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Association between molecular diversity and hybrid performance in sunflower (*Helianthus annuus* L.)

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Abstract

The present study was aimed to assess the molecular genetic diversity in 14 parental lines (10 maintainers and 4 restorers) of sunflower crop using 51 SSR markers. A total of 147 alleles were produced with the range of 1-6 alleles per locus. The polymorphic information content (PIC) values ranged from 0.12(ORS1198 in LG5) to 0.74(ORS384 in LG15) with an average PIC of 0.51. A phylogenetic tree was constructed by neighbour- joining method which grouped the parents into four clusters. Among the maintainer and restorer combinations, the dissimilarity index was maximum between CMS 519B and CSFI 99 (0.67) which indicateed high divergence between them. Lower dissimilarity value was observed between RCR CMS 38B and LTRR 341 (0.40) suggested less divergence between them. Mahalanobis D² analysis was also carried out among the parental lines. The D² analysis grouped the parental lines into six clusters. Among them genotype pairs CMS104B vs RHA 1-1 and CSFI 99 vs LTRR341 had highest and lowest D² values respectively. Dissimilarity and D² values for parental combinations, mean performance, mid and better parental heterosis of 40 hybrids for yield and its component traits were subjected to simple correlation analysis. The D² analysis did not show any association with performance of hybrids. However, the dissimilarity values showed association with hybrid performance and heterosis for plant height and volume weight. Hence, the present study indicated that the selection of parental genotypes with high molecular diversity could result in improved performance of plant height and volume weight in sunflower hybrids. However further study is required with multi environment data sets.

Keywords: Sunflower, molecular diversity, hybrid, association, heterosis.

INTRODUCTION

Sunflower is an important oilseed crop in the world. Sunflower competes in the world oilseed complex with the other major oilseed crops such as soybean, groundnut, and rapeseed. It has about 46-52% oil content (Das *et al.*, 2020). Ever since sunflower was introduced in 1969 in India, as a supplement to traditional oilseed crops, its area has shown a steady increase and presently, India has emerged as the second major sunflower producing country in Asia after China. It is a cross pollinated crop. The commercial success of the production was archived by hybrid breeding. Hybrid breeding in Sunflower was started economically after the discovery of CMS by

Leclercq in 1960 and restorer genes by Kinman in 1970 (Fick and Miller, 1997). and currently, hybrid breeding program is well developed (Chandirakala *et al.*,2016; Nichal *et al.*, 2017; Ghaffari and Shariati, 2018; Hilli *et al.*,2020; Karande *et al.*, 2020).

The choice of the parents is a crucial step in the heterosis breeding program. The genetic diversity of parental lines is an important criterion in the manifestation of heterosis (Melchinger, 1999). The genetic diversity of plant material can be observed from morphological as well as molecular data. D² statistic is a powerful tool for

the identification of genetically diversified parents which can be used in hybridization programs (Punitha *et al.*, 2010). DNA markers *viz.*, RFLP, RAPD, SSR, ISSR *etc.*, can be used to assess the diversity studies. Among them, SSR markers have great potential in genetic and breeding studies (Shoba *et al.*, 2010; Ramanaiah and Kadirvel, 2021).

In recent years, molecular method of diversity research has been conducted in most of oilseed crops by SSR markers (Ramanaiah and Kadirvel, 2021; Yihan *et al.*, 2022; Ahmed *et al.*, 2022 in sunflower, Teklu *et al.*,2022 in sesame, Tomar *et al.*, 2022 in groundnut and Kumar *et al.*, 2022 in soybean) due to its codominant and highly reproducible characteristics. The relationship between molecular diversity with hybrid performance and heterosis was studied in several crops to access the reliability of markers-based prediction in hybrid performance (Xiao *et al.*, 1996; Sureja *et al.*, 2006; Gupta *et al.*, 2018 and Somashekhar *et al.*, 2020). Hence, an attempt was made to study the molecular genetic diversity among parental lines and their association with the hybrid performance in sunflower.

MATERIALS AND METHODS

This research work consists of 14 parents which includes ten maintainer lines viz., COSF 6B, COSF 10B, COSF 12B, COSF 13B, CMS 104B, CMS 207B, CMS 519B, ARM 248B and RCR CMS 38B and four restores viz., IR 6, CSFI 99, RHA-1-1 and LTRR341. A field experiment was carried out at the Department of Oilseeds, Tamil Nadu Agricultural University, Coimbatore (11° N latitude and 77° E longitude). Forty hybrids were produced from these parents using a line x tester mating design. During July-September season, 2021, 40 hybrids and their parents were evaluated in a randomized complete block design with two replications. Morphological observations were taken on nine quantitative traits viz., days to 50% flowering, plant height (cm), head diameter (cm), days to maturity, volume weight (g/100ml), hundred seed weight (g), seed yield /plant (g), oil content (%) and oil vield/plant (g) with randomly selected five plants in each entry in each replication. The oil content of the seeds was estimated by using a Near-infrared (NIR) spectroscopy instrument (Make: M/s ZEUTEC, Germany; Model: SPA 1.0) available at the Centre of Excellence in Molecular Breeding, TNAU, Coimbatore.

The DNA was extracted from young leaves of 14 parents by following CTAB method (Doyle and Doyle, 1987). The DNA quality and quantity were checked on 0.8% agarose gel, and DNA concentration was normalized to 10 ng/μ l.

In the present study, publicly available ORS markers mapped by Tang *et al.* (2002) were used. A total of 51 SSR primers were used for analyzing the diversity among parents. These markers were well distributed among the 17 linkage groups (LGs) in sunflower at the rate of 3 to 5

markers per LG. The Polymerase Chain Reaction (PCR) mixtures were prepared as per Subramaniyan *et al.* (2022). The DNA was amplified in a thermocycler (Applied Biosystems, Veriti) under the following conditions: Initially single cycle of 94°C for 3 min followed by touch down step (0.5° C decrease in the annealing temperature after each cycle) (Hube *et al.* 2005) in 20 cycles (of 94°C for 30 s, 63°C for 30 s, 72°C for 1 min) followed by 20 cycles of 94°C for 15 s, 55°C for 30 s, 72°C for 1 min with a final extension step of 72°C for 10 min. The PCR products were separated on 3% agarose gel and photographed using GELSTAIN 49 advanced gel documentation unit (M/s Medicare, India).

In each parent, the amplified bands of 51 markers were scored as present (1) or absent (0) for each unique allele. Polymorphism information content was estimated for each marker using the formula suggested by Botstein *et al.* (1980). The allelic data was subjected for estimating the genetic distance. A neighbour-joining tree (Saitou and Nei, 1987) was created using the DARwin 6.0 software package (Perrier and Jacquemoud-Collet, 2006). Data on the 14 parental lines were subjected into D² analysis as suggested by Mahalanobis (1928) and clustered by Tocher method as suggested by Rao (1952).

The mean value of nine biometrical traits of 40 hybrids and dissimilarity values and D² values of 40 parental combinations were used for correlation analyses. Simple correlation analysis was performed as per the standard method (Pearson, 1896) with MS Excel.

RESULTS AND DISCUSSION

In the present study, 51 markers were taken for molecular diversity analysis in a set of 14 sunflower parental lines (10 maintainer lines and 4 restorer lines). All the linkage groups had three polymorphic markers except the LG8 and LG10 which had two polymorphic markers (Table 1). A total of 147 alleles were produced from 51 SSR markers. The number of bands produced per primer ranged from 1 to 6 with an average of 2.88 alleles. The Polymorphic Information Content (PIC) values ranged from 0.12 (ORS1198 in LG5) to 0.74 (ORS384 in LG15) with an average PIC of 0.51 (Zia et al., 2014; Zeinalzadeh-Tabrizi et al., 2018). The monomorphic markers (ORS762 in LG8; ORS1130 in LG10) had zero PIC values. The highest PIC value (0.74) was recorded by the polymorphic markers viz., ORS384 and ORS565 and they were located in LG15 and LG17 respectively. Hence, these markers are highly suitable for distinguishing individuals and understanding the genetic diversity among them (Darvishzadeh et al., 2010; Zeinalzadeh-Tabrizi et al., 2018). The genetic dissimilarity indices ranged from 0.30 (COSF 12B vs COSF 10B) to 0.68 (RHA-1-1 vs CSFI 99) (Table 2). High dissimilarity indices were recorded between CSFI 99 vs RHA-1-1 (0.68) followed by CMS 519B vs CSFI 99 (0.67), CSFI 99 vs IR 6 (0.67) and CMS 104B vs CSFI 99 (0.66) which indicated high

