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GENETIC CHARACTERIZATION OF NORTH-WEST HIMALAYAN LANDRACES AND ESTABLISHED DOUBLED HAPLOIDS OF BREAD WHEAT (*Triticum aestivum* L.)

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ABSTRACT

Wheat is one of the most important cereal crops and is the staple food of millions of people. Understanding the evolutionary linkages and diversity patterns among the germplasm is crucial for improving wheat and ensuring the long-term sustainability of important genetic resources. Therefore, the objective of this study is to interpret genetic diversity and identify various genotypes for future breeding programmes. 50 genotypes of bread wheat, including native landraces, cultivars and developed doubled haploids, were evaluated in this investigation for quality profiling and various agro-morphological parameters. Grain yield and its component traits showed sufficient genetic variability indicating the scope of selection for these traits. For harvest index, high PCV and GCV, along with high heritability and moderate genetic advance were observed and grain yield per plant showed high PCV and moderate GCV coupled with high heritability and moderate genetic advance. D² statistics analysis grouped 50 wheat genotypes into 3 clusters. Maximum genotypes were placed in cluster I. Principal component analysis revealed 78.45 per cent variation elaborated by the first five significant principal components. On the basis of mean performance for grain yield, three genotypes viz., DH 114, DH 776 and T 32 were found to be statistically at par. Quality analysis revealed that protein content was observed highest for genotype BK-2 followed by DH 100 and wet gluten was found highest in DH-51 followed by DH 84 and DH 7, whereas genotype DH 1 showed highest glute strength followed by DH 51.

Keywords : Wheat, diversity, heritability, double haploids, landraces, quality, yield

Introduction

Bread wheat (*Triticum aestivum* L.) is a segmental allohexaploid ($2n = 6x = 42$, AABBDD genome), annual and self-pollinated cereal crop. Millions of Indians rely on it as a staple diet, especially in the northern and northwestern regions of the country. It is the most significant food grain of India, second only to rice. Bread wheat is cultivated in various agro-climatic regions of north-west Himalayas encompassing the hill states viz., Himachal Pradesh, Jammu & Kashmir and Uttarakhand. Most of the area of these states is under rainfed cultivation. Wheat provides a superior source of nourishment in terms of carbohydrates, minerals and proteins, making it a special gift from nature to humanity. It contains 75–80% carbohydrates, 9–18% protein, fibre, vitamins, calcium, iron and many macro and micro-nutrients.

Genetic base of wheat has been reduced over time by the widespread monoculture of elite cultivars, which has also made the crop more susceptible to a variety of biotic and abiotic challenges. Hence, genetic improvement should be based on the exploitation of the genetic diversity in the existing and untapped gene pools, making diversity studies imperative for future wheat breeding programmes. Utilizing new variety is crucial to overcoming limited genetic base in wheat. Since landraces have a wider genetic base and can thus offer desirable traits, they have emerged as a result of both natural selection and artificial selection by the farmers. In addition to enhancing agronomic and quality qualities in breeding programs, wheat landraces are a useful genetic material for diversity and specialized adaptability to local environmental conditions. Hence, there is an urgent need to assess the

variability amongst these gene pools so as to utilize the novel genetic resource.

Wheat quality has been linked to reduced protein and gluten content. The processing properties of wheat are largely determined by gluten proteins. Beccari was the first to successfully isolate gluten proteins around the mid-eighteenth century (Bailey, 1941). Gliadins and glutenins make up gluten, which makes up around 80% of the endosperm protein. While gliadins primarily affect the extensibility and viscosity of the dough, glutenins were found to impart elasticity. (Finney 1943; Wall 1979). Other factors determining the quality of processed wheat food products are grain storage protein (about 80% of the grain total protein) content and composition (Shewry, 2002). Protein content has been consistently considered as the primary determinant of wheat bread quality, (Shewry, 1994), therefore protein quality is also an important character to be considered.

