

Assessment of heterosis for yield and its component traits in Pearl Millet (*Pennisetum glaucum* (L.) R. Br. in pooled environments

Comment [R1]: Title can be improved

Abstract:

An experiment comprised of twelve male sterile lines, three inbred testers of pearl millet, their 36 hybrids and a standard check (Kaveri Super Boss) was conducted at ICAR- Indian Institute of Millets Research, Hyderabad in *Kharif* 2022 and summer 2023 for studying the extent of hybrid vigour in F_1 for grain yield and its components. The cross 274A \times 123R showed highest standard heterosis for total number of effective tillers per plant, dry biomass, grain yield per plot, harvest index and 1000 seed weight. Among 36 hybrids studied, five hybrids namely 274A \times 123R, 260A \times 124R, 252A \times 124R, 269A \times 124R and 246A \times 123R were selected as best crosses since they expressed high better parent, mid parent and standard heterosis over standard check for grain yield per plot. These hybrids can be further recommended for evaluation to improve yield performance.

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Introduction:

Pearl millet (*Pennisetum glaucum*), $2n=14$ (Sattler *et al.*, 2019) is a little cereal grain that is high in nutrients and resistant to heat and drought. Its efficient use of resources and ability to produce substantial yields under low-input conditions contribute to its role in food security and sustainable agriculture, especially in areas prone to drought and soil degradation. Pearl millet grains are frequently produced in many Asian and African countries. India has been the world's biggest producer of this crop for the past five years, harvesting 7.3 million tons from 10.6 million hectares of land ((Rani *et al.*, 2023). The crop is mostly grown in the country during the *Kharif* season, while it is also grown to a lesser amount during the Rabi season. The C4 mechanism allows pearl millet to efficiently capture carbon dioxide and convert it into energy even under high temperatures and low moisture conditions. It can thrive in regions with very low annual rainfall (less than 250 mm), it shows resilience to soils with high aluminium saturation, which often affects root development and crop productivity. Heterosis breeding is the most practical method for pearl millet due to its extensively cross-pollinated nature, which is aided by protogynous flowering and the availability of CMS systems. In pearl millet, successful heterosis breeding mainly relies on the development of diverse sets of A-, B- and R-line pools distinguished with wide genetic variability. In heterosis breeding, choosing appropriate parents and determining the degree of heterosis in the resulting crosses are crucial steps. The heterosis over mid parental value has limited scope in practical breeding. Therefore, the heterosis is more beneficial when compared to the better parent and the conventional check hybrid. As a result, the kind and degree of heterosis aid in the identification of the best cross-combinations to produce the best transgressive segregants.

Materials and Methods:

The present study involved twelve male sterile lines viz., 04999A, 843-22A, 221A, 242A, 246A, 252A, 260A, 262A, 264A, 269A, 274A, 291A and three restorer pollinators viz., 123R, 124R and 132R; developed at ICAR- Indian Institute of Millets Research,

Hyderabad and a single standard check hybrid Kaveri Super Boss. The parental lines and testers were crossed in line x tester mating design during summer 2021 and kharif 2022, and evaluated in randomized complete block design during kharif 2022 and summer 2023. Five competitive plants from each experimental unit of every replication were selected randomly for recording observations on component characters viz., Days to 50 % flowering, days to maturity, plant height(cm), number of effective tillers per plant, flag leaf length (cm), flag leaf width (cm), leaf length (cm), leaf width (cm), panicle length (cm), panicle width (cm), actual photosystem II efficiency (Φ PSII), maximum photosystem II efficiency (F_v/F_m), fresh biomass (kg/plot), dry biomass (kg/plot), harvest index (%), grain yield (kg/plot), and 1000 seed weight (g). The expression of heterosis in 36 hybrids involving twelve CGMS lines and three testers was measured in terms of heterobeltiosis in relation to better parent and standard heterosis in comparison with standard check Kaveri Super Boss. The analysis of variance for the RBD was carried out for each character by following the model given by Panse and Sukhatme (1967).