S.No	Markers	LG	Sequences	Allele size (bp)	Number of alleles	PIC	Remark
1	ORS822	1	F: CAATGCCATCTGTCATCAGCTAC	140-170	3	0.55	Polymorphic
			R: AAACAAACCTTTGGACGAAACTC				
2	ORS965	1	F: TTGGATTACCTTGGATAGTCAGC	240-700	3	0.58	Polymorphic
			R: CTTACCCTCCTCAGACCCTACCT				
3	ORS543	1	F: CCAAGTTTCAGTTACAATCCATGA	300-310	2	0.27	Polymorphic
			R: GGTCATTAGGAGTTTGGGATCA				
4	ORS1264	2	F: TAGAAGCGGTTGGGTTGACAGTA	260-400	3	0.51	Polymorphic
			R: TGAACTCGGTTGATTCTCTAGCC				
5	ORS1152	2	F: CCCAAGCTCCCTCTTCATCTTA	250-270	3	0.58	Polymorphic
			R: TGTTCATAGCCTCATTCATGTTG				
6	ORS1073	2	F: AGAGTGTTGGTCTTGATTTGTGG	100-200	3	0.58	Polymorphic
			R: AAAGAAAGGCGCACGTTAGTC				
7	ORS1040	3	F: CTGCTGATCGTTTCTTGGATAGA	200-210	2	0.25	Polymorphic
			R: TGCTAATCCTTCTAATCAACTTCCAC		_		
8	ORS1080	3	F: TTTGTTTCTTTGTGTGGTTATCG	400-1000	2	0.38	Polymorphic
	000/00		R: GGGTGTTGTCGGAGGTATAAAGT				
9	ORS488	3		190-450	2	0.29	Polymorphic
4.0	0004407			000 070	0		
10	ORS1197	4		200-370	3	0.41	Polymorphic
44	000550	4		200.240	2	0.00	Delume empleie
11	085558	4		300-310	2	0.23	Polymorphic
10	000224	4		220 200	2	0.45	Dolumorphia
12	0K3334	4		230-300	3	0.45	Polymorphic
12	0001100	5		140 150	2	0 1 2	Dolumorphia
15	0K31190	5		140-150	Z	0.12	Polymorphic
1/	0851024	5		200-350	1	0.65	Polymorphic
14	0101024	5		200-330	4	0.05	i olymorphic
15	ORS 1159	5	F TTTCGTGATGGTGATGATGATG	200-260	3	0.53	Polymorphic
10		Ũ	R ⁻ CAGCAACTCTGACCGTTTCATTA	200 200	0	0.00	i olymorphie
16	ORS1193	6	F: GACGGTGATGGATGATAAAGAGA	100-210	5	0.60	Polymorphic
		-	R: TTATTGTCACTCATCGAGAAATTGA		-		
17	ORS1219	6	F: CAAACTCTTGCATGTTATGTCCTTT	160-170	2	0.36	Polymorphic
			R: GATCTATCTCAACCACCACTGATCT				
18	ORS725	6	F: TCCGACGACCAAAGAAACTT	490-510	3	0.57	Polymorphic
			R: CACAATGAAGGGAAATGGAGA				
19	ORS1259	7	F: ACCCATGTTGATAGCCAACCTT	140-500	5	0.72	Polymorphic
			R: TAGATTCCGAGGTGTGAGGGTAT				
20	ORS331	7	F: TGAAGAAGGGTTGTTGATTACAAG	300-400	3	0.59	Polymorphic
			R: GCATTGGGTTCACCATTTCT				
21	ORS328	7	F: GACCTGTAGGCCAATATGAGACTT	200-270	2	0.37	Polymorphic
			R: TTATACCGGTGTTGTATCGTATCC				
22	ORS185	8	F: AGCCGCTCCTAACTTGAACCA	290-320	2	0.37	Polymorphic
			R: TCACCCTTAAACATCACCCACC				
23	ORS780	8	F: TGATTACAACCCTAATTCGCATAC	100-290	4	0.69	Polymorphic
			R: GATACTGGTGGGACAGATGTTG				
24	ORS762	8	F: TGCACATGAGGGTATTCTTGTC	210	1	0.00	Monomorphic
			R: TCGAGGAGAGTGTGACGTTG				
25	ORS805	9	F: CATGGATTATAAGAACGGGTGTT	320-600	2	0.37	Polymorphic
		-	R: AA ICCCAGGGGTAAAATTGC			• • •	
26	ORS739	9	H: CICCGICGCGTATGATAATG	120-190	3	0.43	Polymorphic
			R: CAAGAAGT (GTTCACTCTTGATCC				

Table 1. List of SSR markers used for diversity analysis in sunflower

Table1. Continued..