Using genetic distance to estimate genetic diversity, parental selection in wheat breeding can be used to promote new genetic recombination and enhance grain production. An efficient breeding programme must take into account the presence of genetic diversity. Genetic diversity available in the existing germplasm determines the success of any crop improvement programme. In order to evaluate the degree of genetic distance between genotypes, a cluster analysis is a suitable tool for establishing family relationships and genetic affinity. D^2 Statistical analysis is an effective method for determining the degree of population divergence. The objective of principle component analysis is to preserve most of the information in the original variables while creating a finite number of linear combinations (principal components) of a set of variables. Identifying wheat lines with high yielding and quality-enhancing traits will undoubtedly be made easier by the researcher's understanding of the patterns of current genetic variability, the trend of character association, the identification of promising traits and the degree of genetic divergence.

The primary breeding target for wheat improvement continues to be yield enhancement, with the demand to incorporate targets for grain quality trait enhancement in breeding program to cater for unique and distinct quality feature requirements for various types of wheat-based products. Besides assessing variability, genetic relationship amongst the various traits will be worked out for isolation of potential genotypes to be used as such or for future breeding programmes so as to introgress the targeted novel genes to mitigate the biotic & abiotic stresses posed by

changing climatic scenario. Hence, the present investigation was undertaken to assess the genetic diversity among various north-west Himalayan potential landraces, winter $\times$ spring and spring $\times$ spring wheat derived doubled haploids for various quality and yield-related traits.

Materials and Methods

The experimental material comprises 50 diverse bread wheat genotypes (Table 1). This includes 29 potential landraces collected from the north-western Himalayas, 16 established doubled haploids by chromosome elimination technique (Chaudhary *et al.* 2005) and five popular Indian cultivars viz., HPW 360, HPW 368, HPW 373, VL 892 and C 306. The 50 genotypes were evaluated for different morpho-physiological traits in Randomized Block Design with three replications during rabi 2021-2022. Two rows of 1 m length for each genotype were grown with a spacing of 20 $\times$ 10 cm. Recommended package of practices were followed for raising the crop. Observations were recorded for several morpho-physiological traits viz., Plant height (cm), Effective tillers per plant, Spikelets per spike, Grains per spike, 1000-grain weight (g), Biological yield per plant (g), Harvest index (%), Grain yield per plant (g); and for days to 50% flowering and 75% maturity, which were recorded on a plot basis. Quality parameters namely Moisture content (%), Total ash content (%), Crude fibre content (%), Crude protein content (%), Crude fat content (%), Wet glucan content (%) and Glucan strength (ml) were also estimated using standard procedures. Various statistical analyses were performed on the observations recorded for the various yield and yield contributing characteristics. The model proposed by Panse and Sukhatme (1984) was used to analyze the data for the analysis of variance. The estimation of the genotypic, phenotypic and environmental coefficients of variation was done in accordance with Burton and De Vane (1953). Heritability in broad sense (h^2 bs) was calculated as per the following formula given by Burton and De Vane (1953) and Johnson *et al.* (1955). The expected genetic advance (GA) resulting from the selection of 5 per cent superior individuals was calculated as per Burton and De Vane (1953) and Johnson *et al.* (1955). A measure of group distance based on multiple characters was given by Mahalanobis (1936). Using D^2 values, different genotypes were grouped into various clusters following Tocher's method as suggested by Rao (1952).

Results and Discussion

Significant variations between the genotypes for each assessed agronomic attribute were revealed by the analysis of variance, indicating the presence of significant variation for the traits under study (Table 2). High variability for different traits in wheat has earlier been reported by Patial *et al.* (2021) and Tomar *et al.* (2019). The various parameters of variability viz., phenotypic coefficient of variation (PCV) and genotypic coefficient of variation (GCV) along with heritability in broad sense (h^2_{bs}) and genetic advance (GA) were estimated and expressed as percentage of mean for different traits are presented in Table 3 & Fig 1. For every trait, the estimate of PCV was greater than the corresponding value of GCV, suggesting that environmental factors in addition to genetic factors are responsible for the observed variation.

Highest PCV and GCV were observed for tillers per plant, harvest index, crude fibre, gluten strength and grain yield per plant, whereas, low estimates of PCV and GCV were recorded for days to 50% flowering, days to 75% maturity, 1000-grain weight, biological yield per plant, moisture content and wet gluten content. Similar findings were reported by Singh *et al.* (2014) and Nukasani *et al.* (2013) for several traits.