Comment [R2]: Give number of replications and the spacing row to row and plant to plant

The heterosis was estimated in relation to mid parent, and check i.e as percentage increase or decrease of F_1 's over the mid parent (MP), better parent (BP) and check (Ch) values following the method of Fonesca and Patterson (1968), Liang *et al.* (1972) and Virmani *et al.* (1982).

(a) Heterosis over mid parent (H1)

$$\% H1 = \frac{\overline{F_1} - \overline{MP}}{\overline{MP}} \times 100$$

(b) Heterosis over better parent (H2)

$$\% H2 = \frac{\overline{F_1} - \overline{BP}}{\overline{BP}} \times 100$$

(c) Heterosis over check (H3)

$$\% H3 = \frac{\overline{F_1} - \overline{\text{Check}}}{\overline{\text{Check}}} \times 100$$

Results and discussion:

Pooled Analysis of variance:

The analysis of variance for yield and its components traits in pooled analysis revealed that the mean square values due to genotypes were highly significant for all the characters except maximum photosystem II efficiency, which indicated existence of sufficient genetic variability in the experimental material for all the characters except maximum photosystem II efficiency (Table 1).

Comment [R3]: What about other source of variations as the main aspect is heterosis so discuss about other variations also... Explain it more in detail as per table 2 with proper discussion . .it's not enough

Mean performance:

Mean values of grain yield and yield component characters of parents (lines and testers) and their hybrids is presented in table 2. The range of heterobeltiosis and standard heterosis as well as the number of hybrids showing significant heterosis in desirable direction is presented in table 3 and list of top 5 hybrids for various characters showing high range of heterosis in table 4.

Comment [R4]: Explain and discuss about mean performance...hybrids mean for what traits showing high or low performance w.r.t parents

Estimation of Heterobeltiosis and Standard Heterosis:

Heterobeltiosis (HB) and standard heterosis (SH) in negative direction for days to 50% flowering are desirable for earliness. The hybrid 274A × 123R (-16.58%) showed the least estimate of HB followed by hybrids 04999A × 123R (-14.29%), 252A × 124R and 264A × 124R (-11.78%). One hybrid 274A × 123R (-4.39) registered significant and negative SH. The results are in accordance with findings of (Athoniet *et al.* (2022), Kumar *et al.* (2017) for SH. Negative estimates of heterobeltiosis (HB) and standard heterosis (SH) for days to maturity of grain is desired to escape terminal drought. The estimates of HB ranged from -11.78 (274A × 123R) to 0 per cent (252A × 132R). The cross 274A × 123R (-11.78) exhibited the least heterotic effect followed by 260A × 124R (-10.27). One hybrid 274A × 123R (-2.24) registered significant and negative SH. The results are in accordance with findings of Kumar *et al.* (2017) for SH. The minimum and maximum values for heterobeltiosis were -14.04 (242A × 124R) and 26.91 (269A × 132R) per cent for plant height. For effective tillers per plant minimum and maximum values of heterobeltiosis were -41.18 (264A × 124R) and 114.29 (274A × 123R) per cent, respectively. High heterosis for productive tillers per plant were also reported by Rafiq *et al.* (2016). For flag leaf length, the heterobeltiosis ranged from -34.47 (274A × 123R) to 6.68 (242A × 124R). High significant positive SH reported by 246A × 124R. Similar findings were reported by Pareek *et al.* (2020). For flag leaf width heterobeltiosis ranged from -42.09 (264A × 124R) to 36.02 (274A × 123R). For leaf length heterobeltiosis ranged from -18.75 (242A × 123R) to 20.27 (274A × 123R), for leaf width heterobeltiosis range between -14.63 (246A × 124R) to 21.37 (843-22A × 132R and 264A × 132R). The actual photosystem II efficiency recorded highest SH in 252A × 132R (51.7) and about 35 hybrids showing high significant positive SH. The better parent heterosis for panicle length ranged between -23.11 (246A × 132R) and 20.17 (260A × 123R). Two hybrids recorded significant positive heterobeltiosis (15.54) 260A × 132R and (20.17) 260A × 123R. Only one hybrid recorded significant positive SH in panicle width i.e., 260A × 132R (18.37). Similar results reported by Kumar *et al.* (2017). In case of fresh biomass, about 17 hybrids recorded significant positive heterobeltiosis of which highest was recorded by 274A × 123R (44.13). Similar findings reported by Srivastava *et al.* (2020). For dry biomass, about 15 hybrids recorded significant positive heterobeltiosis of which highest was recorded by 274A × 123R (75.61). Similar results shown by Jethva *et al.* (2012). Highest better parent heterosis for grain yield was recorded by 274A × 123R (46.01) followed by 246A × 123R (32.71). For harvest index, the better parent heterosis recorded the highest for 843-22A × 132R (47.50), 1000 seed weight showed the range of heterobeltiosis between -28.42 (242A × 124R) and 48.32 (274A × 123R). None of the hybrids recorded high significant positive SH for characters such as plant height, effective tillers per plant, flag leaf width, leaf length and width, fresh biomass, dry biomass, harvest index and 1000 seed weight.