27 ORS1265 9 F: GGGTTTAGCAATAATAGCGACA 190-220 2 0.37 Polymorphic 28 ORS1110 10 F: CATTCAAGGGGCTATTGTGTAAG 200-600 2 0.15 Polymorphic 29 ORS437 10 F: GACTCGACTTCAATAACG 320-550 3 0.22 Polymorphic 30 ORS1130 10 F: ACACCAACATCACAACAACAACAACACACACC 230 1 0.00 Menomorphic 31 ORS621 11 F: CCCCATTGGGGATCCATATAACG 230 0.84 0.64 Polymorphic 320 ORS930 11 F: CCCCATCAGGGAACAACATCACACGC 230 0.38 Polymorphic 33 ORS630 11 F: CCCCAACACATATATCACCCTTTC 130-140 2 0.38 Polymorphic 34 ORS520 12 F: ATCCCAACAGGCACATATATTCACCCTT 100-270 2 0.35 Polymorphic 35 ORS538 12 F: CACTGAACTCACACTCACACCT 270-300 2 0.37 Polymorphic 36 ORS559	S.No.	Markers	LG	Sequences	Allele size (bp)	Number of alleles	PIC	Remark
R. ACCCT IDAGE IT INSIGAT IT AGE 20-600 2 0.15 Pelymorphic 29 ORS110 10 F: CACTGAAGGCGCATTGTGTAAG 220-500 3 0.22 Polymorphic 30 ORS1130 10 F: AGGCATCACATCAACAATCAACAGC 230 1 0.00 Monomorphic 31 ORS6130 10 F: AGGCAACAACATCAACAATCAACAGC 230 1 0.00 Monomorphic 32 ORS9101 F: CGCATTGGTGAGGAAAA 180-380 4 0.64 Polymorphic 33 ORS630 11 F: GGGCAACTATATTGCTGAGGAGAAA 130-350 2 0.13 Polymorphic 34 ORS502 12 F: ACCATGACCGGAATAT 100-270 2 0.35 Polymorphic 35 ORS358 12 F: CACTACCCACACACACATCATC 2 0.37 Polymorphic 36 ORS359 12 F: CACTGCACTCATCCCT 270-300 2 0.37 Polymorphic 37 ORS573 13 F: TGGCACTTATCCCACTCATC 20-210 2 2	27	ORS1265	9	F: GGGTTTAGCAAATAATAGGCACA	190-220	2	0.37	Polymorphic
28 ORST110 TO FLOATIGANGSGGCIATATGATAG 200-800 2 0.15 Polymorphic 29 ORS437 10 F: GACGTCTTCGCAGGTCGATCGA 320-550 3 0.22 Polymorphic 30 ORS1130 10 F: AGCACAACATCAACAATCAAGC 230 1 0.00 Monomorphic 31 ORS621 11 F: CCGCATCGGGATCATATATAAC 7 0.38 Polymorphic 32 ORS980 11 F: GCGGAACATACTCCCTTGATGTT 130-140 2 0.38 Polymorphic 33 ORS630 11 F: GCGGAACATACTCCCCTTGATGTT 100-270 2 0.38 Polymorphic 34 ORS502 12 F: ATCCCAACGACGCCATTAT 100-270 2 0.37 Polymorphic 35 ORS559 12 F: CACTGAACCTACACCACTCCTTG 70-300 2 0.37 Polymorphic 36 ORS559 12 F: CACTGAACCTACACTCTTGTA 160-500 4 0.68 Polymorphic 37 ORS673 13 F: TGGTGCTATCCCA	00	0004440	10		000 000	0	0.45	Dahmannikia
R: BGTTTIGBAGAAGBICGATICATACC 320-550 3 0.22 Polymorphic 29 0RS1130 10 F: AGGCATCACATCAACATCAACGC 230 1 0.00 Monomorphic 30 0RS1130 10 F: AGGCATCACATCAACAATCAACACC 230 1 0.00 Monomorphic 31 0RS621 11 F: CGCATCGGGATCATTATACC 230 1 0.00 Monomorphic 32 0RS990 11 F: GGGAAACTCCTCCCTGATGGT 130-140 2 0.38 Polymorphic 33 0RS630 11 F: TGTGGTGAGGATATTCCACTTTC 3 0.85 Polymorphic 34 0RS502 12 F: CACGACCGGATATTAT 100-270 2 0.35 Polymorphic 35 0RS503 12 F: CAATGAATGCACGAAAGGTC 110-200 3 0.58 Polymorphic 36 0RS579 13 F: TGGTGCTTACACGAGAAGGTC 110-200 3 0.58 Polymorphic 37 0RS673 13 F: TGGTGCTACCCACTGACACGG 160-500 4 <td>28</td> <td>ORS1110</td> <td>10</td> <td></td> <td>200-600</td> <td>2</td> <td>0.15</td> <td>Polymorphic</td>	28	ORS1110	10		200-600	2	0.15	Polymorphic
29 ORS437 10 F: GACGITICITACTCTCG 320:550 3 0.22 Polymorphic 30 ORS1130 10 F: AGCAACACATCAACAATCAAGC 230 1 0.00 Monomorphic 31 ORS621 11 F: CGCATCGAGCTAATTAAAC 230 1 0.00 Monomorphic 32 ORS990 11 F: CGCACTGAGCTAGATTAACC 2 0.13 Polymorphic 33 ORS630 11 F: TOTCGTCAGGGGAGCATAT 100-270 2 0.37 Polymorphic 34 ORS502 12 F: ATCCCAACAGACGGAGCATA 100-270 2 0.37 Polymorphic 35 ORS530 12 F: CACTGACTCACATCACTC 270:300 2 0.37 Polymorphic 7 TOTGCACACGAGAGAGACTCATTA 100-270 2 0.22 Polymorphic 35 ORS539 12 F: CACTGACTCACATTCACACGCTTA 270:300 2 0.37 Polymorphic 36 ORS573 13 F: TOTGCACTTTACACCTTTTA 200:210 <td< td=""><td></td><td>000/00</td><td></td><td>R: GGIIIIGGAGAGAGGICGAIGIG</td><td></td><td></td><td></td><td></td></td<>		000/00		R: GGIIIIGGAGAGAGGICGAIGIG				
Bit	29	ORS437	10	F: GACGICIICACAGIICAAAIAACG	320-550	3	0.22	Polymorphic
30 ORS1130 10 F: AGCAACAACATCAAGC 230 1 0.00 Monomorphic 31 ORS621 11 F: CGCACTGGGGATCCTATTAAAC 180-380 4 0.64 Polymorphic 32 ORS990 11 F: GGGAACTTACTCCCTGATGTT 130-140 2 0.38 Polymorphic 33 ORS630 11 F: TGTGCTOAGGGAGGAGGAGGAG 310-350 2 0.13 Polymorphic 33 ORS630 11 F: TGTGCTOAGGGGAGCATA 100-270 2 0.35 Polymorphic 34 ORS550 12 F: CACTGACCGAGAGGCCATA 100-270 2 0.37 Polymorphic 35 ORS558 12 F: CACTGACAGCAGACAGTC 110-200 3 0.58 Polymorphic 36 ORS559 12 F: CACTGACGTCATCTTCA 110-200 3 0.58 Polymorphic 37 ORS673 13 F: TTGGGCACTCATCTTCA 200-210 2 0.22 Polymorphic 38 ORS17 13 F:				R: GCATCGACTCTGTTCTTCTCG				
H: CCCATICGGGATCCTATTATACAC 11 F: CCCCTTATCTGAGAGGAAA 180-380 4 0.64 Polymorphic 32 ORS920 11 F: CCCCTTGAGAGGAAA 180-380 4 0.64 Polymorphic 32 ORS930 11 F: CCCCTTGAGGATGATAGCAG 310-350 2 0.38 Polymorphic 33 ORS502 12 F: ATCCCAACAGAGAGCCATTAT 100-270 2 0.35 Polymorphic 34 ORS502 12 F: ATCCCAACAGCAACGCAATGCT 100-270 2 0.35 Polymorphic 35 ORS559 12 F: CACTGACCTCAACGCATGCT 270-300 2 0.37 Polymorphic 36 ORS559 12 F: CACTGACACTAATCCACGCTTCAT 110-200 3 0.58 Polymorphic 37 ORS570 13 F: TGGTGCTATCCACCTTCAT 200-210 2 0.22 Polymorphic 38 ORS570 13 F: TGGTGATTGCTGACTCACCTTCAT 200-210 2 0.22 Polymorphic 39 ORS707 13<	30	ORS1130	10	F: AGCAACAACATCAACAATCAAGC	230	1	0.00	Monomorphic
31 ORS21 11 F: CGCTTATGCTGAGAGGAAA 180-380 4 0.64 Polymorphic 32 ORS90 11 F: GGGAAACTACTCCTCGATGTT 130-140 2 0.38 Polymorphic 33 ORS830 11 F: GGGACACTATTATCCACCACTTTC 130-140 2 0.38 Polymorphic 34 ORS502 12 F: ATCCCAACGGGCATTAT 100-270 2 0.35 Polymorphic 7 R: GCACGACCCGGATATGTAAC 20-37 Polymorphic R: ACATTGGAGGGGGGCCAATA 20-37 Polymorphic 36 ORS59 12 F: CACTGACCTCACACTGTTGC 270-300 2 0.37 Polymorphic 7 R: GTCGTATTATCTCCCCTCTTGC 110-200 3 0.58 Polymorphic 7 R: GGTCGTATGCTACCATCTTGTA 200-210 2 0.22 Polymorphic 7 R: GGTCGTATGCTAACTGAGCATCG 150-170 2 0.38 Polymorphic 7 R: GCGACATGGGAGCAGCA 160-200 3 0.57 Polymorphic 7				R: CCCACTCGGGATCCTATATAAAC				
Sec Construction Construct	31	ORS621	11	F: CGCCTTATGCTGAGAGGAAA	180-380	4	0.64	Polymorphic
32 ORS99 11 F: GGGAAACTTACTCCCTTGATT 130-140 2 0.38 Polymorphic 33 ORS630 11 F: TGTGCTGAGGATGATATGCAAC 310-350 2 0.13 Polymorphic 34 ORS502 12 F: ATCCCAACGACGCCATTAT 100-270 2 0.35 Polymorphic 35 ORS508 12 F: CACTGACCCGACCTCATCT 270-300 2 0.37 Polymorphic 36 ORS559 12 F: CACTGACCCCACACTCACTTG 2 0.38 Polymorphic 37 ORS673 13 F: TGGGCATCTCCACCCTTCCTC 10-200 3 0.58 Polymorphic 38 ORS317 13 F: TTGGCACTTGCACCGTTCA 200-210 2 0.22 Polymorphic 39 ORS317 13 F: TTGGCACTTGAACCCCG 150-170 2 0.38 Polymorphic 39 ORS707 13 F: CCTGAACCGATATCG 100-170 2 0.38 Polymorphic 30 ORS578 14 F: CCTGGAACCAGACAC <td></td> <td></td> <td></td> <td>R: CCTGAAGCGAAGAAGAATCG</td> <td></td> <td></td> <td></td> <td></td>				R: CCTGAAGCGAAGAAGAATCG				
33 ORS630 11 F: TGTGCTGAGGATGATATGCAG 310-350 2 0.13 Polymorphic 34 ORS502 12 F: TGTGCTGAGGAGGATGATAGCAG 100-270 2 0.35 Polymorphic 35 ORS581 12 F: CACTGACCTCACACTOATGCT 270-300 2 0.37 Polymorphic 36 ORS559 12 F: CACTGACCTCACACCTCATGCT 270-300 2 0.58 Polymorphic 37 ORS559 12 F: CATGAATGCACCAAAAGGTC 110-200 3 0.58 Polymorphic 38 ORS573 13 F: TGGGCACTCCATCCTGA 160-500 4 0.68 Polymorphic 39 ORS707 13 F: TGGGCAGTTAATTCTTTCTCT 200-210 2 0.22 Polymorphic 39 ORS707 13 F: CTGGAAGCTGAAGCAGGA 150-170 2 0.38 Polymorphic 30 ORS78 14 F: CCTGAACGCAAAGGAGAGGA 10-130 2 0.26 Polymorphic 30 ORS812 15	32	ORS990	11	F: GGGAAACTTACTCCCTTGATGTT	130-140	2	0.38	Polymorphic
33 ORS630 11 F: IGTECTGAGGATGATACCG 310-350 2 0.13 Polymorphic 34 ORS502 12 F: ATCCCAAGACCGGGATATAT 100-270 2 0.35 Polymorphic 35 ORS58 12 F: CACTGACCTGACATGAT 110-200 3 0.58 Polymorphic 36 ORS59 12 F: CATGACCTGACATCATGCT 270-300 2 0.37 Polymorphic 37 ORS673 13 F: TGGGGATATTCCTCACCTTCAT 110-200 3 0.58 Polymorphic 38 ORS317 13 F: TTGGCAGTTGCTTAATTCCTCTCT 200-210 2 0.22 Polymorphic 39 ORS707 13 F: GCAGTGACACAGAGACCC 2 0.38 Polymorphic 40 ORS578 14 F: CTCTCAATCCTAAGCATCG 120-210 2 0.22 Polymorphic 41 ORS694 14 F: CCTGGAACAGAGACCC 200-600 4 0.6 Polymorphic 42 ORS307 14 F: CCTGGAACAGAGAGAC <td></td> <td></td> <td></td> <td>R: GCGACACTATIATITCACTCACTTC</td> <td></td> <td></td> <td></td> <td></td>				R: GCGACACTATIATITCACTCACTTC				
34 ORS502 12 F: ATCCCAACCCGCATTAT 100-270 2 0.35 Polymorphic 35 ORS358 12 F: CACTGAACCTCATGCT 270-300 2 0.37 Polymorphic 36 ORS559 12 F: CACTGAACCTCATGCT 270-300 2 0.37 Polymorphic 36 ORS559 12 F: CAATGAATGCACGAAAGGTC 110-200 3 0.58 Polymorphic 37 ORS673 13 F: TIGGTGCATATCCCTCCTTGA 160-500 4 0.68 Polymorphic 38 ORS517 13 F: TIGTGGCATTTCTCACCGTTGA 200-210 2 0.22 Polymorphic 39 ORS707 13 F: GCAGTCAATTGCTAAGCATGG 150-170 2 0.38 Polymorphic 39 ORS578 14 F: CCTGGAAGTGAGACAACCC 2 0.28 Polymorphic 41 ORS694 14 F: CCTGGAACTGAACCAGAGAGAC 160-200 3 0.57 Polymorphic 42 ORS307 14 F: CCAGGAGGAAAC <td>33</td> <td>ORS630</td> <td>11</td> <td>F: IGIGCIGAGGAIGAIAIGCAG</td> <td>310-350</td> <td>2</td> <td>0.13</td> <td>Polymorphic</td>	33	ORS630	11	F: IGIGCIGAGGAIGAIAIGCAG	310-350	2	0.13	Polymorphic
34 ORS502 12 F: ATCCCAACGACGCCCATTAT 100-270 2 0.35 Polymorphic 35 ORS358 12 F: CACTGACCTCACACTCATGCT 270-300 2 0.37 Polymorphic 36 ORS559 12 F: CACTGACCTCACACTCATGCT 270-300 2 0.37 Polymorphic 37 ORS673 13 F: TGGTGCTACTCCATCTTGA 160-500 4 0.68 Polymorphic 38 ORS317 13 F: TTTGGCAGTTGGTGCTTA 200-210 2 0.22 Polymorphic 39 ORS707 13 F: GCAGTAGCTAAGCATACCT 200-210 2 0.22 Polymorphic 39 ORS707 13 F: GCAGTAGCTAAGCATACC 150-170 2 0.38 Polymorphic 40 ORS578 14 F: CCTGGAACGACAGAGTCC 200-600 4 0.6 Polymorphic 41 ORS694 14 F: CCTGGAACCAACAGAGGGA 110-130 2 0.26 Polymorphic 42 ORS307 14				R: GCACGACCCGGATATGTAAC				
B: AACATTGGAGGGAGCCAATA 35 ORS358 12 F: CACTGACCTCACACTTGGT 270-300 2 0.37 Polymorphic 36 ORS559 12 F: CACTGACCTCACCTTCAGC 110-200 3 0.58 Polymorphic 37 ORS673 13 F: TGGTGCTACTCCATCCTCTCT 160-500 4 0.68 Polymorphic 38 ORS317 13 F: TTTGGCAGTTGGTGGCTTA 200-210 2 0.22 Polymorphic 39 ORS707 13 F: GCAGTATTCGTAGCACTCG 150-170 2 0.38 Polymorphic 7 GGTGGAAGCGAGAGAGAGCC 160-200 3 0.57 Polymorphic 7 CGTGGAAGCGGAAGAGAGAGGA 110-130 2 0.26 Polymorphic 41 ORS381 15 F: TGGCAGTGAAGAGAGAGGA 110-130 2 0.26 Polymorphic 42 ORS381 15 F: CCGGGAAAGAGAGAGGAGA 110-130 2 0.26 Polymorphic 8 GCGGGGATGAATGAGAGAGAGAGAGAGAGA 110-130 <td< td=""><td>34</td><td>ORS502</td><td>12</td><td>F: ATCCCAACAGACGCCATTAT</td><td>100-270</td><td>2</td><td>0.35</td><td>Polymorphic</td></td<>	34	ORS502	12	F: ATCCCAACAGACGCCATTAT	100-270	2	0.35	Polymorphic
35 ORS358 12 F: CACTGACCTCACCCATGCT 270-300 2 0.37 Polymorphic 36 ORS559 12 F: CAATGAATGCACGAAAGGTC 110-200 3 0.58 Polymorphic 37 ORS673 13 F: TGGGCATCCTCCATCCTTGA 160-500 4 0.68 Polymorphic 38 ORS317 13 F: TTTGGCAGTTTGGTGCTTA 200-210 2 0.22 Polymorphic 39 ORS707 13 F: GCAGTCATCCTTGACTC 200-600 4 0.6 Polymorphic 7 ORS578 14 F: CCTGAAGCTGAAGCACGATCC 2 0.38 Polymorphic 40 ORS578 14 F: CCTGGAACTGAAGCAGAACC 160-200 3 0.57 Polymorphic 41 ORS694 14 F: CCTGGAACCGAGAAC 110-130 2 0.26 Polymorphic 8 CCGSTGGAACTGAAGAGGGGAGAT 110-130 2 0.26 Polymorphic 42 ORS312 15 F: TTGCACATGAGGGTGGTT 130-500 6 <td></td> <td></td> <td></td> <td>R: AACATTGGAGGGAGCCAATA</td> <td></td> <td></td> <td></td> <td></td>				R: AACATTGGAGGGAGCCAATA				
36 ORS559 12 F: CAATGAAGGACGAAGGTC 110-200 3 0.58 Polymorphic 37 ORS673 13 F: TGGGCACATTACTCCCATCCTTGA 160-500 4 0.68 Polymorphic 38 ORS317 13 F: TTGGCACTTGGTGCTCAT 200-210 2 0.22 Polymorphic 38 ORS707 13 F: GCGGAGGCTAATCGTGGCTTA 200-210 2 0.38 Polymorphic 39 ORS707 13 F: GCGGAGGCGAAGGCGG 150-170 2 0.38 Polymorphic 40 ORS578 14 F: CTCCAATCCCTAAAGCCCGT 200-600 4 0.6 Polymorphic 8: GCGGAGACGGAGACCCC 10 CCGGAGACGAACCGAGGACC 160-200 3 0.57 Polymorphic 41 ORS694 14 F: CCGGGAACGAGGAGA 160-200 3 0.47 Polymorphic R: GCGGTGAACCGAGAGGAC 180-800 3 0.47 Polymorphic R: CCGGTGGAACGAGGAGGA 42 ORS812 15 F: TTCGCAGAGAGGAGGAGGA 110-130 2 0.26 Polymorphic R: CCGGTGGTCGAATAACGGGGAG	35	ORS358	12	F: CACTGACCTCACACTCATGCT	270-300	2	0.37	Polymorphic
36 ORS559 12 F: CAATGAATGCACGAAAGGTC 110-200 3 0.58 Polymorphic R: TCTGCACATTATCTCCCTCTCTC 37 ORS673 13 F: TGGTGCTACTCCATCTTGA 160-500 4 0.68 Polymorphic R: GGCATGTTCTCACCGTTCAT 38 ORS317 13 F: TTTGGCACTTTGGTGGCTTA 200-210 2 0.22 Polymorphic R: GGTGATGCTTAATTCTTCTCT 39 ORS707 13 F: GCAGTCATTCGTAGCATCG 150-170 2 0.38 Polymorphic R: GCTGAAGCTGAAGACAGATCC 40 ORS578 14 F: CTCCAAAGCCGAAACCAGATCC 200-600 4 0.6 Polymorphic R: TGGTGGAATGTGGTTGTGAT 41 ORS694 14 F: CCTGGAACTGAACCAAAGGGAAC 160-200 3 0.57 Polymorphic R: GCAGTGAAACAGAGAGGG 42 ORS307 14 F: CAGTTCACTGAAGCAGGAGGG 110-130 2 0.26 Polymorphic R: GCAGTGAAGAGGGAGAGGG 43 ORS812 15 F: TTGGAAAGAGGGGAGATG 130-500 6 0.74 Polymorphic R: CCAGTGAGGGGAGGTGCTT 130-500 6 0.74 Polymorphic R: CCAGTGGAGCGTGTA				R: GTTCTGTTCCTTACTCAGCTTTG				