Heritability in broad sense h^2 (b.s) was observed highest for days to 50% flowering, days to 75% maturity, plant height, spikelets per spike, grains per spike, harvest index, moisture content, total ash content, crude fibre, crude protein, crude fat, gluten strength and grain yield per plant; and moderate (50-70%) for tillers per plant, 1000-grain weight and wet gluten content. These findings corroborate the results of Kumar *et al.* (2017) and Ahmad *et al.* (2016) for grains per spike and plant height.

Genetic advance as percent of mean was found high for plant height, tillers per plant, spikelets per spike, harvest index, total ash content, crude fibre, crude protein, crude fat, gluten strength and grain yield per plant. High heritability along with high genetic advance was noted for spikelets per spike, harvest index, total ash content, crude fibre, crude protein, crude fat, gluten strength and grain yield per plant. These results are in conformity with Prakash and Keketta 2023.

Genetic diversity among 49 wheat genotypes was studied using the Mahalanobis D^2 approach, which was

based on thirteen quantitative features. Using the Tocher's technique, the 50 genotypes were divided into three clusters with the criterion that intra-cluster average D^2 values be fewer than inter-cluster D^2 values. The distribution of 50 wheat genotypes in three clusters is presented in Table 4. Cluster 1 has the highest number of genotypes, followed by Cluster 2 with seven genotypes and Cluster 3 with only one genotype. The estimates of average intra and inter cluster distance for the three clusters is presented in Table 5. Highest inter-cluster distance was observed between cluster II and cluster III (1450.85), followed by cluster I and cluster III (717.16). The highest intra-cluster distance was observed for cluster I (227.82) followed by cluster II (185.01), thus genotypes within the same cluster were relatively different. These results are in conformity with Elahi *et al.* (2021) and Rajaneesh *et al.* (2019).

The per cent contribution towards genetic divergence by all the 17 characters is presented in Table 7. Among all the characters, crude protein (33.06%) followed by moisture content (25.88%) showed the highest percent contribution towards total genetic divergence. The first five principal components explained a total of 78.45% per cent variation. PC1 was the most significant and explained 43.41 per cent of the total variance which was mainly contributed by crude fat. PC2 accounted for 11.47 per cent variation and was attributed mainly due to harvest index (Table 8). The scatter biplot of PC1 against PC2 showed that the majority of genotypes were distinct, as they were dispersed throughout the diagram (Fig. 2.)

Conclusion

The analysis of variance revealed significant differences among genotypes for all morpho-physiological traits studied. High heritability along with high genetic advance was observed for spikelets per spike, harvest index, total ash content, crude fibre, crude protein, crude fat, gluten strength and grain yield per plant thus these character show additive gene action in their expression. Crude protein and moisture content contributed maximum towards genetic divergence. D^2 statistical analysis grouped genotypes into three clusters. Maximum genotypes, i.e., 42 genotypes were placed in cluster I. Protein content was highest for genotype BK-2 followed by DH-100. Genotype DH-1 showed highest gluten strength followed by DH-51. Hence, these superior genotypes can be further utilized in hybridization programmes.

Table 1 : Details of material used in the present study

S. No.	Wheat Genotypes	Source
1	DH 1	CSKHPKV, Palampur
2	DH 3	CSKHPKV, Palampur
3	DH 7	CSKHPKV, Palampur
4	DH 37	CSKHPKV, Palampur
5	DH 40	CSKHPKV, Palampur
6	DH 51	CSKHPKV, Palampur
7	DH 52	CSKHPKV, Palampur
8	DH 59	CSKHPKV, Palampur
9	DH 65	CSKHPKV, Palampur
10	DH 84	CSKHPKV, Palampur
11	DH 86	CSKHPKV, Palampur
12	DH 100	CSKHPKV, Palampur
13	DH 114	CSKHPKV, Palampur
14	DH 195	CSKHPKV, Palampur
15	DH 206	CSKHPKV, Palampur
16	DH 776	CSKHPKV, Palampur
17	Mandi landrace 3	-
18	Mandi landrace 4	-
19	Mandi landrace 5	-
20	Mandi landrace 10	-
21	Kinnaur landrace 12	-
22	Mandi landrace 19	-
23	Kullu landrace 20	-
24	Kullu landrace 21	-
25	Kullu landrace 22	-
26	Kullu landrace 26	-
27	Kullu landrace 31	-
28	Kullu landrace 32	-
29	Kullu landrace 39	-
30	Kullu landrace 41	-
31	Kullu landrace 47	-
32	Kullu landrace 50	-
33	Kullu landrace 51	-
34	Chamba landrace 60	-
35	Kinnaur landrace 64	-
36	Chamba landrace 71	-
37	Chamba landrace 78	-
38	Chamba landrace 79	-
39	Chamba landrace 93	-
40	Chamba landrace 94	-
41	Chamba landrace 95	-
42	Chamba landrace 96	-
43	Chamba landrace 101	-
44	BK 1	-
45	BK 2	-
46	HPW 360	WL 711/HPW 89S
47	HPW 368	NAC/TH.AC//3 × PVN/3/MIRLO/BUC/4/2 × PASTOR
48	HPW 373	MILAN//BUC/CHRC/3/METSO
49	VL 892	WH 542/PBW 226
50	C 306	REGENT 1974/3 × CHZ// × 2C591/3/P19/C 281

