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Diallel analysis of seven promising genotypes of opium poppy for assessment of their combining ability and efficacy in future genetic improvement programmes

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Abstract

Opium poppy (*Papaver somniferum* L.) is a widely cultivated plant species, producing a number of pharmaceutically important alkaloids and highly nutritive seeds. In the present investigation, combining ability analysis and reciprocal effect were studied in a 7 × 7 diallel set of opium poppy. Analysis of variance revealed the presence of significant variance due to general combining ability (GCA), specific combining ability (SCA) and reciprocal effect among the parents for all the traits except for codeine content. Combining ability analysis revealed the involvement of both additive and non-additive gene action in the inheritance of most of the traits. On the basis of GCA, SCA effects and per se performance, genotype SPS-20, Sanchita and Ajay were identified as the most eligible parents for future hybridization programmes for better seed yield, development of morphine-less cultivars and quantitative improvement of different types of alkaloid in poppy straw, respectively.

Key words: combining ability, diallel analysis, genetic improvement, opium poppy, *Papaver sominferum*, reciprocal effect.
Abbreviations: GCA, general combining ability; SCA, specific combining ability; gi, estimate of GCA effect; Sij, estimate of SCA effect; Srj, estimate of reciprocal effect.

Introduction

Opium poppy (*Papaver somniferum* L., Papaveraceae) is a medicinal plant cultivated globally for the production of pharmaceutically important opiates and heroin (Marciano et al. 2018). *P. somniferum* is considered as the only source for several high-value pharmaceutically important benzylisoquinoline alkaloids including narcotic analgesics morphine and thebain, antitussive agent codeine, anti-cancer drug noscapine, antimicrobial agent sanguinarine and vasodilator agent papaverine (Pathak et al. 2013), which are utilized as a major feedstock for synthesis of important drugs (Frick et al. 2005). Moreover, the species is also acknowledged for nutraceutical value of its seeds as a rich source of essential minerals, protein (24%), and linoleic acid (68%), which help to lower blood cholesterol levels (Vos, Cunnne 2003; Sacks, Compos 2006). Owing the economical value, there is a significant interest for genetic improvement of *P. somniferum*. Among the unexplored aspects, evaluation of different germplasm for their agronomic traits, yield component and combining ability are most imperative ones as offering precious information necessary for development of elite genotypes with desirable traits. In this context, traditional diallel analysis is more efficient and frequently utilized to acquire information on genetic effects, an approximate estimate of general combining ability (GCA) and specific combining ability (SCA), heritability for a population of parental lines, and to identify the potential heterotic combinations as well as heterotic patterns (Bertoria et al. 2006; Rastogi et al., 2013).

Several recent studies attempted diallel crosses in diverse genotypes of *P. somniferum* to understand inheritance of various agronomic traits and their combining ability (Lal et al. 2014; Shukla et al. 2019; Yazici, Yilmaz 2020), but still there are unresolved issues in this respect. To obtain further information necessary for identification of suitable genetic stocks for future hybridization programmes, more concerted efforts are needed. In the present investigation, seven genotypes of opium poppy were crossed in diallel fashion and screened for performance of their F_1 progeny, their breeding potential in specific combination, and for the whole performance of the hybrids, in order to select the promising genotypes for future yield improvement programmes.

Materials and methods

In the present study, seven genotypes of *P. somniferum* (Shweta, Sujata, SPS-20, Sanchita, Ab-1, CIMAP-Ajay, and Dr-44) belonging to different genetic stocks, improved varieties and land races were collected or procured from different eco-geographical regions of the world. In the milieu of qualitative and quantitative yield, the performance of all accessions was evaluated by growing them at the experimental field of Genetics & Plant Breeding Unit of CSIR, CIMAP, Lucknow, located at 26.5° N and 80.50° E, 120 m above sea level, in November to April during 2013 – 2014 and 2014 – 2015. Later, a pure line progeny was selected as a parent and used for diallel analysis (P_1 – P_7, Table 1).

All parents were crossed in a diallel mating design in all possible combinations, including reciprocals to obtain F_1 seeds (Griffing 1956a; Griffing 1956b). The hybridization and evaluation of F_1 hybrids were performed in 2015 – 2016 and 2016 – 2017, respectively. All hybrids, including parents, F_1 and reciprocals, were grown in a randomized block design with three replications from November to April. One row of each entry was grown in each replication with spacing of 10 cm between plants within rows and 40 cm between rows. Standard cultural practices were followed throughout the cropping season, which included pre-sowing addition of farmyard manure at a rate of 10 t ha^{-1} nitrogen, 80 kg ha^{-1} phosphorus, and 40 kg ha^{-1} potassium. The data for 13 agronomic traits (days to 50% flowering, plant height, peduncle length, days to maturity, number of capsules per plant, capsule index, seed yield per plant, capsule husk yield per plant, straw alkaloid content i.e. morphine, codeine, thebaine, papaverine and narcotine) were recorded as per the adopted methodology (Yadav et al. 2009a; b)