37 ORS673 13 F: TGGGCATATTCCCCTCCTGA 160-500 4 0.68 Polymorphic 38 ORS317 13 F: TTGGCAGTTGGTGGCTTA 200-210 2 0.22 Polymorphic 38 ORS317 13 F: TTGGCAGTTGGTGGCTTA 200-210 2 0.22 Polymorphic 39 ORS707 13 F: GCGAGAGCTGAAGCAGATCC 150-170 2 0.38 Polymorphic 40 ORS578 14 F: CTCTCAATCCCTAAGTCCCCT 200-600 4 0.6 Polymorphic 7 TGGTGAAGTGGAGTGGGTTGTTGAT TCTGGAACTGAACCGAGAGAG 160-200 3 0.57 Polymorphic 8: GCGGTGAAACAGAGAGAGAGAGA 110-130 2 0.26 Polymorphic 8: GCGGTGGTCGAAACAGAGAGAGAGAGA 110-130 2 0.26 Polymorphic 8: CGRS307 14 F: CAGTTCCCTGAAACGAGAGAGAGA 180-800 3 0.47 Polymorphic 8: CGRS384 15 F: TTCGACATGAGAGGGTGCTT 130-500 6 0.74 Polymorphic 45 ORS668 15 F:	36	ORS559	12	F: CAATGAATGCACGAAAGGTC	110-200	3	0.58	Polymorphic
37 ORS673 13 F: TGGTGCTACTCCATCCTTGA 160-500 4 0.68 Polymorphic 38 ORS317 13 F: TTGGCAGTTTGGTGGCTTA 200-210 2 0.22 Polymorphic 39 ORS707 13 F: GCAGTCAATTCGTAGCATCG 150-170 2 0.38 Polymorphic 40 ORS578 14 F: CTCCAATCCCTAAAGTCCCCT 200-600 4 0.6 Polymorphic 741 ORS694 14 F: CCTGAATCCCTAAAGTCCCCT 200-600 4 0.6 Polymorphic 741 ORS694 14 F: CCTGAACCGAAACAGAGAGGGA 160-200 3 0.57 Polymorphic 742 ORS812 15 F: TTGCACATGAGGAGAGGGA 110-130 2 0.26 Polymorphic 743 ORS812 15 F: TTGGGAATGAGAGAGTGAACGG 180-800 3 0.47 Polymorphic 743 ORS812 15 F: TTGGAACAGAGAGTGAATCG 180-800 3 0.47 Polymorphic 744 ORS841 15 F: TTGGAATAGAGAGAGTGAATCG 180-800 3 0.47 Polymorphic				R: TCTGCACATTATCTCCCTCTCTC				
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38 ORS317 13 F: TTTGCCAGTTTGGTGGCTTA 200-210 2 0.22 Polymorphic 39 ORS707 13 F: GCGATGCTTAATTCTTTCTCT 2 0.38 Polymorphic 40 ORS578 14 F: CTCTCAATCCCTAAGCTCCCT 200-600 4 0.6 Polymorphic 41 ORS694 14 F: CTCTGGAACTGAAACCGAGAAC 160-200 3 0.57 Polymorphic 42 ORS307 14 F: CTGGGAGTGGTGGTTGAT 110-130 2 0.26 Polymorphic 43 ORS812 15 F: TTGCACATGAGGGAGGAGAGG 180-800 3 0.47 Polymorphic 44 ORS841 15 F: CTGGGAATAGAGAGGAGGAGGA 180-800 3 0.47 Polymorphic 43 ORS812 15 F: TTCGATGGGTGGATGCT 130-500 6 0.74 Polymorphic 44 ORS688 15 F: TTCGATGGCTGTAAA 180-200 2 0.26 Polymorphic 7 CCACTCATGTGTGTGTAAA 180-200 2 0.26 Polymorphic 7 CCAS688 15 <td< td=""><td></td><td></td><td></td><td>R: GGCATGTTCTCACCGTTCAT</td><td></td><td></td><td></td><td></td></td<>				R: GGCATGTTCTCACCGTTCAT				
R: GGTCGTATGCTTAGTTCTTCTCT39ORS70713F: GCAGAAGCTGAAGCAGACGC150-17020.38Polymorphic40ORS57814F: CTCTCAATCCCTAAGCTCCCT200-60040.6Polymorphic8: GCGTGGAAGTGGATGGGTGTTGTGATR: GCGTGGAACGAGAGAC160-20030.57Polymorphic41ORS69414F: CCTGGAACGAGAGAGGAGGA110-13020.26Polymorphic42ORS30714F: CCGGGAAACAGAGGAGGAGGA110-13020.26PolymorphicR: GCAGTAGAAGATGACGAGGAGGA110-13020.26PolymorphicR: CCGGTGGTCGAATATGAGAGGGAGGAGGA130-50060.74PolymorphicR: CCGGTGGTCGAATATGGAAGGTGCTT130-50060.74PolymorphicR: CCGGTGGTCGAATATGGAACTGATGGCA180-20020.26PolymorphicR: CCACTCATGTTGTATACTGACCA300-34040.54PolymorphicR: CGATATACGGACAATTTCGACCA190-26060.73PolymorphicR: CTGGTGTGTTGTTGTGCCTCGTAT190-26060.73PolymorphicR: TCCAGTTTGGCTCGATAACGGACA190-26060.73PolymorphicR: TCCAGTTTGGTGTGTGTGTGACA160-20050.74PolymorphicR: TCCAGTTGGCACGATTACTGCGATAACGACA190-26060.73PolymorphicR: TCGGTGAAGGGACAATTTCGCCGATATTCGG190-26060.73PolymorphicR: TCCAGTTTGGCTCGATA190-26050.74PolymorphicR: TCGGTGAAGGGGATTATAGGGGCAAT	38	ORS317	13	F: TTTGGCAGTTTGGTGGCTTA	200-210	2	0.22	Polymorphic
39 ORS707 13 F: GCAGTCAATTCGTAGCATCG 150-170 2 0.38 Polymorphic 40 ORS578 14 F: CTCTCAATCCCTAAAGTCCCT 200-600 4 0.6 Polymorphic 41 ORS578 14 F: CTCTCAATCCCTAAAGTCCCT 200-600 4 0.6 Polymorphic 41 ORS694 14 F: CCTGGAACTGAACCGAGAAC 160-200 3 0.57 Polymorphic 42 ORS307 14 F: CCTGGAACTGAACGAGAGGAG 110-130 2 0.26 Polymorphic 43 ORS812 15 F: TTGCACATGAGGGTAGATCG 180-800 3 0.47 Polymorphic 44 ORS384 15 F: CTGGGATAGAGAGGTGCTT 130-500 6 0.74 Polymorphic 7 CCGGTGGTCGAATAGAGAGGTTGCTA 180-800 3 0.47 Polymorphic 8 CCGGTGGTCGAATGAGAGGGATGGTGCTT 130-500 6 0.74 Polymorphic 8 CCGGTGATAGAGAGGCATTTATGGCA 180-200 2 0.26 Polymorphic 8 CAS689 16 F: GCACGATATAACGACACA 30				R: GGTCGTATGCTTAATTCTTTCTCT				
R: GCTGAAGACGATCC40ORS57814F: CTCTCAATCCCTAAAGTCCCCT200-60040.6Polymorphic41ORS69414F: CCTGGAACTGAACCGAGAAC160-20030.57Polymorphic42ORS30714F: CAGTTCCCTGAAACCAAGGAGAGGA110-13020.26Polymorphic43ORS81215F: TTGCACATGAGGGAGGAGGAGGA110-13020.26Polymorphic44ORS38415F: CCGGGGTGGAAACAGAGAGGTGCTT130-50060.74Polymorphic8: CCACTCATGTTGTATTGGGAAT180-20020.26Polymorphic20.26Polymorphic45ORS66815F: TTTCGATTGGACTGTTGCTAAA180-20020.26Polymorphic20.26Polymorphic46ORS89916F: GCCACGTATAACTGACTATGACCA300-34040.54Polymorphic20.28Polymorphic47ORS90216F: GCAGAGTGGACAATTTCACTACAA280-29020.28Polymorphic20.28Polymorphic48ORS80716F: CCGATATTTGACCGATATTTCC190-26060.73Polymorphic20.57Polymorphic50ORS72717F: GGTGGCAAGTGGTGGTAGA160-20050.74Polymorphic2AAGCTGGTCAATGGTGGTTGATAGGGGA51ORS124517F: GAGGGGAGGAGGGGGGGA170-19030.57Polymorphic30.57Polymorphic51ORS124517F: GAGG	39	ORS707	13	F: GCAGTCAATTCGTAGCATCG	150-170	2	0.38	Polymorphic
40 ORS578 14 F: CTCTCAATCCCTAAAGTCCCCT 200-600 4 0.6 Polymorphic R: TGGTGGATGTGGTTGTTGAT 41 ORS694 14 F: CCTGGAACTGAACCGAGAAC 160-200 3 0.57 Polymorphic R: GCCGTGAAACAGAGAGAGGA 42 ORS307 14 F: CAGTTCCCTGAAACCAATTCA 110-130 2 0.26 Polymorphic R: GCAGTAGAAGATGACGGGATG 43 ORS812 15 F: TTGCACATGAGGGTAGATCG 180-800 3 0.47 Polymorphic R: CCGGTGGTCGAATATGAGAG 44 ORS384 15 F: CCTGGAATGAGGGTGCTT 130-500 6 0.74 Polymorphic R: CACTCATGTGTATTGGGAAT 45 ORS668 15 F: TTTCCATTGGACTGTTGCTAAA 180-200 2 0.26 Polymorphic R: CATGTGAGGGCATTTATGTCA 46 ORS899 16 F: GCACGTATAACTGACTATGACCA 300-340 4 0.54 Polymorphic R: CTGAATACTGACTCATGACCACA 47 ORS902 16 F: GCGAAGTGGACAATTTCACTACAA 280-290 2 0.28 Polymorphic R: CTGACCTTCATCTCCTCC 48 ORS807 16 F: CGGATATTTGGACTAT 190-260 6 0.73 Polymorphic R: TCCAGTTTGGTCTTGATTT				R: GCTGAAGCTGAAGACAGATCC				
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41 ORS694 14 F: CCTGGAACTGAACCGAGAAC 160-200 3 0.57 Polymorphic 42 ORS307 14 F: CAGTTCCCTGAAACCAATTCA 110-130 2 0.26 Polymorphic 43 ORS812 15 F: TTGCACATGAGGGTAGATCG 180-800 3 0.47 Polymorphic 44 ORS384 15 F: CCTGGGATAGAGAGGTGCTT 130-500 6 0.74 Polymorphic 45 ORS668 15 F: TTTCGATTGGACTGTTGCTAAA 180-200 2 0.26 Polymorphic 46 ORS899 16 F: GCCACGTATAACTGACCA 300-340 4 0.54 Polymorphic 47 ORS902 16 F: GCAGAGTGGACATTTAGACAA 280-290 2 0.28 Polymorphic 48 ORS807 16 F: CCGATATTTGACCAA 280-290 2 0.28 Polymorphic 9 ORS565 17 F: TGGTCAACGGATTTTGCTCAA 160-200 5 0.74 Polymorphic 6 ORS727 17 F: GGAGTGGCATTTAGAGTCAA 160-200 5 0.74 Polymorphic				R: TGGTGGATGTGGTTGTTGAT				
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42ORS30714F: CAGTTCCCTGAAACCAATTCA110-13020.26Polymorphic43ORS81215F: TTGCACATGAGGGAAGATGACGGGATG180-80030.47Polymorphic44ORS38415F: CCTGGGATAGAGAGGTGCTT130-50060.74Polymorphic45ORS66815F: TTTCGATTGGACTGTTGCTAAA180-20020.26Polymorphic46ORS89916F: GCCACGATAACGACTATGACAA180-20020.26Polymorphic47ORS90216F: GCCACGATAACTGACTATGACCA300-34040.54Polymorphic48ORS80716F: CCGATATTTTGGCCTGGTAT190-26060.73Polymorphic49ORS56517F: GGTGACAGGATTTAGAGTCAA160-20050.74Polymorphic50ORS72717F: GGTGGCAAGTGGTCAATGTGGA170-19030.57Polymorphic51ORS124517F: GAGAGTGGACAATGTGGGGA170-20040.68PolymorphicAverage2.880.51				R: GCCGTGAAACAGAGAGAGAGA				
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43 ORS812 15 F: TTGCACATGAGGGTAGATCG 180-800 3 0.47 Polymorphic 44 ORS384 15 F: CCTGGGATAGAGAGGTTGCTT 130-500 6 0.74 Polymorphic 45 ORS668 15 F: TTTCGATTGGACTGTTGCAAA 180-200 2 0.26 Polymorphic 46 ORS899 16 F: GCCACGTATAACTGACTATGACCA 300-340 4 0.54 Polymorphic 47 ORS902 16 F: GCGAGTGGACAATTTCACTACAAA 280-290 2 0.28 Polymorphic 48 ORS807 16 F: CCGATATCCGCTCGTAT 190-260 6 0.73 Polymorphic 49 ORS565 17 F: TGGTCAACGGATTTAGGTCAA 160-200 5 0.74 Polymorphic 50 ORS727 17 F: GGTGGCAAGTGGTGGTAGA 170-190 3 0.57 Polymorphic 51 ORS1245 17 F: GAAGTGGAGCAATGTTGGTGA 170-200 4 0.68 Polymorphic 71 Average 2.88 0.51 0.57 Polymorphic				R: GCAGTAGAAGATGACGGGATG			- ·	
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44ORS38415F: CCTGGGATAGAGAGGTTGCTT130-50060.74Polymorphic45ORS66815F: TTTCGATTGGACTGTTGCTAAA180-20020.26Polymorphic46ORS89916F: GCCACGTATAACTGACTATGACCA300-34040.54Polymorphic47ORS90216F: GCAGAGTGGACAATTTCACTACAA280-29020.28Polymorphic48ORS80716F: CCGATATTTTGACCGATATTTCC190-26060.73Polymorphic49ORS56517F: TGGTCAACGGATTTAGAGTCAA160-20050.74Polymorphic50ORS72717F: GGTGGCAAGTGGTGGTTAGA170-19030.57Polymorphic51ORS124517F: GAAGTGGAGCAATGTTGGTGA170-20040.68PolymorphicR: CCCCCAAGATATTAGTGTGATGATTAverage2.880.51		00000		R: CCGGTGGTCGAATATGAGAG		•	<u> </u>	
45ORS66815F: TTTCGATTGGACTGTTGCTAAA R: CATGTGAGGGCATTTATGTCA180-20020.26Polymorphic Rolphic R: CATGTGAGGGCATTAACTGACTATGACCA46ORS89916F: GCCACGTATAACTGACTATGACCA300-34040.54Polymorphic R: CGAATACAGACTCGATAAACGACA47ORS90216F: GCAGAGTGGACAATTTCACTACAA R: CTTGGTTGTTGCTGCTCGTAT280-29020.28Polymorphic R: CTTGGTTGTTGTTGCCTCGTAT48ORS80716F: CCGATATTTTGACCGATATTTGC R: TCTCACCCTTCATCTCCTCC190-26060.73Polymorphic R: TCTCACCCTTCATCTCCTCC49ORS56517F: TGGTCAACGGATTTAGAGTCAA R: TCCAGTTTGGTCTTGATTTGG160-20050.74Polymorphic R: TCCAGTTTGGTCTTGATTTGG50ORS72717F: GGTGGCAAGTGGTGGTGAGA R: AAAGCTGGTCATCTCAAGGGTA170-19030.57Polymorphic R: CGCCAAGATATTAGTGTGATGATT R: CGCCAAGATATTAGTGTGATGATT51ORS124517F: GAAGTGGAGCAATGTTGGTGA R: CGCCAAGATATTAGTGTGATGATT R: CGCCAAGATATTAGTGTGATGATT2.880.51	44	ORS384	15		130-500	6	0.74	Polymorphic
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46ORS89916F: GCCACGTATAACTGACTATGACCA300-34040.54Polymorphic47ORS90216F: GCAGAGTGGACAATTTCACTACAA280-29020.28Polymorphic47ORS90216F: GCAGAGTGGACAATTTCACTACAA280-29020.28Polymorphic48ORS80716F: CCGATATTTTGACCGATATTTGC190-26060.73Polymorphic49ORS56517F: TGGTCAACGGATTTAGAGTCAA160-20050.74Polymorphic50ORS72717F: GGTGGCAAGTGGTGGTTAGA170-19030.57Polymorphic51ORS124517F: GAAGTGGAGCAATGTTGGTGA170-20040.68PolymorphicR: CGCCAAGATATTAGTGTGATGAT170-20040.68PolymorphicAverage2.880.51	45	ORS668	15	F: TTTCGATTGGACTGTTGCTAAA	180-200	2	0.26	Polymorphic
46ORS89916F: GCCACGTATAACTGACTATGACCA300-34040.54Polymorphic47ORS90216F: GCAGAGTGGACAATTTCACTACAA280-29020.28Polymorphic48ORS80716F: CCGATATTTTGACCGATATTTGC190-26060.73Polymorphic49ORS56517F: TGGTCAACGGATTTAGAGTCAA160-20050.74Polymorphic50ORS72717F: GGTGGCAAGTGGTGGTTAGA170-19030.57Polymorphic51ORS124517F: GAAGTGGAGCAATGTTGGTGA170-20040.68PolymorphicR: CCCCAGGTATTAGAGTGAGAGTAverage2.880.51	10						<u> </u>	
47ORS90216F: GCAGAGTGGACAATTTCACTACAA R: CTTGGTTGTTGCTCGTAT280-29020.28Polymorphic Polymorphic R: CTTGGTTGTTGCCCCGTAT48ORS80716F: CCGATATTTTGACCGATATTTTGC R: TCTCACCCTTCATCTCCTTCC190-26060.73Polymorphic R: TCTCACCCTTCATCTCCTTCC49ORS56517F: TGGTCAACGGATTTAGAGTCAA R: TCCAGTTTGGTCTTGATTTGG160-20050.74Polymorphic R: TCCAGTTTGGTCTTGATTTGG50ORS72717F: GGTGGCAAGTGGTGGTTAGA R: AAAGCTGGTCATCTCAAGGGTA170-19030.57Polymorphic R: AAAGCTGGTCATCTCAAGGGTA51ORS124517F: GAAGTGGAGCAATGTTGGTGA R: CGCCAAGATATTAGTGTGATGATT170-20040.68Polymorphic R: 2.880.51	46	ORS899	16		300-340	4	0.54	Polymorphic
47ORS90216F: GCAGAGTGGACAATTTCACTACAA280-29020.28PolymorphicR: CTTGGTTGTTGTTGCTGCTCGTATR: CTTGGTTGTTGTTGCCTCGTAT190-26060.73Polymorphic48ORS80716F: CCGATATTTTGACCGATATTTGC190-26060.73Polymorphic49ORS56517F: TGGTCAACGGATTTAGAGTCAA160-20050.74Polymorphic50ORS72717F: GGTGGCAAGTGGTGGTTAGA170-19030.57Polymorphic51ORS124517F: GAAGTGGAGCAATGTTGGTGA170-20040.68PolymorphicR: CGCCAAGATATTAGTGTGATGATTAverage2.880.51	47	000000	40			0	0.00	
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48 ORS807 16 F: CCGATATTTTGACCGATATTTTGC 190-260 6 0.73 Polymorphic 49 ORS565 17 F: TGGTCAACGGATTTAGAGTCAA 160-200 5 0.74 Polymorphic 50 ORS727 17 F: GGTGGCAAGTGGTGGTTAGA 170-190 3 0.57 Polymorphic 51 ORS1245 17 F: GAAGTGGAGCAATGTTGGTGAA 170-200 4 0.68 Polymorphic Average 2.88 0.51	10	00000			100.000	•		
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49 ORS565 17 F: IGGTCAACGGATTTAGAGTCAA 160-200 5 0.74 Polymorphic R: TCCAGTTTGGTCTTGATTTGG R: TCCAGTTTGGTCTTGATTTGG 170-190 3 0.57 Polymorphic 50 ORS727 17 F: GGTGGCAAGTGGTGGTTAGA 170-190 3 0.57 Polymorphic 51 ORS1245 17 F: GAAGTGGAGCAATGTTGGTGA 170-200 4 0.68 Polymorphic R: CGCCAAGATATTAGTGTGATGATGATT Average 2.88 0.51	40	000505	4-		100.000	-	0.74	
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50 ORS727 17 F: GGTGGCAAGTGGTGGTTAGA 170-190 3 0.57 Polymorphic R: AAAGCTGGTCATCTCAAGGGTA R: AAAGCTGGTGGTGGTGGTGGGGGGGGGGGGGGGGGGGGG	50	000707	4-	R: ICCAGIIIGGICIIGAIIIGG	470.400	0	0	
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Average 2.88 0.51	51	UK51245	17	R: CGCCAAGATATTAGTGTGATGATT	170-200	4	U.08	Polymorphic
		Average				2.88	0.51	