Comment [R5]: Give the table reference in this section

Conclusion:

The highest better parent was exploited by the cross 274A × 123R for almost all the characters under study except for flag leaf length (246A × 124R) and panicle length (260A × 123R). The five hybrids namely 274A × 123R, 260A × 124R, 252A × 124R, 269A × 124R and 246A × 123R recorded highest values for better parent and standard heterosis for grain yield and also for most of the traits. These hybrids can be further recommended for yield improvement.

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Table 1. Pooled Analysis of Variance for combining ability (Line × Tester) for grain yield and other traits in pearl millet

Source of Variation	d.f	Days to 50% flowering	Days to Maturity	Plant height (cm)	Effective Tillers per plant	Flag leaf length (cm)	Flag leaf Width (cm)	Leaf length (cm)	Leaf width (cm)
Environments	1	502.170**	53.542**	21291.690**	23.334**	234.194**	5.242**	110.641	2.834**
Rep * Env.	2	41.199**	2.366	183.285	0.099	10.119	0.154	32.497	0.129
Treatments	50	33.979**	67.386**	2289.662**	2.506**	83.162**	2.173**	99.543**	0.601**
Parents	14	52.892**	29.290**	1015.925**	2.493**	27.612**	1.328**	50.721	0.323**
Parent vs Crosses	1	234.964**	1477.869**	65595.480**	34.035**	1400.933**	45.660**	1584.590**	11.216**
Crosses	35	20.671**	42.325**	990.419**	1.610**	67.731**	1.269**	76.643**	0.409**
Line effect	11	35.045*	47.035	1181.756	0.922	74.627	1.312	111.929	0.531
Tester effect	2	13.644	17.810	101.772	0.760	24.436	0.206	3.252	0.019
Line * Tester effect	22	14.123**	42.199**	975.537**	2.032**	68.219**	1.343**	65.671**	0.383**
Env * Parents	14	21.854**	9.500**	578.592**	2.469**	21.070*	0.435**	10.359	0.061
Env * Parent vs Cross	1	121.083**	0.851	5163.368**	0.271	3.912	0.154	1.196	0.019
Env * Crosses	35	21.994**	36.932**	1120.525**	1.834**	70.036**	1.417**	75.598**	0.616**
Env * Line effect	11	44.446**	33.934	904.889	1.205	43.207	1.047	48.763	0.398
Env * Tester effect	2	7.764	82.394	3383.483	3.212	133.036	2.622	155.802	1.105
Env * L * T effect	22	12.062**	34.298**	1022.619**	2.024**	77.723**	1.492**	81.724**	0.680**
Error	200	5.216	1.648	147.937	0.203	10.998	0.160	30.152	0.140
Total	305	14.833	17.003	715.805	0.948	31.161	0.663	45.874	0.274
σ^2_{gca}		0.455	0.680	10.359	0.013	0.819	0.012	0.487	0.002
σ^2_{sca}		1.705	6.728	133.320	0.297	9.257	0.188	5.002	0.036
$\sigma^2_{gca} / \sigma^2_{sca}$		0.26	0.10	0.07	0.04	0.08	0.06	0.09	0.05
$(2 \sigma^2_{gca}) / (2 \sigma^2_{gca} + \sigma^2_{sca})$		0.34	0.16	0.13	0.08	0.15	0.11	0.16	0.10