Table 1. Parent *Papaver somniferum* genotypes used in diallel analysis

No.	Abbreviation	Name	Source of origin
1	P1	Shweta	CIMAP, Lucknow, India
2	P2	Sujata	CIMAP, Lucknow, India
3	P3	SPS- 20	CIMAP, Lucknow, India
4	P4	Sanchita	CIMAP, Lucknow, India
5	P5	Ab-1	Kosice, Slovak Republic
6	P6	Ajay	CIMAP, Lucknow, India
7	P7	Dr 44	CIMAP, Lucknow, India

For chemical analysis, the dried powder of capsule husk (1 g) was first dissolved in methanol and sonicated for 30 min in an ultrasonic bath. The solution was then centrifuged at 10 000 rpm for 10 min and then later used for high-performance thin-layer chromatography analysis. Each standard was separately weighed and a stock solution was prepared. From each standard stock solution, an equal volume was taken and mixed to prepare a working standard. Thin-layer chromatography densitometric procedure was used to analyze the five major opium alkaloids: morphine, codeine, thebaine, papaverine and narcotine (Gupta, Verma 1996). Toluene-acetone-methanol-ammonia (40:40:6:2) was used as a mobile phase. Silica gel plates 60 F_{254} (Merck, Darmstadt, Germany) were scanned after derivatization using Dragendorff reagent No. llC to detect alkaloid at 540 nm (Wagner et al. 1984).

The pooled data was statistically analyzed for mean value, ANOVA, general combining ability (GCA), specific combining ability (SCA) and reciprocal effect using Statistical Software 4.0 version and the methodology adopted by Yadav et al. (2009a).

Table 2. Results of ANOVA analysis (mean sum squares) for general combining ability (GCA), specific combining ability (SCA) and reciprocal effect for 13 traits in 7 × 7 diallel cross of opium poppy. d.f., degrees of freedom, *, $p < 0.05$; **, $p < 0.01$

No.	Characteristic	Source of variation			
		GCA (d.f. = 6)	SCA (d.f. = 21)	Reciprocal (d.f. = 21)	Error (d.f. = 96)
1	Days to 50% flowering	64.542**	31.12**	19.064**	8.063
2	Plant height	242.55**	20.85**	45.043**	0.181
3	Peduncle length	12.26**	5.35**	9.530**	0.364
4	Days to maturity	303.58**	34.07**	84.465**	3.567
5	Number of capsules	4.53×10^{-2}*	9.90×10^{-2}**	1.58×10^{-2}**	6.3×10^{-2}
6	Capsule index	1.0×10^{-2}**	2.0×10^{-2}**	2.0×10^{-2}**	0.000
7	Seed yield	4.21×10^{-2}**	4.45×10^{-2}*	3.46×10^{-2}**	4.30×10^{-2}
8	Poppy husk yield	6.49×10^{-2}**	7.45×10^{-2}**	5.32×10^{-2}**	2.00×10^{-1}
9	Morphine concentration	3.30×10^{-4}**	1.90×10^{-4}**	3.30×10^{-2}**	5.20×10^{-4}
10	Codeine concentration	8.50×10^{-4}	2.20×10^{-6}	5.00×10^{-4}	1.30×10^{-7}
11	Thebaine concentration	6.70×10^{-6}**	6.71×10^{-5}*	9.90×10^{-6}**	2.00×10^{-7}
12	Papaverine concentration	2.26×10^{-6}**	5.03×10^{-6}**	4.60×10^{-4}**	1.32×10^{-6}
13	Narcotine concentration	1.61×10^{-6}**	1.17×10^{-6}**	1.88×10^{-5}**	1.27×10^{-9}

Results

Analysis of variance for combining ability revealed that mean square values were significant for GCA, SCA and reciprocals, thus signifying the role of both additive and non-additive genetic variance, as well as variance due to cytoplasmic differences, for all the thirteen traits studied except for codeine concentration (Table 2).

Early flowering is a desirable characteristic, and therefore a negative value of GCA is desirable for this trait. Three parents showed negative GCA effect where GCA was most pronounced by P_3 followed by P_7 and P_1 (Table 3). P_1 had comparatively higher mean (101.93) and negative GCA (gi) (−0.514) values indicating that for this trait P_1 could be a most desirable parent genotype. Eleven hybrid crosses showed negative SCA effect whereas for earliness the best specific combinations was $P_5 \times P_6$ (−6.03) followed by and $P_5 \times P_7$ (−4.33). Six crosses, namely $P_1 \times P_4$, $P_1 \times P_5$, $P_2 \times P_5$, $P_2 \times P_6$, $P_3 \times P_5$ and $P_3 \times P_6$, had positive and high SCA effect for days to 50% flowering, and were considered as undesirable for this trait (Table 4). In context of earliness, desirable negative and significant reciprocal effect were shown by nine crosses (Table 5).