Parents	COSF 6B	COSF 10B	COSF 12B	COSF 13B	CMS 104B	CMS 112B	CMS 207B	CMS 519B	ARM 248B	RCR CMS 38B	IR 6	CSFI 99	RHA- 1-1	LTRR 341
COSF 6B	0	0.38	0.43	0.49	0.4	0.45	0.54	0.56	0.49	0.52	0.53	0.65	0.53	0.52
COSF 10B		0	0.3	0.38	0.45	0.51	0.52	0.51	0.55	0.54	0.56	0.65	0.59	0.5
COSF 12B			0	0.37	0.49	0.52	0.54	0.54	0.54	0.56	0.55	0.64	0.56	0.54
COSF 13B				0	0.5	0.57	0.57	0.59	0.58	0.54	0.52	0.63	0.5	0.48
CMS 104B					0	0.31	0.47	0.5	0.46	0.46	0.53	0.66	0.56	0.46
CMS 112B						0	0.49	0.57	0.52	0.57	0.55	0.64	0.58	0.55
CMS 207B							0	0.53	0.62	0.54	0.57	0.66	0.61	0.58
CMS 519B								0	0.57	0.51	0.64	0.67	0.58	0.57
ARM 248B									0	0.45	0.56	0.63	0.61	0.44
RCR CMS 38B										0	0.52	0.63	0.47	0.4
IR 6											0	0.67	0.41	0.49
CSFI 99												0	0.68	0.64
RHA-1-1													0	0.4
LTRR 341														0