Table 2 : Analysis of variance for different traits in wheat genotypes

	Traits	Source	Mean sum of squares		
			Replication	Genotype	Error
		df	2	49	98
1	Days to 50% flowering		72.65	73.43*	6.39
2	Days to 75% maturity		1.32	137.57*	2.48
3	Plant height (cm)		9.92	581.21*	34.49
4	Tillers per plant		0.94	1.78*	0.34
5	Spikelets per spike		2.40	29.25*	2.36
6	Grains per spike		0.74	177.15*	18.82
7	1000-grain weight (g)		32.52	73.06*	11.33
8	Grain yield per plant (g)		0.58	6.52*	0.59
9	Biological yield per plant (g)		11.02	21.54 *	7.80
10	Harvest index (%)		1.55	120.43*	13.96
11	Moisture content (%)		0.24	3.78*	0.05
12	Total ash content (%)		0.02	0.28*	0.01
13	Crude fibre (%)		0.23	0.23*	0.02
14	Crude protein (%)		0.08	8.94*	0.13
15	Crude fat (%)		0.01	0.19*	0.001
16	Wet gluten content (%)		1.81	28.78*	5.39
17	Gluten strength (ml)		0.31	92.62*	1.77

*P ≤ 0.05

Table 3 : Estimates of parameters of variability for various traits in wheat genotypes

Traits	Range	Mean ± SE	GCV (%)	PCV (%)	h ² (b.s) (%)	GA as (%) of mean
Days to 50% flowering	144.00-121.67	130.27±2.06	3.62	4.11	77.77	6.59
Days to 75% maturity	181.00-130.33	164.99±1.28	4.06	4.17	94.77	8.15
Plant height (cm)	106.44-58.72	78.69±4.79	17.15	18.70	84.09	32.40
Tillers per plant (no.)	5.12-2.28	3.24±0.47	21.38	28.01	58.28	33.63
Spikelets per spike	21.07-10.06	15.51±1.25	19.29	21.68	79.16	35.35
Grains per spike	75.50-38.05	53.11±3.54	13.67	15.93	73.71	24.18
1000-grain weight (g)	54.90-34.75	42.45±2.74	10.68	13.30	64.49	17.67
Biological yield per plant (g)	31.83-19.78	26.17±2.28	8.17	13.44	37.00	2.68
Harvest index (%)	35.36-10.37	22.25±3.05	26.76	31.59	71.77	46.71
Moisture content (%)	14.73-10.57	12.03±0.18	9.25	9.44	95.97	18.67
Total ash content (%)	2.07-1.16	1.29±0.06	23.24	24.03	93.53	46.31
Crude fibre (%)	2.04-0.74	1.18±0.11	22.62	25.52	78.56	41.30
Crude protein (%)	12.44-7.03	9.46±0.28	18.11	18.50	95.92	36.55
Crude fat (%)	1.60-0.72	1.08±0.07	22.78	24.11	89.26	44.35
Wet gluten content (%)	35.22-22.13	28.85±1.89	9.67	12.58	59.13	15.32
Gluten strength (ml)	27.17-16.00	22.46±1.08	24.50	25.20	94.48	49.06
Grain yield per plant (g)	8.82-2.68	5.71±0.55	24.57	28.02	76.94	44.14

Table 4 : Distribution of genotypes among different clusters on the basis of Mahalanobis D² – analysis