Tall plants are always susceptible to lodging and thus plants having medium plant stature were considered for selection and the negative component of combining ability was preferred. Parents P_3 and P_7 showed desirable negative GCA effect for plant height (Table 3). Seven crosses possessed high negative SCA effect for plant height (Table 4). The desirable reciprocal effect for height, i.e. negative significant values, were shown by several crosses ($P_3 \times P_1$, $P_3 \times P_2$, $P_5 \times P_1$, $P_5 \times P_3$, $P_6 \times P_1$, $P_6 \times P_2$, $P_6 \times P_3$, $P_6 \times P_5$, $P_7 \times P_1$, $P_7 \times P_2$, $P_7 \times P_3$, $P_7 \times P_4$, $P_7 \times P_5$ and $P_7 \times P_6$; Table 5).

In context of peduncle length, P_1 showed the highest mean (22.43) with positive GCA (gi = 0.43). The negative GCA value for parents P_3, P_4 and P_7 indicated their poor general combination ability for peduncle length (Table 3). Crosses $P_1 \times P_3$, $P_1 \times P_5$, $P_2 \times P_4$, $P_3 \times P_4$, $P_4 \times P_5$, $P_4 \times P_6$ and $P_6 \times P_7$ showed high SCA effect for peduncle length (Table 4). For peduncle length, eight crosses had desirable positive and significant reciprocal effect (Table 5).

Early maturity is a desirable character and therefore negative estimates can be selection criteria. The highest negative GCA effect (gi = −6.394) for this trait was exhibited by the parent P_3. High negative SCA were shown by $P_1 \times P_2$, $P_1 \times P_3$, $P_1 \times P_6$, $P_2 \times P_3$, $P_2 \times P_4$, $P_2 \times P_5$, $P_4 \times P_5$, $P_4 \times P_6$, $P_4 \times P_7$, $P_5 \times P_7$ and $P_6 \times P_7$, while $P_4 \times P_2$, $P_4 \times P_3$, $P_5 \times P_2$, $P_5 \times P_3$ and $P_6 \times P_1$ showed high negative reciprocal effect for days to maturity (Tables 4 & 5). The parent P_6 followed by P_3 and P_4 had the highest mean value (3.00) and positive GCA effect (0.026), for number of capsules/plant. The cross between $P_1 \times P_3$, $P_1 \times P_4$, $P_1 \times P_5$, $P_1 \times P_6$, $P_4 \times P_6$ and $P_4 \times P_7$ had high SCA for number of capsules per plant while desirable reciprocal effect was found for $P_3 \times P_2$, $P_5 \times P_1$, $P_5 \times P_3$, and $P_6 \times P_4$. In respect to capsule index, the parent P4 recorded ahigh mean value (0.971) and higher GCA effect (0.009). High significant SCA were seen in the crosses $P_1 \times P_3$, $P_1 \times P_5$, $P_1 \times P_6$, $P_2 \times P_3$, $P_2 \times P_4$, $P_2 \times P_5$, $P_2 \times P_7$, $P_3 \times P_6$, $P_4 \times P_5$, $P_4 \times P_7$, $P_5 \times P_6$, $P_5 \times P_7$ and $P_6 \times P_7$. For this trait high reciprocal effect was expressed by hybrids $P_2 \times P_1$ and $P_5 \times P_1$ (Tables 3, 4, 5).

In perspective of the economically important trait like seed yield per plant, P_1 was the most desirable parent as it had notably a high mean value along with the highest desirable positive GCA effect (0.20). For seed yield, significant high SCA was recorded for all crosses except for $P_1 \times P_2$, $P_1 \times P_4$ and $P_1 \times P_7$, while high reciprocal effect was seen for $P_2 \times P_1$, $P_3 \times P_1$, $P_4 \times P_1$, $P_5 \times P_1$, $P_5 \times P_2$, $P_5 \times P_3$, $P_6 \times P_1$, $P_6 \times P_4$, $P_6 \times P_5$, $P_7 \times P_1$, $P_7 \times P_4$ and $P_7 \times P_6$. For capsule husk yield, the parent P1 had a notable mean value (6.22) and the highest positive GCA effect (0.32). For capsule husk yield, 16 and 9 crosses showed significant positive GCA and reciprocal effect, respectively (Tables 4 & 5).