Table 2. Genetic dissimilarity coefficient values between 14 parents in sunflower

divergence between them. In general, the restorer inbred CSFI 99 had more divergence with other parental lines. Low dissimilarity indices were recorded between COSF 15B vs COSF 12B (0.30) followed by COSF 12B vs CMS 104B (0.31), COSF 12B vs COSF 13B (0.37) and RCR CMS 38B vs LTRR 341(0.40) which indicated less divergence between them. Similar results were reported by various scientists (Zia *et al.*, 2014; Zeinalzadeh-

Tabrizi *et al.*, 2018; Yihan *et al.*, 2022; Ahmed *et al.*, 2022) in sunflower.

A phylogenetic tree was constructed by neighbour-joining method which grouped the parents into four clusters (**Fig. 1**). Among the four clusters, cluster I had 5 genotypes, clusters III and IV had four genotypes each and cluster II had one genotype. The clustering pattern



Fig. 1. Neighbour-joining tree of 14 sunflower parental genotypes based on SSR marker data

indicated that three out of four restorer inbreds were grouped into one cluster while the restorer inbred CSFI 99 was grouped separately from other parental lines. Likewise, the COSF series of maintainer inbreds were grouped into a single cluster. It might be due to the similar selection pressure for various traits during the breeding programme. This result was in accordance with the study conducted by Lochner, (2011) in sunflower in which the SSR cluster analysis grouped the A and B lines (female) into one cluster and the restorers (male) were grouped in a separate cluster.

All the 14 parental lines were analysed for phenotypic diversity through D^2 analysis. The D^2 values e ranged between 65.96 (CMS 112A vs CSFI 99) to 2598.85 (CMS 104A vs RHA-1-1) (**Table 3**). The D^2 analysis grouped the parental lines into six clusters (**Table 4**). Among them, genotype pairs CMS104B vs RHA-1-1 and CSFI 99 vs LTRR341 had highest and lowest D^2 values respectively. Among the restorer inbreds, RHA-1-1 was grouped into

a separate cluster. Likewise, the maintainer inbred CMS 207B was also grouped into a separate cluster. Other restorer inbreds CSFI 99 and LTRR 341 were clustered along with other maintainer lines. Likewise, the restorer IR 6 was clustered along with maintainer inbred CMS 519B.

The dissimilarity values, D² values, and mean performance of hybrids for yield and its component traits are presented in **Table 5**. Among the parental combinations, the dissimilarity values ranged from 0.4 (RCR CMS 38A *vs* LTRR 341) to 0.67 (CMS519A *vs* CSFI 99). The D² values ranged between 65.96 (CMS 112A *vs* CSFI 99) to 2598.85 (CMS 104A *vs* RHA-1-1). Mid-parent heterosis and better parent heterosis of 40 hybrids for yield and its component traits are presented in **Table 6 and 7**.