Comment [R6]: Format other tables also

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Table 1 (continued.). Pooled Analysis of Variance for combining ability (Line × Tester) for grain yield and other traits in pearl millet

Source of Variation	d.f	Actual PS II efficiency	Maximum PS II efficiency	Panicle length (cm)	Panicle width (cm)	Fresh biomass (kg/plot)	Dry biomass (kg/plot)	Grain Yield per plant (kg)	Harvest index (%)	1000 seed weight (g)
Environments	1	0.066**	1.520**	590.000**	46.667**	2.400**	0.234**	0.161**	470.059**	19.065**
Rep * Env.	2	0.001	0.003	17.041	0.629*	0.030	0.005	0.000	29.309	0.161
Treatments	50	0.038**	0.005	30.159**	0.428**	2.218**	0.139**	0.096**	472.301**	19.000**
Parents	14	0.010**	0.007	45.422**	0.736**	1.424**	0.102**	0.052**	441.314**	9.622**
Parent vs Crosses	1	0.690**	0.003	41.939*	0.330	46.934**	2.619**	2.088**	9448.069**	441.508**
Crosses	35	0.030**	0.005	23.717**	0.308*	1.257**	0.083**	0.056**	228.246**	10.680**
Line effect	11	0.031	0.004	51.349**	0.643**	1.293	0.088	0.061	236.145	9.827
Tester effect	2	0.006	0.007	30.638	0.588*	0.268	0.013	0.016	59.963	3.876
Line * Tester effect	22	0.031**	0.005	9.272	0.115	1.330**	0.087**	0.058**	239.595**	11.725**
Env * Parents	14	0.009**	0.006	10.184	0.178	0.364**	0.047**	0.013**	99.579**	3.859**
Env * Parent vs Cross	1	0.0001	0.001	90.848**	0.484	0.126	0.031**	0.002	133.163	5.148**
Env * Crosses	35	0.031**	0.005	11.779**	0.275	1.041**	0.079**	0.049**	179.031**	9.684**
Env * Line effect	11	0.024	0.004	12.734	0.240	0.603	0.044	0.03	89.404	5.446
Env * Tester effect	2	0.058	0.0009	26.017	0.537	2.456	0.201	0.152	542.363	26.837
Env * L * T effect	22	0.032**	0.005	10.007	0.268	1.131**	0.086**	0.050**	190.814**	10.243**
Error	200	0.000	0.007	6.674	0.197	0.048	0.004	0.003	43.796	0.261
Total	305	0.011	0.011	13.537	0.411	0.540	0.038	0.024	133.584	4.656
σ^2_{gca}		0.000	0.000	0.746	0.009	0.016	0.001	0.001	2.850	0.146
σ^2_{sca}		0.005	0.000	0.309	-0.012	0.212	0.014	0.009	36.630	1.904
$\sigma^2_{gca}/\sigma^2_{sca}$		0.00	0.00	2.41	-0.75	0.07	0.07	0.11	0.07	0.07
$(2\sigma^2_{gca})/(2\sigma^2_{gca} + \sigma^2_{sca})$		0	0	0.82	3	0.13	0.12	0.18	0.13	0.12