For the pharmaceutically important alkaloid concentration like that of morphine, parents P_5 and P_6 exhibited a high mean value as well as high GCA effect (9.6×10^{-4}, 6.12×10^{-4}) indicating they usefulness for development of high morphine containing varieties. Parents P_1, P_2 and P_4 showed negative GCA effect for morphine concentration, signifying their potential use in development of morphine-less varieties. For morphine concentration, high SCA was recorded for crosses $P_1 \times P_6$, $P_2 \times P_5$, $P_2 \times P_6$, $P_3 \times P_4$, $P_3 \times P_5$, $P_3 \times P_6$, $P_3 \times P_7$ and $P_5 \times P_7$, while high reciprocal effect was noted for $P_2 \times P_1$, $P_4 \times P_1$, $P_2 \times P_3$, $P_5 \times P_1$, $P_5 \times P_4$, $P_6 \times P_5$ and $P_7 \times P_5$. For codeine concentration, P6 had the highest mean value (1.0×10^{-1}) and high GCA effect (1.49×10^{-4}) while high SCA was evident for $P_1 \times P_5$, $P_1 \times P_6$, $P_2 \times P_4$, $P_2 \times P_7$, $P_3 \times P_4$, $P_3 \times P_7$, and $P_4 \times P_7$. Several crosses ($P_3 \times P_2$, $P_4 \times P_3$, $P_5 \times P_1$, $P_5 \times P_4$, $P_6 \times P_1$, $P_6 \times P_4$, $P_7 \times P_4$, $P_7 \times P_5$) expressed significant reciprocal effect for codeine concentration. For thebaine concentration, the parent genotype P6 possessed the highest mean value (4.3×10^{-3}) as well as positive GCA effect (3.6×10^{-4}) and promising hybrids were $P_1 \times P_4$, $P_1 \times P_5$, $P_1 \times P_6$, $P_2 \times P_5$, $P_2 \times P_7$, $P_3 \times P_4$, $P_3 \times P_7$, $P_4 \times P_5$ and $P_4 \times P_7$. Besides this, the hybrid lines $P_2 \times P_1$, $P_3 \times$ P1, $P_3 \times P_2$, $P_4 \times P_2$, $P_4 \times P_3$, $P_5 \times P_1$, $P_5 \times P_4$, $P_6 \times P_1$, $P_6 \times P_4$, $P_6 \times P_5$, $P_7 \times P_2$, $P_7 \times P_4$ and $P_7 \times P_5$ possessed high reciprocal effect. In context of papaverine the parent genotype P_6 had a high mean value (2.3×10^{-3}) and positive GCA (3.6×10^{-3}). For papaverine concentration, the crosses $P_1 \times P_5$, $P_2 \times P_4$, $P_2 \times P_5$, $P_2 \times P_6$, $P_3 \times P_5$, $P_3 \times P_6$, $P_3 \times P_7$ and $P_4 \times P_7$ showed high SCA, whereas out of 21 crosses 12 had positive reciprocal effect and the remaining had negative effect. For narcotine, the parent genotype P_6 did not have the highest mean value but showed positive and desirable GCA (2.0×10^{-2}). Hence it was good combiner with other parents. The crosses $P_1 \times P_6$, $P_1 \times P_7$, $P_2 \times P_5$, $P_2 \times P_6$, $P_2 \times P_7$, $P_3 \times P_5$, $P_3 \times$ P7, $P_4 \times P_5$ and $P_5 \times P_7$ exhibited high SCA for narcotine concentration, while crosses with significant reciprocal effect were $P_3 \times P_2$, $P_6 \times P_4$, $P_7 \times P_3$ and $P_7 \times P_5$ (Tables 3, 4, 5).

Table 3. General combining ability (GCA) effects, GCA and specific combining ability (SCA) variances for thirteen traits in seven parents of opium poppy. SE, standard error; gi, general combining ability effect