The mean value of nine biometrical traits, mid-parental heterosis, better parental heterosis, D^2 values and dissimilarity values of 40 hybrids were subjected to

Table 3. D ² v	alues between	14 parents	in sunflower
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Parents	COSF 6B	COSF 10B	COSF 12B	COSF 13B	CMS 104B	CMS 112B	CMS 207B	CMS 519B	ARM 248B	RCR CMS 38B	IR 6	CSFI 99	RHA- 1-1	LTRR 341
COSF 6B	0.00	983.43	417.70	565.27	460.62	115.23	261.70	186.90	60.99	305.42	211.79	140.79	1100.58	119.34
COSF 10B		0.00	137.34	154.65	2342.28	1284.71	434.67	1168.35	1345.98	2001.40	821.31	1135.42	239.62	986.79
COSF 12B			0.00	117.42	1406.79	627.82	151.74	655.53	667.49	1142.09	457.42	518.79	333.49	420.20
COSF 13B				0.00	1826.95	887.15	361.86	646.50	863.04	1505.37	334.00	872.14	143.96	752.75
CMS 104B					0.00	186.40	921.06	745.21	241.97	59.88	1141.23	307.19	2598.85	344.89
CMS 112B						0.00	342.45	325.39	40.20	115.88	527.52	65.96	1468.83	84.85
CMS 207B							0.00	366.36	378.86	663.48	396.03	239.74	772.29	180.06
CMS 519B								0.00	217.02	452.63	126.76	403.69	1142.70	374.04
ARM 248B									0.00	143.03	369.84	128.09	1455.74	121.11
RCR CMS 38B										0.00	843.08	182.64	2201.41	226.80
IR 6											0.00	604.94	739.92	511.70
CSFI 99												0.00	1449.64	39.38
RHA-1-1													0.00	1283.11
LTRR 341														0.00

Table 4. Clustering pattern of parents by D² analysis

Clusters	Number of genotypes	Name of the genotypes				
l	5	CSFI 99	LTRR 341	CMS 112B	ARM 248B	COSF 6B
II	2	CMS 104B	RCR CMS 38B	3		
111	3	COSF 12B	COSF 13B	COSF 10B		
IV	2	CMS 519B	IR6			
V	1	RHA-1-1				
VI	1	CMS 207B				

Table 5. Dis-similarity and D^2 values and Mean performance of hybrids for yield and its component traits in sunflower

Lines x testers	Days to 50% flowering	Plant height (cm)	Head diameter (cm)	Days to maturity	Volume weight (g/100ml)	100 seed weight (g)	Seed yield / plant (g)	Oil content (%)	Oil yield / plant (g)	Dis- similarity	D ² values
COSF 6A x IR 6	58.50	169.00	11.55	90.50	43.30	4.18	52.97	45.14	23.91	0.53	211.79
COSF 6Ax CSFI 99	53.00	174.95	15.50	84.50	42.54	5.42	62.54	39.75	24.87	0.65	140.79
COSF 6A x RHA-1-1	50.50	151.50	13.38	79.50	40.00	5.91	75.46	34.72	26.23	0.53	1100.58
COSF 6A x LTRR 341	54.50	126.30	12.35	85.50	29.30	3.37	40.12	35.31	21.24	0.52	119.34
COSF 10A x IR 6	61.00	185.10	15.48	91.00	43.35	4.87	51.71	39.32	28.92	0.56	821.31
COSF 10A x CSFI 99	49.50	177.83	14.72	81.50	42.00	6.25	69.73	39.68	27.15	0.65	1135.42
COSF 10A x RHA-1-1	45.50	168.70	17.41	76.00	41.15	7.06	51.69	40.55	19.66	0.59	239.62
COSF 10A x LTRR 341	52.00	165.40	14.20	83.00	44.10	4.88	72.88	42.26	31.80	0.50	986.79
COSF 12A x IR 6	57.50	187.68	18.64	89.50	39.25	4.53	88.59	39.68	33.64	0.55	457.42
COSF 12A x CSFI 99	51.00	185.45	14.00	82.50	47.25	5.32	53.94	40.90	23.02	0.64	518.79
COSF 12A x RHA-1-1	43.00	131.93	12.49	76.50	36.40	5.10	50.43	39.55	23.51	0.56	333.49
COSF 12A x LTRR 341	51.50	182.00	14.33	83.50	40.40	4.98	79.45	40.85	32.34	0.54	420.20
COSF 13A x IR 6	55.50	183.00	18.06	87.50	37.55	4.66	84.53	38.87	32.89	0.52	334.00
COSF 13A x CSFI 99	49.00	178.02	17.24	79.00	46.30	5.79	69.06	41.68	28.74	0.63	872.14
COSF 13A x RHA-1-1	47.00	154.05	13.69	78.00	42.85	5.69	55.89	40.66	21.50	0.50	143.96
COSF 13A x LTRR 341	53.50	154.00	15.36	86.00	35.10	4.90	69.85	36.57	25.55	0.48	752.75
CMS 104A x IR 6	60.00	176.20	14.40	93.50	41.65	4.12	52.23	38.90	20.27	0.53	1141.23
CMS 104A x CSFI 99	50.00	162.00	16.23	82.00	40.35	3.96	63.80	35.97	23.11	0.66	307.19
CMS 104A x RHA-1-1	48.50	149.45	13.40	80.00	40.55	5.28	34.29	35.92	12.35	0.56	2598.85
CMS 104A x LTRR 341	56.00	162.82	15.65	86.50	41.70	5.83	63.04	40.17	25.27	0.46	344.89
CMS 112A x IR 6	57.50	171.30	15.41	88.00	39.70	4.05	79.58	40.87	32.56	0.55	527.52
CMS 112A x CSFI 99	54.50	175.70	18.55	85.50	35.45	4.68	78.70	36.22	28.71	0.64	65.96
CMS 112A x RHA-1-1	49.00	167.82	14.59	81.50	36.95	4.93	74.87	37.87	28.30	0.58	1468.83
CMS 112A x LTRR 341	49.00	157.42	16.68	80.50	39.90	7.55	48.10	36.35	17.47	0.55	84.85
CMS 207A x IR 6	53.00	167.02	17.37	84.00	39.20	4.50	55.84	38.26	21.42	0.57	396.03
CMS 207A x CSFI 99	51.50	159.90	15.36	83.50	38.30	4.74	59.00	35.09	20.77	0.66	239.74
CMS 207A x RHA-1-1	49.50	143.20	16.38	80.00	37.35	6.02	85.52	37.37	31.94	0.61	772.29
CMS 207A x LTRR 341	58.00	158.39	16.91	90.00	32.95	5.22	82.61	34.72	28.71	0.58	180.06
CMS 519A x IR 6	60.50	191.35	19.63	93.00	42.45	4.95	74.66	35.83	26.75	0.64	126.76
CMS 519A x CSFI 99	57.00	199.98	15.03	88.50	43.40	5.11	60.90	35.54	21.65	0.67	403.69
CMS 519A x RHA-1-1	55.50	166.20	14.63	87.50	36.35	4.95	56.83	29.38	16.72	0.58	1142.70
CMS 519A x LTRR 341	54.50	174.10	17.50	88.50	35.00	5.35	58.45	33.58	19.63	0.57	374.04
RM 248A x IR 6	59.00	183.80	17.76	90.00	39.90	4.64	80.87	40.09	30.41	0.56	369.84
RM 248A x CSFI 99	53.00	174.00	16.75	86.00	38.20	6.25	77.75	40.51	31.60	0.63	128.09
RM 248A x RHA-1-1	57.00	168.84	22.70	89.00	37.10	8.45	31.22	31.94	9.91	0.61	1455.74
RM 248A x LTRR 341	53.00	129.32	13.59	83.50	32.65	6.97	32.09	31.52	9.63	0.44	121.11
RCR CMS 38A x IR 6	58.00	178.00	19.24	89.50	42.55	5.56	87.19	38.52	33.54	0.52	843.08
RCR CMS 38A x CSFI 99	51.50	171.57	13.68	85.00	41.55	5.12	64.99	36.62	23.83	0.63	182.64
RCR CMS 38A x RHA-1-1	50.50	159.95	20.19	83.00	36.80	5.45	65.53	37.28	24.40	0.47	2201.41
RCR CMS 38A x LTRR 341	57.50	166.10	16.47	89.00	35.40	5.91	84.47	35.40	29.84	0.40	226.80

Table 6. Mid-p	arental heterosis	of hybrids	for yield and	its component	traits in sunflower

Lines x testers	Days to 50% flowering	Plant height (cm)	Head diameter (cm)	Days to maturity	Volume weight (g/100ml)	100 seed weight (g)	Seed yield / plant (g)	Oil content (%)	Oil yield / plant (g)
COSF 6A x IR 6	-2.9	20.71	-9.09	-2.43	28.58	15.73	121.17	21.1	161.31
COSF 6Ax CSFI 99	-1.85	18.49	6.02	-2.87	11.57	22.35	55.61	-1.2	55
COSF 6A x RHA-1-1	-1.94	20.35	13.92	-4.22	11.58	11.04	140.4	-9.22	119.14
COSF 6A x LTRR 341	-4.8	-15.21	-10.99	-4.74	-19.45	-31.08	-7.2	-7.82	31.09
COSF 10A x IR 6	5.63	58.07	23.89	1.68	45.59	28.07	66.11	7	143.59
COSF 10A x CSFI 99	-3.88	42.55	2.15	-2.69	22.72	35.03	47.2	-0.05	44.64
COSF 10A x RHA-1-1	-7.14	63.81	50.93	-4.7	28.79	27.87	34.02	7.5	33.83
COSF 10A x LTRR 341	-5.02	31.2	3.92	-4.05	35.8	-3.89	44.56	11.86	68.03
COSF 12A x IR 6	0.44	48.89	50.5	0.56	18.42	14.61	194.51	6.57	189.08
COSF 12A x CSFI 99	0	38.71	-2.1	-0.9	25.68	11.31	16.45	1.75	24.2
COSF 12A x RHA-1-1	-11.34	17.86	9.32	-3.47	3.06	-10.01	34.41	3.5	62.67
COSF 12A x LTRR 341	-5.07	34.8	5.72	-2.91	12.71	-4.73	60.94	6.72	73.03
COSF 13A x IR 6	-4.72	49.14	46	-3.31	25.17	15.92	257.35	5.74	270.64
COSF 13A x CSFI 99	-5.77	36.57	20.69	-6.78	34.4	19.46	73.1	4.94	82.27
COSF 13A x RHA-1-1	-5.05	41.87	19.98	-3.41	33.18	-0.87	79.74	7.74	83.88
COSF 13A x LTRR 341	-3.17	16.97	13.44	-1.71	7.34	-7.55	62.67	-3.25	60.44
CMS 104A x IR 6	-3.61	12.7	1.19	0	35.45	-6.42	81.83	8.57	92.41
CMS 104A x CSFI 99	-10.71	-1.22	0.5	-6.55	14.63	-24.31	41.89	-7.28	32.59
CMS 104A x RHA-1-1	-9.35	5.07	0.98	-4.48	23.16	-13.77	-5.18	-2.41	-7.51
CMS 104A x LTRR 341	-5.49	-1.51	1.62	-4.42	24.66	2.6	31.31	8.94	43.65
CMS 112A x IR 6	-2.54	17.89	12.52	-3.83	35.84	0.43	197.38	14.06	232.53
CMS 112A x CSFI 99	3.32	14.87	18.83	-0.29	5.27	-3.6	83.02	-6.63	72.07
CMS 112A x RHA-1-1	-2.49	27.92	14.57	-0.31	17.68	-14.22	118.92	2.89	124.47
CMS 112A x LTRR 341	-12.5	2.05	12.21	-9.04	24.98	42.16	4.46	-1.41	3.7
CMS 207A x IR 6	-10.92	28.53	29.92	-8.45	43.72	39.92	63.75	15.77	92.5
CMS 207A x CSFI 99	-3.29	16.21	0.49	-2.91	20.72	17.35	17.2	-2.53	15.22
CMS 207A x RHA-1-1	-2.46	23.62	31.99	-2.44	26.83	22.07	105.87	9.84	129.04
CMS 207A x LTRR 341	2.65	14.02	16.3	1.41	9.92	16.32	54.74	1.87	57.92
CMS 519A x IR 6	-2.42	39.97	41.89	-0.8	54.64	61.37	297.02	16.49	366.75
CMS 519A x CSFI 99	2.24	38.53	-4.57	0.57	36.05	31.36	73.78	5.42	71.45
CMS 519A x RHA-1-1	4.23	35.56	13.63	4.17	22.7	3.5	116.54	-7.41	95.55
CMS 519A x LTRR 341	-7.63	19.51	16.63	-2.48	16.09	23.09	53.45	5.61	53.53
RM 248A x IR 6	-2.48	25.63	41.57	-3.49	37.35	33.24	261.92	15.17	287.08
RM 248A x CSFI 99	-2.3	13.02	15.84	-1.71	14.03	45.06	101.5	7.26	114.24
RM 248A x RHA-1-1	10.14	27.72	95.94	6.59	18.82	62.58	4.82	-10.75	-7.19
RM 248A x LTRR 341	-7.83	-16.71	-0.95	-7.48	2.83	46.51	-22.92	-12.07	-35.44
RCR CMS 38A x IR 6	-4.13	18.47	32.55	-2.45	34.65	34.5	190.73	13.82	229.42
RCR CMS 38A x CSFI 99	-5.07	8.66	-16.74	-1.16	15.26	3.28	40.58	-0.49	39.53
RCR CMS 38A x RHA-1-1	-2.42	17.49	48.95	1.22	8.96	-6.76	75.07	7.08	87.69
RCR CMS 38A x LTRR 341	0	4.33	5	0.28	3.21	9.34	71.43	1.51	73.16