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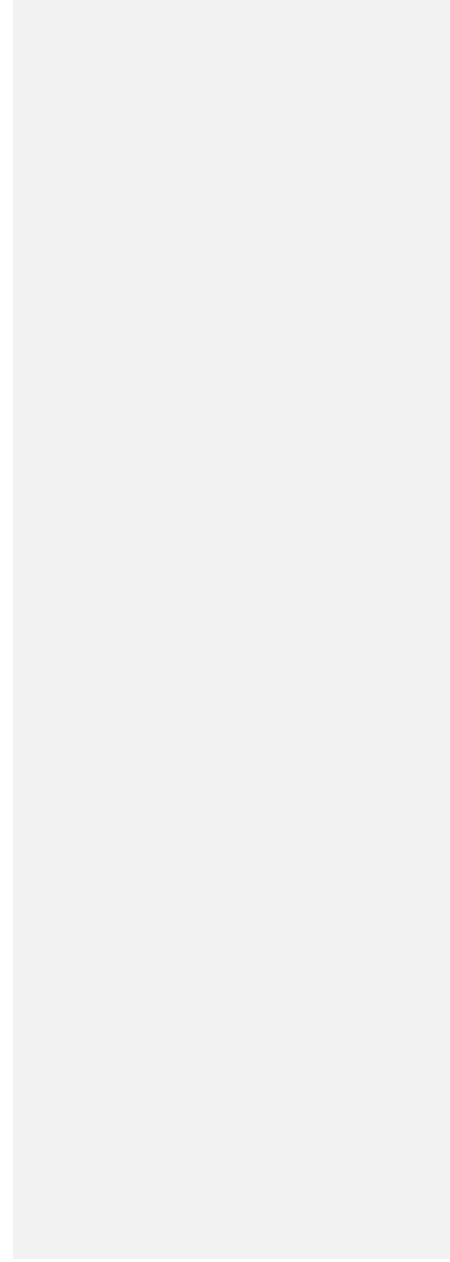


Table 2: Mean values of lines, testers and hybrids

Characters	Mean values				
	lines	testers	hybrids	CD at 5%	CV
Days to 50% flowering	59.91	61.88	58.42	3.80	4.01
Days to maturity	98.45	94.39	92.81	2.07	1.37
Plant height	105.17	132.77	142.82	19.92	9.22
Effective tillers per plant	1.92	2.39	2.74	0.73	17.7
Flag leaf length	37.91	34.69	32.57	5.33	9.79
Flag leaf width	1.79	2.73	2.82	0.64	15.33
Leaf length	38.92	44.55	45.04	8.91	12.63
Leaf width	2.85	3.19	3.34	0.62	11.83
Actual PS II efficiency	0.79	0.70	0.66	0.03	2.73
Panicle length	18.78	19.94	19.82	4.20	13.28
Panicle width	2.69	2.57	2.59	0.72	17.17
Fresh biomass	1.10	1.94	2.12	0.35	11.59
Dry biomass	0.22	0.45	0.47	0.10	15.31
Grain yield	0.33	0.48	0.54	0.09	11.10
Harvest index	60.72	53.74	54.27	7.86	8.78
1000 seed weight	6.13	8.33	9.20	0.85	6.20

Table.3 Range of heterosis and number of crosses showing significant heterosis in required direction in pearl millet

Characters	Heterobeltiosis (%)		Standard heterosis (%)	
	Range	No of significant hybrids	Range	No of significant hybrids
Days to 50% flowering	-16.58 to 5.83	29	-4.39 to 9.36	1
Days to maturity	-11.78 to 0.00	35	-2.24 to 9.70	1
Plant height	-14.04 to 26.91	15	-34.88 to -6.51	-
Effective tillers per plant	-41.18 to 114.29	18	-52.38 to 7.14	-
Flag leaf length	-34.47 to 6.68	1	-7.16 to 40.80	25
Flag leaf width	-42.09 to 36.02	7	-55.53 to -4.99	-
Leaf length	-18.75 to 20.27	1	-35.49 to -4.51	-
Leaf width	-14.63 to 21.37	5	-28.39 to -1.25	-
Actual PS II efficiency	-34.21 to 1.72	31	-0.25 to 51.7	-
Panicle length	-23.11 to 20.17	2	-31.39 to 5.84	-
Panicle width	-21.85 to 10.11	1	-18.98 to 18.37	1
Fresh biomass	-48.73 to 65.61	17	-59.37 to 0.86	-
Dry biomass	-63.43 to 78.92	15	-66.35 to 1.65	-
Grain yield	-37.67 to 48.97	18	-49.23 to 6.45	-