Parents	Traits	Days to flower (50%)	Plant height (cm)	Peduncle length (cm)	Days to maturity	Number of capsules per plant	Capsule index (width / length)	Seed yield (g)	Capsule husk yield (g)	Morphine concen–tration (%)	Codeine concen–tration (%)	Thebaine concen–tration (%)	Papaverine concen–tration (%)	Narcotine concen–tration (%)
P_1	Mean	101.93	108.46	22.43	134.15	1.89	0.95	4.54	6.22	2.3×10^{-2}	2.2×10^{-4}	1.5×10^{-3}	1.1×10^{-3}	3.0×10^{-3}
	gi	–0.51	2.26	0.43	–3.50	–0.12	–0.010	0.200	0.320	-1.4×10^{-3}	-5.1×10^{-4}	1.0×10^{-3}	1.0×10^{-2}	-8.0×10^{-3}
P_2	Mean	106.10	96.70	20.09	128.27	2.89	0.86	4.30	6.48	0	9.6×10^{-4}	0	0	0
	gi	2.13	0.05	0.29	–3.55	–0.17	0.010	0.150	0.160	-5.8×10^{-3}	-1.1×10^{-3}	-4.6×10^{-4}	-2.4×10^{-3}	-3.0×10^{-3}
P_3	Mean	97.06	88.26	16.83	117.33	2.78	0.937	5.17	5.18	9.3×10^{-3}	4.8×10^{-4}	0	3.3×10^{-3}	2.8×10^{-3}
	gi	–3.16	–2.31	–0.72	–6.39	0.29	0.009	–0.03	0.010	1.0×10^{-4}	4.7×10^{-4}	-1.9×10^{-4}	2.2×10^{-3}	4.0×10^{-3}
P_4	Mean	107.00	92.49	14.22	142.55	2.33	0.971	3.45	4.57	8.4×10^{-3}	2.3×10^{-4}	0	2.2×10^{-3}	2.1×10^{-3}
	gi	0.42	0.94	–1.08	2.32	0.19	0.009	–0.180	0.150	-4.5×10^{-4}	-1.7×10^{-4}	2.2×10^{-4}	-1.2×10^{-3}	-7.0×10^{-3}
P_5	Mean	101.06	107.60	22.10	148.77	2.56	0.850	3.73	4.59	5.5 × 10–3	6.3×10^{-4}	0	1.2×10^{-3}	1.1×10^{-3}
	gi	0.24	6.27	1.43	5.81	–0.17	–0.006	0.001	0.004	9.6×10^{-4}	-6.4×10^{-4}	1.5×10^{-4}	-9.0×10^{-3}	-9.0×10^{-3}
P_6	Mean	110.33	94.77	20.56	141.33	3.00	0.860	3.63	4.22	3.0×10^{-3}	1.0×10^{-1}	4.3×10^{-3}	2.3×10^{-3}	0
	gi	2.91	0.17	0.59	5.01	0.03	0.000	0.116	0.001	6.2×10^{-4}	1.5×10^{-4}	3.6×10^{-4}	3.6×10^{-3}	2.0×10^{-2}
P_7	Mean	101.00	81.01	21.72	135.39	2.56	0.880	3.54	4.36	3.7×10^{-3}	6.3×10^{-3}	9.0×10^{-3}	5.4×10^{-3}	0
	gi	–2.03	–7.38	–0.94	0.30	–0.04	–0.010	–0.265	–0.360	4.7×10^{-4}	4.6×10^{-4}	-1.6×10^{-3}	-2.0×10^{-3}	-4.0×10^{-3}
	GCA variance	4.9×10^{-3}	1.1×10^{-2}	2.2×10^{-2}	2.2×10^{-2}	3.8×10^{-3}	7.0×10^{-5}	3.0×10^{-2}	1.0×10^{-2}	3.0×10^{-5}	1.0×10^{-6}	1.0×10^{-10}	8.0×10^{-8}	7.0×10^{-9}
	SCA variance	59.2×10^{-1}	1.3×10^{-1}	2.7×10^{-1}	26.2×10^{-1}	4.6×10^{-4}	8.0×10^{-4}	3.1×10^{-2}	1.5×10^{-2}	4.0×10^{-4}	1.6×10^{-5}	1.0×10^{-9}	9.0×10^{-7}	9.0×10^{-10}
	SE (gi)	7.0×10^{-1}	1.1×10^{-1}	1.5×10^{-1}	4.7×10^{-1}	6.2×10^{-2}	3.0×10^{-2}	5.1×10^{-2}	3.5×10^{-2}	1.7×10^{-3}	3.1×10^{-4}	1.0×10^{-4}	8.9×10^{-6}	8.4×10^{-7}

Table 4. Estimation of specific combining ability effects of 13 traits in 7 × 7 diallel cross of opium poppy. S_{ij}, the specific combining ability effect for the cross between i^{th} and j^{th} parent; S_{ik}, specific combining ability effect for the cross between i^{th} and k^{th} parent; *, $p < 0.05$; **, $p < 0.01$