Table 1. Detter parental neterosis of hybrids for yield and its component traits in sumowe
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Lines x testers	Days to 50% flowering	Plant height (cm)	Head diameter (cm)	Days to maturity	Volume weight (g/100ml)	100 seed weight (g)	Seed yield / plant (g)	Oil content (%)	Oil yield / plant (g)
COSF 6A x IR 6	-7.14	15.2	-11.43	-5.24	13.95	5.83	72.54	10.6	91.36
COSF 6Ax CSFI 99	-7.83	17.73	-4.32	-6.11	11.2	10.27	25.89	-2.61	26.92
COSF 6A x RHA-1-1	-12.17	3.27	2.61	-11.67	5.26	-11.79	135.22	-14.92	109.88
COSF 6A x LTRR 341	-5.22	-16.48	-16.04	-5	-22.89	-42.18	-28.06	-13.48	6.68
COSF 10A x IR 6	-3.17	38.86	22.66	-4.71	43.54	12.34	14.76	-1.11	61.2
COSF 10A x CSFI 99	-5.71	19.67	-9.14	-2.98	9.8	27.06	40.36	-0.19	38.53
COSF 10A x RHA-1-1	-13.33	60.55	37.96	-8.98	22.11	5.3	14.71	2	9.59
COSF 10A x LTRR 341	-8.77	9.37	-3.47	-7.26	26.91	-16.15	30.68	6.3	59.72
COSF 12A x IR 6	-8.73	40.8	50.32	-6.28	6.25	-2.27	106.22	-2.6	92.59
COSF 12A x CSFI 99	-0.97	24.8	-13.58	-1.79	23.53	8.14	8.57	0.38	17.45
COSF 12A x RHA-1-1	-16.5	11.05	0.73	-7.27	-1.46	-23.88	17.39	-2.92	34.61
COSF 12A x LTRR 341	-9.65	20.35	-2.58	-6.7	9.37	-14.43	42.46	0.26	62.41
COSF 13A x IR 6	-11.9	37.28	46	-8.38	22.51	-2.31	180.74	-2.31	175.42
COSF 13A x CSFI 99	-8.41	19.8	6.42	-7.6	21.05	17.7	39.01	4.75	46.67
COSF 13A x RHA-1-1	-12.15	37.42	10.67	-8.77	27.15	-15.15	74.22	2.19	80.03
COSF 13A x LTRR 341	-6.14	1.84	4.42	-3.91	1.01	-15.89	25.25	-8.09	28.33
CMS 104A x IR 6	-4.76	-1.78	-10.5	-2.09	29.55	-25.56	29.76	2.56	32.79
CMS 104A x CSFI 99	-18.7	-9.7	0.15	-10.38	5.49	-28.55	28.42	-9.28	17.94
CMS 104A x RHA-1-1	-21.14	-16.69	-16.72	-12.57	20.33	-21.27	-14.81	-5.3	-19.1
CMS 104A x LTRR 341	-8.94	-9.24	-2.73	-5.46	20	0.09	13.04	5.89	26.9
CMS 112A x IR 6	-8.73	8.9	2.6	-7.85	35.26	-15.54	119.11	7.75	136.33
CMS 112A x CSFI 99	-0.91	11.7	14.51	-2.29	-7.32	-4.78	58.41	-8.65	46.52
CMS 112A x RHA-1-1	-10.91	6.69	-2.86	-6.86	9.64	-26.42	106.14	-0.15	105.44
CMS 112A x LTRR 341	-14.04	0.08	11.05	-10.06	14.82	29.64	-13.75	-4.17	-12.28
CMS 207A x IR 6	-15.87	25.3	20.88	-12.04	33.56	37.46	9.49	13.42	30.21
CMS 207A x CSFI 99	-8.04	7.6	-5.19	-5.11	0.13	-3.66	15.69	-11.49	5.97
CMS 207A x RHA-1-1	-11.61	13.11	13.99	-9.09	10.83	-10.22	67.69	4.72	94.16
CMS 207A x LTRR 341	1.75	4.74	14.96	0.56	-5.18	-10.31	48.13	-3.03	44.2
CMS 519A x IR 6	-3.97	36.56	28.3	-2.62	44.63	51.38	265.8	6.21	360.72
CMS 519A x CSFI 99	-6.56	34.58	-7.22	-3.8	13.46	3.97	22.58	-10.35	10.46
CMS 519A x RHA-1-1	-9.02	18.61	-4.38	-4.89	7.86	-26.12	77.15	-17.67	46.11
CMS 519A x LTRR 341	-10.66	15.13	14.38	-3.8	0.72	-8.16	4.81	-6.23	-1.43
RM 248A x IR 6	-6.35	15.38	39.62	-5.76	35.95	25.58	194.18	11.72	206.97
RM 248A x CSFI 99	-8.62	9.23	3.4	-5.49	-0.13	27.06	56.5	2.17	61.27
RM 248A x RHA-1-1	-1.72	5.99	78.46	-2.2	10.09	26.12	-2.68	-10.99	-13.42
RM 248A x LTRR 341	-8.62	-18.82	-7.65	-8.24	-6.04	19.76	-42.46	-12.17	-51.66
RCR CMS 38A x IR 6	-7.94	6.46	15.49	-6.28	25.7	11.32	103.81	13.46	130.4
RCR CMS 38A x CSFI 99	-11.21	2.61	-17.89	-3.41	8.63	2.51	30.82	-7.64	21.59
RCR CMS 38A x RHA-1-1	-12.93	-4.34	21.19	-5.68	8.71	-18.66	53.18	4.47	67.61
RCR CMS 38A x LTRR 341	-0.86	-0.66	-1.14	-0.56	1.87	1.55	51.46	-1.13	49.87

Characters		D ² values		Dis	similarity value	S
	Mean performance	Mid-parent heterosis	Better parent heterosis	Mean performance	Mid-parent heterosis	Better Parent heterosis
Days to 50% flowering	-0.14	0.13	-0.25	-0.13	0.16	0.03
Plant height (cm)	-0.06	0.08	-0.25	0.39**	0.29*	0.32*
Head diameter (cm)	0.09	0.22	0.01	0.13	0.01	0.01
Days to maturity	-0.13	0.29*	-0.18	-0.09	0.12	0.1
Volume weight (g/100ml)	0.08	0.08	0.15	0.31*	0.22	0.03
100 seed weight (g)	0.1	-0.21	-0.27*	-0.03	0.23	0.25
Seed yield / plant (g)	-0.17	-0.09	-0.06	0.01	0.01	0.05
Oil content (%)	-0.13	-0.11	-0.01	-0.01	-0.09	-0.15
Oil yield / plant (g)	-0.19	-0.11	-0.09	-0.02	-0.01	0.02

Table 8. Correlations of D² and dissimilarity values with hybrid performance and heterosis

*, ** Significance at 5 and 1 % level respectively.

correlation analysis (Table 8). The D² values did not show any association with hybrid performance for all traits. Hence, the results indicated that mean performance of hybrids could not be predicted based on phenotypic diversity analysis. However, D² values were correlated significantly with mid-parental heterosis for days to maturity and better parent heterosis for hundred seed weight. Hence, the diverse parental combinations at phenotypic level may produce more mid parental or better parent heterosis for these traits. In contrast, the dissimilarity indices had significant and positive association with hybrid performance for plant height (0.39) and volume weight (0.31). Also, dissimilarity indices recorded significant and positive association with mid (0.29) and better parent (0.32) heterosis for plant height. These results indicated that diverse parental combinations at the molecular level may produce hybrids with increased performance for plant height and volume weight. Similar superior performance of hybrids can be expected for mid and better parent heterosis for plant height. This result in in accordance with the findings of Xiao et al. (1996) in rice; Sureja et al. (2006) in ash gourd and Gupta et al. (2018) in pearl millet and Somashekhar et al. (2020) in cotton. Hence, the molecular level diversity could be correlated with heterotic expression of two traits only and not for oil yield and other component traits. This study may be extended further with more number of parental lines and molecular markers over various environments for utilization of these results in the heterosis breeding program of sunflower.

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