Clusters	Number of genotypes	Genotypes
I	42	T-71, T-93, T-5, T-10, T-31, T-96, T-95, T-78, T-21, T-22, T-101, T-79, T-20, T-3, T-4, T-94, T-26, BK-1, T-12, T-64, T-50, DH-40, DH-86, DH-84, T-60, T-39, T-32, T-51, T-47, T-41, DH-10, DH-51, DH-37, DH-59, DH-52, DH-3, DH-65, BK 2, DH-195, DH-7, DH-776, DH-1
II	07	HPW-373, C-306, HPW-368, HPW-360, VL-892, DH-206, DH-114
III	01	T-19

Table 5 : Average intra and inter-cluster distance among three clusters

Cluster	I	II	III
I	227.82	622.01	717.16
II		185.01	1450.85
III			00.00

Table 6 : Cluster means of six clusters for different traits of wheat genotypes

Traits	I	II	III	Mean	Max	Min
Days to 50% flowering	129.35	136.19	126.00	131.00	136.19	126.00
Days to 75% maturity	165.29	168.14	130.33	155.00	168.14	130.33
Plant height (cm)	76.27	93.45	77.33	82.40	93.45	76.27
Tillers per plant (no.)	3.11	4.10	2.88	3.36	4.10	2.88
Spikelets per spike	15.12	18.25	13.09	15.50	18.25	13.09
Grains per spike	52.22	58.87	50.33	53.80	58.87	50.33
1000-grain weight (g)	41.33	48.08	50.40	46.60	50.40	41.33
Biological yield per plant (g)	26.43	24.48	27.34	26.10	27.34	24.48
Harvest index (%)	21.15	29.31	19.34	23.30	29.31	19.34
Moisture content (%)	12.13	11.56	11.79	11.80	12.13	11.56
Total ash content (%)	1.20	1.94	1.14	1.430	1.94	1.14
Crude fibre (%)	1.10	1.66	1.13	1.30	1.66	1.13
Crude protein (%)	9.11	11.84	7.58	9.51	11.84	7.58
Crude fat (%)	1.02	1.52	0.96	1.17	1.52	0.96
Wet gluten content (%)	28.25	33.08	24.81	28.70	33.08	24.81
Gluten strength (ml)	20.69	33.45	19.83	24.70	33.45	19.83
Grain yield per plant (g)	5.50	7.12	5.32	5.98	7.12	5.32

Table 7 : Relative contribution (%) of individual trait to the genetic divergence among wheat genotypes

S. No.	Traits	Contribution (%)
1	Days to 50% flowering	0.98
2	Days to 75% maturity	12.24
3	Plant height (cm)	0.73
4	Tillers per plant (no.)	0.08
5	Spikelets per spike	0.49
6	Grains per spike	1.55
7	1000-grain weight (g)	0.41
8	Biological yield per plant (g)	0.08
9	Harvest index (%)	1.63
10	Moisture content (%)	25.88
11	Total ash content (%)	9.22
12	Crude fibre (%)	0.73
13	Crude protein (%)	33.06
14	Crude fat (%)	1.39
15	Wet gluten content (%)	0.57
16	Gluten strength (ml)	10.86
17	Grain yield per plant (g)	0.08

Table 8 : Eigenvectors for the first five components of different traits in 50 genotypes of Wheat

	PC1	PC2	PC3	PC4	PC5
Days to 50% flowering	0.222	-0.236	0.304	-0.104	0.180
Days to 75% maturity	0.121	-0.222	0.571	0.002	0.283
Plant Height (cm)	0.306	-0.003	-0.146	-0.049	-0.081
No. of tillers per plant	0.291	0.036	0.018	-0.133	0.023
Spikelets per spike	0.299	-0.198	0.095	-0.220	-0.211
Grains per spike	0.110	0.334	0.090	-0.373	0.306
1000-grain weight (g)	0.226	0.110	-0.454	0.007	-0.259
Biological yield (g)	-0.082	-0.161	-0.345	-0.312	0.574