Parents	Days to flower (50%)	Plant height (cm)	Peduncle length (cm)	Days to maturity	Number of capsules per plant	Capsule index (width / length)	Seed yield (g)	Capsule husk yield (g)	Morphine concen–tration (%)	Codeine concen–tration (%)	Thebaine concen–tration (%)	Papaverine concen–tration (%)	Narcotine concen–tration (%)
$P_1 \times P_2$	–1.12**	–2.36**	0.95**	–4.310**	–0.154**	–0.021**	0.050	0.4093**	–0.0087**	–0.00100**	–0.00046**	–0.00213**	–0.00185**
$P_1 \times P_3$	–3.38**	–0.13*	1.05**	–3.768**	0.496**	0.023**	0.351**	0.2841**	–0.0126**	–0.00140**	–0.00050**	–0.00318**	–0.00187**
$P_1 \times P_4$	3.30**	–1.30**	–1.28**	4.615**	0.545**	–0.047**	0.033	0.3187**	0.0005	0.000340	0.00043**	–0.00137**	–0.00155**
$P_1 \times P_5$	8.50**	0.04	1.64**	2.566**	0.346**	0.025**	0.094**	0.1896**	–0.0004	0.001800**	0.00059**	0.00290**	–0.00129**
$P_1 \times P_6$	–2.27**	1.92**	–2.34**	–5.737**	0.408**	0.010**	0.139**	0.3184**	0.0211**	0.001450**	0.00011**	–0.00250**	0.00887**
$P_1 \times P_7$	–3.01**	–5.12**	–2.01**	0.214	–0.734**	–0.049**	0.026	0.0155	–0.009**	–0.00162**	–0.00070**	–0.00044**	0.00058**
$P_2 \times P_3$	–2.83**	1.49**	0.17*	–3.957**	0.099**	0.025**	0.055*	–0.0358	–0.0073**	–0.00052*	–0.00022**	–0.00233**	–0.00020**
$P_2 \times P_4$	0.71	2.77**	1.482**	–2.119**	–0.574**	0.012**	0.057*	–0.822**	–0.0008	0.00056**	–0.00021**	0.00107**	–0.0003**
$P_2 \times P_5$	3.66**	2.30**	–1.368**	–3.554**	–0.107**	0.013**	0.248**	–0.5613**	0.0203**	0.00034	0.00154**	0.00368**	0.00058**
$P_2 \times P_6$	2.42**	–0.92**	0.217*	6.178**	0.028**	–0.003**	0.272**	0.4298**	0.00334**	–0.00023	–0.00003**	0.00182**	0.00127**
$P_2 \times P_7$	0.297	–2.88**	–1.391**	7.124**	–0.298**	0.030**	0.185**	0.696**	–0.00182	0.00049*	0.00005**	–0.00108**	0.00117**
$P_3 \times P_4$	–1.99**	0.63**	1.009**	3.115**	–0.924**	–0.013**	0.641**	0.6806**	0.00662**	0.00088**	0.00047**	–0.00215**	–0.00025**
$P_3 \times P_5$	2.18**	–0.70**	0.364**	3.903**	1.045**	–0.051**	0.693**	1.0251**	0.0025*	0.00003	–0.00004**	0.00583**	0.00030**
$P_3 \times P_6$	7.62**	0.17**	0.688**	6.265**	–0.044**	0.017**	0.561**	–0.0987**	0.0073**	–0.00119**	–0.00022**	0.00495**	–0.00040**
$P_3 \times P_7$	–0.03	2.66**	0.604**	0.947**	–0.065**	–0.015**	0.250**	0.2013**	0.0108**	0.00114**	0.00089**	0.00391**	0.00012**
$P_4 \times P_5$	0.40	–3.41**	1.304**	–3.314**	–0.107**	0.006**	0.363**	0.2944**	–0.0024*	–0.00021	0.00002**	–0.00135**	0.00032**
$P_4 \times P_6$	–1.43**	5.41**	2.405**	–2.285**	1.003**	–0.010**	0.609**	1.0684**	–0.0027**	–0.00186**	–0.00080**	–0.00131**	–0.00018**
$P_4 \times P_7$	–2.16**	2.28**	–1.716**	–3.191**	1.291**	0.005**	0.760**	1.0708**	–0.0019	0.00049*	0.00040**	0.00652**	–0.00019**
$P_5 \times P_6$	–6.03**	–2.32**	–1.329**	0.114	–1.083**	0.022**	0.369**	0.1491**	–0.0142**	–0.00024	–0.00092**	–0.00604**	–0.00220**
$P_5 \times P_7$	–4.33**	6.01**	–0.261**	–2.128**	0.201**	0.027**	0.537**	0.1698**	0.0073**	–0.00082**	–0.00013**	–0.00236**	0.00061**
$P_6 \times P_7$	0.15	–1.73**	1.956**	–1.126**	–0.217**	0.009**	0.297**	0.5439**	–0.0162**	–0.00224**	–0.00096**	–0.00660**	–0.00184**
S.E. (S_{ij})	2.5×10^{-1}	3.4×10^{-3}	5.3×10^{-2}	1.7×10^{-2}	2.2×10^{-2}	1.0×10^{-2}	1.8×10^{-2}	1.2×10^{-3}	6.5×10^{-4}	1.3×10^{-4}	3.9×10^{-6}	3.1×10^{-5}	3.1×10^{-6}
S.E. ($S_{ij} - S_{ik}$)	2.5×10^{-1}	5.6×10^{-3}	8.0×10^{-2}	2.3×10^{-2}	3.0×10^{-2}	1.0×10^{-2}	2.7×10^{-3}	1.9×10^{-3}	9.8×10^{-4}	2.0×10^{-4}	5.8×10^{-6}	4.8×10^{-5}	4.7×10^{-6}

Table 5. Estimation of reciprocal effect of 13 traits in 7 × 7 diallel cross of opium poppy. *, $p < 0.05$; **, $p < 0.01$; Sr_{ij}, reciprocal effect for the cross between ith and jth parent