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Harvest index	-45.51 to 47.50	15	-47.99 to 6.16	-
1000 seed weight	-28.42 to 48.32	22	-40.34 to 6.01	1

Table 4. Standard Heterosis, Better parent and Mid- parent heterosis for top five crosses for each trait in pearl millet

Character/ Cross	Standard Heterosis	BP Heterosis	MP Heterosis
1. Days to 50% Flowering			
274A × 123R	-4.39*	-16.58**	-12.21**
274A × 132R	-2.63	-5.67**	-3.90*
04999A × 123R	-1.75	-14.29**	-12.27**
252A × 124R	-1.46	-11.78**	-8.55**
264A × 124R	-1.46	-11.78**	-7.03**
2. Days to maturity			
274A × 123R	-2.24*	-11.78**	-9.50**
252A × 123R	-1.12	-8.93**	-7.50**
260A × 124R	-0.56	-10.27**	-7.30**
269A × 124R	-0.19	-7.76**	-5.81**
04999A × 124R	1.49	-8.57**	-5.47**
3. Plant Height (cm)			
269A × 124R	-6.51	17.54**	31.37**
274A × 123R	-7.26	26.20**	40.92**
252A × 124R	-10.00*	13.16*	29.00**
260A × 124R	-10.93*	11.99*	32.53**
04999A × 124R	-11.26**	11.58*	29.36**
4. Effective tillers per plant			
274A × 123R	7.14	114.29**	87.50**
269A × 124R	0	100.00**	44.83**
252A × 124R	0	23.53*	42.37**
264A × 132R	-4.76	90.48**	116.22**
260A × 124R	-4.76	17.65	48.15**
5. Flag leaf length (cm)			
246A × 124R	40.80**	-3.55	6.71
264A × 124R	38.64**	-4.83	5.2
221A × 124R	38.09**	-4.81	5.02
843-22A × 132R	37.96**	-4.49	1.02
260A × 124R	36.79**	-1.51	-0.85
6. Flag leaf width (cm)			
274A × 123R	-4.99	36.02**	62.83**
252A × 124R	-8.03	19.77*	41.81**
269A × 124R	-10.63	16.38	36.42**
843-22A × 132R	-12.8	30.52**	63.75**
260A × 124R	-13.02	13.28	46.35**
7. Leaf length (cm)			
274A × 123R	-4.51	20.27*	28.91**
260A × 123R	-8.87	14.77	21.56**
252A × 124R	-9.32	8.26	14.97*
269A × 132R	-10.38	14.84	18.73**
269A × 124R	-11.13	6.1	13.44
8. Leaf width (cm)			
274A × 123R	-1.25	21.28**	30.12**
260A × 124R	-4.38	11.71	21.97**
252A × 124R	-5.85	10	18.06**
262A × 123R	-9.81	10.77	12.79*

*and**representssignificanceat5%and1%respectively.

Table 4 (continued).Standard Heterosis, Better parent and Mid- parent heterosis for top five

