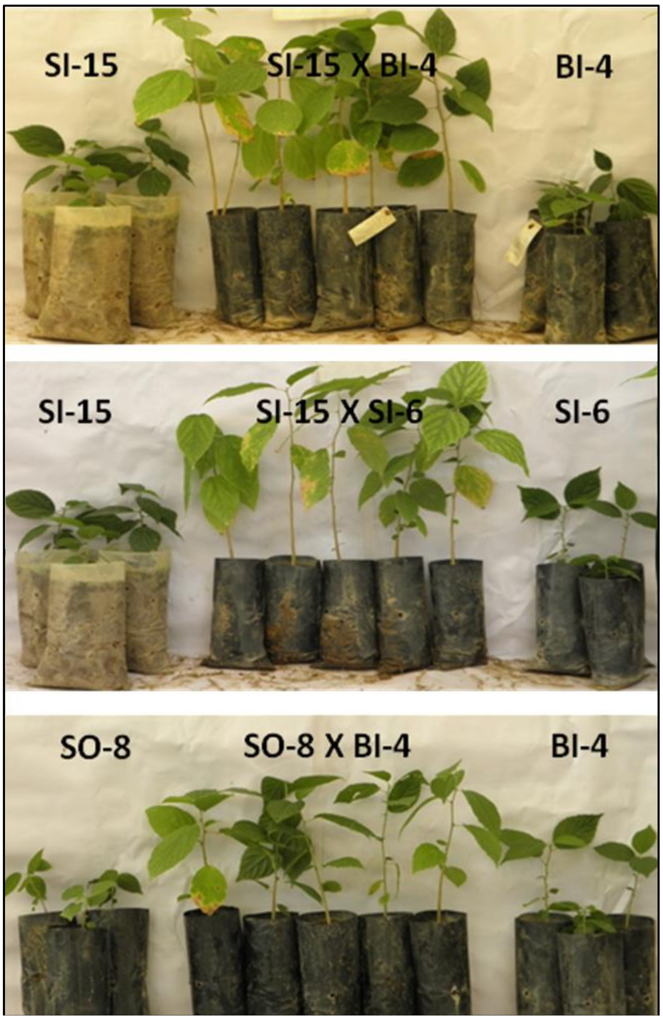

Fig 1: F_1's showing heterosis for plant height character over its parents

Table 2: Analysis of variance for heterosis (plant height, collar diameter, number of leaves, internodel length leaf fresh and dry weight (g)

Sources of variation	df	Plant height (cm)	Collar diameter (mm)	No. of leaves	Internodel length (mm)	Leaf area (cm^2)	Leaf weight (g)		Leaf dry mater production (g)
							Fresh wt	Dry wt	
Rep	2	5.05**	0.94 ns	0.81 ns	0.02 ns	20.06**	0.13 ns	0.02 ns	0.20 ns
Tret	23	76.13**	21.43**	11.06**	1.75ns	4416.25**	36.51**	4.40**	2046.46**
Error	46	20.46	1.87	9.39	0.13	1272.30	1.57	0.06	5.41
total	71	101.64	24.24	21.27	1.89	5708.61	38.22	4.48	2052.07

Table 3: Per cent successful cross and performance of F_1 hybrids for percentage increase (+) or decrease (-) over mid parent (relative heterosis) in terms of plant height (cm), collar diameter (mm), number of leaves, internodel length (cm), leaf area (cm^2) leaf weight and leaf dry matter production

Crosses	Per cent Successful cross	Plant height (cm)	Collar diameter (mm)	No. of leaves	Internodel length (cm)	Leaf area (cm^2)	Leaf weight (g)		Leaf dry mater production(g)
							Fresh wt	Dry wt	
SO-1 X SO-3	52.54	24.58**	31.26**	15.12**	9.92**	26.90**	23.27**	54.24**	27.82**
SO-1 X SI-6	40.00	22.62**	25.22**	10.66**	9.92**	22.07**	-20.71	8.57**	37.46**
SO-1 X BI-4	58.93	20.66**	7.28**	13.61**	6.20**	7.69**	-14.49	37.78**	61.20**
SO-1 X HA-4	35.21	18.09**	1.93**	12.36**	6.23**	27.60**	34.02**	45.65**	8.86**
SO-2 X SO-3	88.10	8.40**	22.75**	0.00	0.00	-18.25	-37.95	-45.21	-11.68
SO-2 X SI-6	54.55	7.44**	20.57**	-4.91	-0.94	-11.17	-33.13	-36.84	-6.23
SO-2 X BI-4	80.85	11.11**	30.45**	0.53	6.93**	6.55**	17.24**	28.81**	10.71**
SO-2 X HA-4	75.56	17.05**	39.40**	3.81**	8.42**	-16.67	-41.32	-18.33	40.14**
SO-4 X SO-3	68.42	22.58**	2.83**	3.47**	10.10**	10.03**	-12.54	0.78	16.12**

SO-4 X SI-6	53.85	26.93**	20.23**	3.70**	11.11**	-2.90	-3.81	-6.90	-3.31
SO-4 X BI-4	62.79	30.68**	25.32**	7.32**	20.86**	-13.46	-16.49	-0.99	18.64**
SO-4 X HA-4	76.19	36.38**	45.93**	11.60**	22.86**	60.73**	62.00**	94.17**	19.68**
SO-8 X SO-3	75.00	31.23**	8.43**	9.89**	12.47**	-11.87	-37.11	-19.67	29.69**
SO-8 X SI-6	69.77	46.20**	86.37**	23.16**	43.00**	122.57**	169.29**	171.56**	1.14
SO-8 X BI-4	61.90	44.56**	98.63**	22.91**	46.63**	64.20**	103.62**	121.28**	8.68**
SO-8 X HA-4	23.81	30.74**	28.89**	7.30**	20.11**	11.02**	-12.71	-12.50	0.15
CH-2 X SO-3	64.79	24.16**	1.61	-1.60	5.56**	16.44**	16.23**	4.48**	-10.78
CH-2 X SI-6	61.22	37.34**	21.09**	3.98**	9.60**	4.52**	0.74	-12.40	-13.13
CH-2 X BI-4	76.47	38.81**	26.81**	6.52**	19.79**	-25.99	-4.51	-13.21	-9.68
CH-2 X HA-4	68.89	35.92**	39.01**	1.24	15.98**	1.49	-11.74	7.41**	19.72**
SI-15 X SO-3	47.06	38.73**	0.73	11.68**	10.86**	19.07**	-5.85	-9.35	-3.52
SI-1 5 X SI-6	45.76	43.06**	16.26**	11.86**	12.90**	34.52**	51.32**	36.51**	-10.10
SI-15 X BI-4	40.58	49.06**	44.14**	17.10**	23.41**	29.40**	60.00**	67.57**	5.21**
SI-15 X HA-4	41.67	29.90**	-2.91	10.12**	6.65**	28.08**	39.05**	55.75**	12.25**

Conclusion

Hybrids SI-15 X BI-4, SO-8 X HA-4, SO-8 X BI-4 and SO-1 X BI-4 have been recommended for field level exploitation for the farmers. Whereas, SI-15, SO-8 and SO-1 (female) and HA-4, BI-4 and SI-6 (male) genotypes may be considered as stable parents in future breeding programmes.

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