Parents	Days to flower (50%)	Plant height (cm)	Peduncle length (cm)	Days to maturity	Number of capsules per plant	Capsule index (width / length)	Seed yield (g)	Capsule husk yield (g)	Morphine concen-tration (%)	Codeine concen-tration (%)	Thebaine concen-tration (%)	Papaverine concen-tration (%)	Narcotine concen-tration (%)
$P_2 \times P_1$	−0.533	1.545**	−0.862**	1.038**	−0.11**	0.021**	0.845**	0.963**	2.3×10^{-4}	7.0×10^{-12}	1.8×10^{-4}**	0	-6.0×10^{-4}**
$P_3 \times P_1$	0.183	−1.671**	1.760**	−1.265**	0	−0.007**	0.568**	0.538**	-5.0×10^{-4}	3.0×10^{-4}	1.6×10^{-4}**	3.1×10^{-3}**	0
$P_3 \times P_2$	−1.983**	−0.389**	1.405**	0.833**	0.665**	−0.003*	−0.565**	−0.243**	-1.2×10^{-4}	7.0×10^{-3}**	2.0×10^{-3}**	1.9×10^{-4}**	1.1×10^{-4}**
$P_4 \times P_1$	−1.955**	2.750**	4.127**	−3.168**	0.055*	−0.081**	0.588**	−0.482**	1.5×10^{-4}*	5.0×10^{-4}	-3.8×10^{-4}	2.3×10^{-4}**	0
$P_4 \times P_2$	−2.833**	−0.073	−0.962**	−13.72**	−0.112**	0.006**	−0.028	−0.348**	9.0×10^{-4}	-4.3×10^{-4}**	2.8×10^{-4}**	-1.9×10^{-4}**	0
$P_4 \times P_3$	1.817**	9.553**	2.978**	−9.110**	−0.112**	0.012**	−0.002	0.555**	1.2×10^{-4}**	2.6×10^{-4}**	6.2×10^{-5}**	2.4×10^{-3}**	0
$P_5 \times P_1$	2.467**	−1.488**	0.950**	−1.612**	0.388**	0.026**	0.125**	−0.737**	1.1×10^{-1}**	2.7×10^{-3}**	1.6×10^{-4}**	6.7×10^{-4}**	0
$P_5 \times P_2$	4.667**	1.931**	1.687**	−8.445**	−0.011	−0.017**	0.222**	−0.045**	-1.3×10^{-1}**	-8.8×10^{-4}**	-1.3×10^{-4}**	6.2×10^{-4}**	-6.3×10^{-4}**
$P_5 \times P_3$	6.833**	−1.405**	−0.612**	−12.723**	0.610**	−0.074**	0.693**	−0.580**	-1.1×10^{-1}**	-1.5×10^{-4}**	-6.8×10^{-4}**	-6.5×10^{-3}**	-3.0×10^{-4}**
$P_5 \times P_4$	0.033	1.257**	2.070**	−3.223**	0.167**	0.002*	−0.363**	−0.052**	2.0×10^{-2}**	8.1×10^{-4}**	5.1×10^{-4}**	8.6×10^{-4}**	0
$P_6 \times P_1$	1.899**	−6.667**	−2.304**	−10.833**	−0.166**	−0.014**	0.193**	−0.595**	-3.1×10^{-1}**	2.1×10^{-4}**	1.3×10^{-4}**	2.7×10^{-4}**	-1.3×10^{-2}**
$P_6 \times P_2$	4.966**	−3.415**	−0.973**	−4.965**	−0.222**	0.0047**	−0.312**	0.200**	-1.7×10^{-1}**	-2.1×10^{-4}**	-4.5×10^{-4}**	-7.8×10^{-3}**	-4.3×10^{-4}**
$P_6 \times P_3$	−0.700*	−0.797**	−1.522**	−8.277**	0.280**	0.008**	−0.561**	−0.413**	-1.7×10^{-4}**	-1.3×10^{-3}**	-5.0×10^{-3}**	-1.2×10^{-3}**	-2.6×10^{-3}**
$P_6 \times P_4$	−0.117	0.972**	2.870**	−3.110**	0.443**	−0.006**	0.6091**	−0.348**	-6.7×10^{-2}**	9.5×10^{-4}**	1.0×10^{-4}**	-1.4×10^{-3}**	3.6×10^{-4}**
$P_6 \times P_5$	1.433**	−1.863**	−0.983**	−1.888**	0	0.015**	0.369**	0.345**	2.0×10^{-2}**	-1.8×10^{-3}**	3.5×10^{-4}**	1.1×10^{-3}**	-1.5×10^{-4}**
$P_7 \times P_1$	−2.883**	−7.762**	−4.028**	−4.590**	−0.223**	−0.069**	0.150**	0.410**	-5.6×10^{-2}**	-3.1×10^{-4}*	0	-9.9×10^{-4}**	-7.6×10^{-4}**
$P_7 \times P_2$	−6.300**	−9.100**	−0.645**	0.057	0.278**	0.0140**	−0.653**	0.378**	-2.4×10^{-2}**	-2.3×10^{-4}**	2.0×10^{-3}**	-2.1×10^{-3}**	-1.4×10^{-3}**
$P_7 \times P_3$	−1.317**	−6.089**	−2.522**	−2.360**	0.167**	0.002	−0.250**	0.008	-2.4×10^{-1}**	-3.0×10^{-2}**	-1.3×10^{-3}**	-7.5×10^{-4}**	6.3×10^{-4}**
$P_7 \times P_4$	−0.380	−7.850**	−4.102**	3.056**	0.333**	0.013**	0.760**	1.161**	-3.4×10^{-2}**	1.0×10^{-2}**	4.5×10^{-4}**	7.7×10^{-3}**	0
$P_7 \times P_5$	−1.483**	−4.438**	−0.702**	7.608**	0	0.012**	−0.537**	0.040**	2.4×10^{-1}**	6.5×10^{-4}**	6.1×10^{-4}**	1.1×10^{-4}**	5.5×10^{-4}**
$P_7 \times P_6$	−3.000**	−6.055**	0.960**	6.194**	0	0.001	0.297**	−0.540**	-6.0×10^{-2}**	-2.0×10^{-3}**	0	-2.7×10^{-4}**	-1.1×10^{-3}**
S.E. (Sr_{ij})	0.287	0.0423	0.061	0.191	0.0253	0.001	0.021	0.0142	7.2×10^{-3}	1.5×10^{-5}	4.4×10^{-6}	3.6×10^{-5}	3.5×10^{-6}