269A × 132R	-10.02	16.49*	19.56**
9. Actual PS II efficiency (ΦPSII)			
252A × 132R	51.7**	1.72	2.15
264A × 124R	49.5**	-1.18	8.94**
242A × 124R	47.9**	-2.25	7.77**
246A × 124R	50.4**	-0.4	9.74**
843-22A × 123R	47.9**	-2.04	3.53**
10. Panicle length (cm)			
260A × 132R	5.84	15.54*	19.83**
260A × 123R	2.19	20.17*	25.84**
291A × 132R	0.73	-3.16	2.99
269A × 123R	-1.09	-2.52	10.61
04999A × 132R	-1.82	7.17	10.7
11. Panicle width (cm)			
260A × 132R	18.37*	3.69	11.81
269A × 123R	8.13	-14.73*	2.87
242A × 132R	5.72	8.33	9.35
260A × 123R	5.42	-7.65	6.71
269A × 132R	4.52	-17.58*	-6.85
12. Fresh biomass (kg/plot)			
252A × 124R	0.86	27.27**	48.94**
274A × 123R	-1.73	44.13**	90.72**
246A × 123R	-3.17	42.01**	109.61**
269A × 124R	-3.46	21.82**	48.89**
262A × 123R	-4.9	39.48**	58.81**
13. Dry biomass (kg/plot)			
274A × 123R	1.65	75.61**	136.07**
252A × 124R	0	8.7	50.18**
269A × 124R	-12.24*	-4.6	30.65**
246A × 123R	-12.94*	50.41**	119.58**
260A × 124R	-15.06**	-7.67	39.11**
14. Grain yield (kg/plot)			
274A × 123R	6.45	46.01**	81.24**
260A × 124R	2.81	26.21**	61.97**
252A × 124R	1.49	24.60**	46.53**
269A × 124R	0.46	23.34**	42.95**
246A × 123R	-3.25	32.71**	77.06**

crosses for each trait in pearl millet

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*and**representssignificanceat5%and1%respectively

15. Harvest index (%)			
274A × 123R	6.16	41.18**	77.42**
252A × 124R	5.96	11.00*	37.08**
269A × 124R	-7.36	-2.95	17.74**
843-22A × 132R	-7.90	47.50**	74.33**
246A × 123R	-8.13	22.17**	65.34**
16. 1000 seed weight (g)			
274A × 123R	6.01	48.32**	75.22**
252A × 124R	0.38	20.43**	39.29**
269A × 124R	-3.88	15.31**	32.20**
262A × 123R	-4.38	33.78**	42.58**
246A × 123R	-4.95	32.98**	51.69**

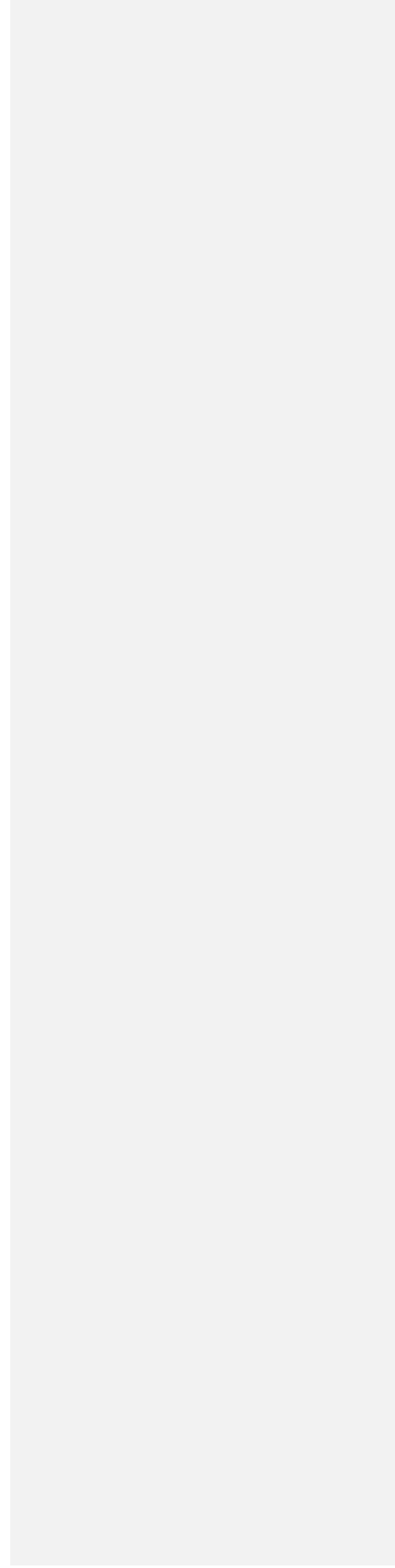
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