Harvest index (%)	0.211	0.513	0.175	-0.107	-0.100
Moisture content (%)	-0.101	0.222	0.403	0.180	-0.278
Total ash content (%)	-0.015	0.269	-0.025	0.632	0.450
Crude fibre (%)	0.307	0.021	-0.118	0.210	0.116
Crude protein (%)	0.312	-0.213	0.019	0.029	-0.067
Crude fat (%)	0.335	-0.101	-0.032	0.137	-0.064
Wet gluten content (%)	0.313	-0.156	0.013	0.133	0.103
Gluten strength (ml)	0.317	0.052	-0.087	0.306	0.086
Grain yield/plant (g)	0.202	0.486	0.037	-0.257	0.126
Eigenvalue	7.381	1.950	1.500	1.356	1.150
Variability (%)	43.415	11.47	8.824	7.979	6.765
Cumulative %	43.415	54.885	63.709	71.688	78.453

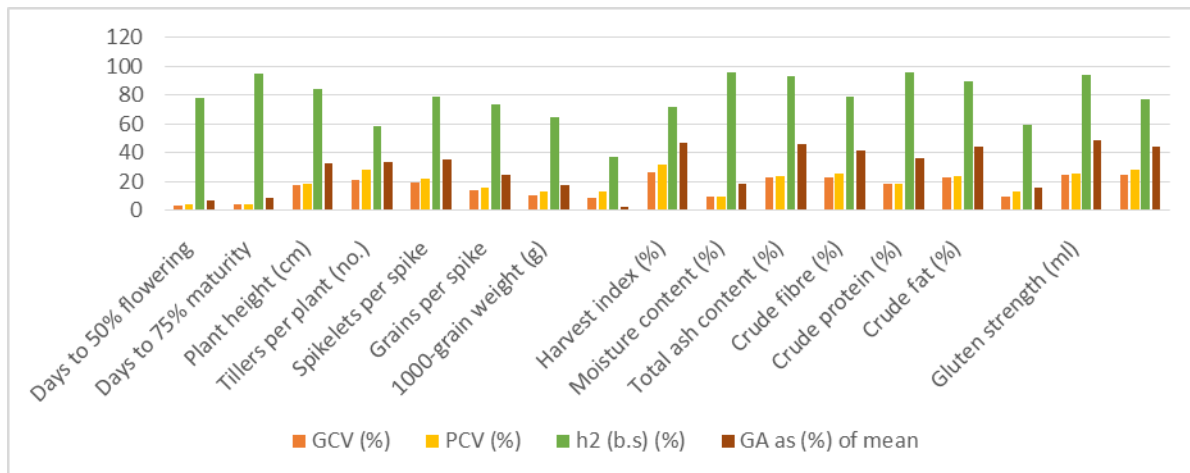


Fig. 1 : Estimates of parameters of variability for various traits in wheat genotypes

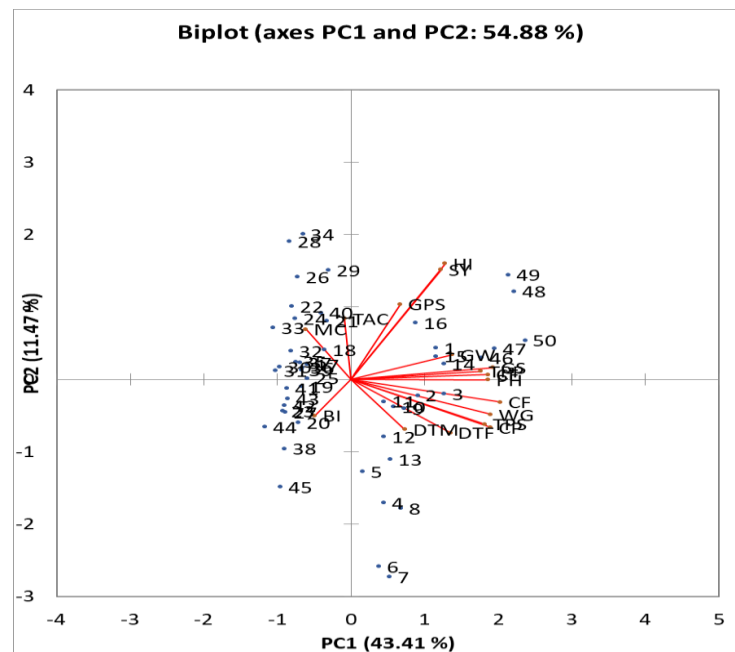


Fig. 2 : Biplot of different variables and genotypes on PC1 and PC2

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