Discussion

It is evident that the majority of the studied traits exhibited significant general combining ability, specific combining ability and reciprocal effect mean squares, and manifested importance of additive, non-additive gene action as well as maternal effects. Earlier studies also reported that in opium poppy both additive and non-additive genetic variation played important roles in the inheritance of poppy latex and straw alkaloid content (Singh et al. 2011; Lal et. al 2014; Shukla et al 2019). Both additive and non-additive genetic components of variance were reported to govern the expression of opium yield and physiological characters (Kandalkar et al. (1992). Early flowering is a desirable character in opium poppy, as it provides sufficient time for seed filling in capsules, which could help in achieving increased seed yield. Similarly, medium plant height prevents lodging of crops, while characters like early maturity helps crops to escape pest losses and unwanted damage> Thus, these characters should be taken into account for negative combing ability. Taller plants are susceptible to lodging, and therefore, medium or short-statured plants are desirable, and for that negative GCA and SCA values are preferred (Singh, Khanna 1993; Singh et al. 2004; Ali et al. 2015). Hence, negative combining ability effects for early days to flowering and early maturity are required to get higher yields. For days to flowering, P_1, P_3 and P_7 showed negative GCA and their hybrids showed high negative SCA, supporting that these three parents are good combiners. Similar findings for GCA were reported for days to flowering, capsule number and latex yield (Dubey et al. 2007). For plant height, P_3, and P_7 had negative GCA and their hybrids also showed significant and high negative SCA and significant reciprocal effect. P_1, P_2 and P_3 showed negative significant GCA for days to maturity and most of the hybrids involving these parents had significant SCA and reciprocal effect. Higher seed yield is the priority of breeding programmes, and hence improvement in yield and its components are prerequisites. Desirable GCA, SCA and reciprocal effects were recorded in selected parental genotypes and their hybrids, respectively. Therefore, positive combining ability effects were considered desirable for seed yield. P_1, P_2, P_5 and P_6 showed positive GCA while P_3, P_4 and P_7 had negative GCA, but SCA was significant for crosses low in GCA, showing that the high × low GCA parents were involved in best specific combinations. Our results were in accordance with the findings of earlier studies with opium poppy (Sharma et al. 1988; Yadav et al. 2014b; Kumar et al. 2014; Lal et al. 2014).

In relation to medicinally important alkaloids in the final tally, positive GCA was evident for parents P_3, P_5, P_6 & P_7 (morphine); P_3, P_6 & P_7 (codeine); P_1, P_4, P_5 & P_6 (thebaine); parents, P_1, P_3 & P_6 (papaverine) and P_6 (narcotine). Among all parents, genotype P_6 had the most desirable level of GCA, SCA and reciprocals for most of considered agronomic and biochemical traits, an consequently it is most important isolate in this study that could be utilized in a variety of ways.

In conclusion, diallel analysis is a useful tool in identification of parents for hybrid combination. The majority of the traits exhibited significant GCA and SCA mean squares, and manifested importance of additive, non-additive gene action and maternal effects. The identified genotypes P_3, P_4 and P_6 are important in relation to seed yield, development of morphine-less cultivars and quantitative alkaloid yield in capsule husk, respectively, and can be utilized for genetic improvement programmes to obtain desired traits of *P. somniferum*.

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