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TRAIT-BASED GENETIC VARIABILITY ANALYSIS IN BRASSICA NAPUS FOR YIELD AND QUALITY IMPROVEMENT

Original Article

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ABSTRACT

Background: Brassica napus represents a major oilseed crop globally and locally, belonging to the economically significant family Brassicaceae. As the world's third most important source of edible oil after soybean and cotton, rapeseed and mustard play a critical role in supplementing domestic oil requirements. Understanding the extent of genetic variability within available germplasm is essential for strengthening breeding pipelines, improving productivity, and enhancing oil quality traits needed to meet growing nutritional and industrial demands.

Objective: To assess the genetic variability among Brassica napus genotypes based on morphological and quality-related traits for effective selection and future breeding application.

Methods: Ten genotypes of Brassica napus were evaluated under a randomized complete block design with three replications. Data were recorded for plant height, shoot diameter, days to 50% flowering, days to maturity, main raceme length, silique length, number of seeds per silique, siliques per main branch, thousand-seed weight, seed yield, oil content, protein content, erucic acid, and glucosinolate concentration. Statistical analyses included analysis of variance (ANOVA) to detect significant differences, followed by LSD tests for pairwise mean comparisons. All procedures ensured accurate assessment of trait variability and genotype performance.

Results: Highly significant differences ($p < 0.05$–0.001) were detected across all traits. Main raceme length ranged from 91.4 to 129.8 cm, silique length from 8.14 to 11.78 cm, and seeds per silique from 19 to 27.6, with Dunkled showing the highest values. Seed yield varied between 1252 and 1764 kg/ha. Genotype RBN-63 possessed the lowest erucic acid (0.42%) and glucosinolates (119 µmol/g) along with high protein content (23.5%), whereas RBN-72 exhibited the highest oil content (45%). Coefficients of variation remained low to moderate, confirming stable trait expression.

Conclusion: The study identified substantial genetic variability among the genotypes, enabling meaningful selection for breeding. RBN-63 and RBN-72 were most promising for enhancing oil quality and protein content, while Dunkled showed superiority in yield-contributing traits. These findings offer valuable direction for developing high-yielding, high-quality Brassica napus cultivars.

Keywords: Brassica napus, Genetic Variation, Oilseed Crops, Plant Breeding, Quantitative Traits, Rapeseed Genetics, Trait Improvement.

INTRODUCTION

The family **Brassicaceae** comprises several agriculturally significant genera, among which *Brassica* holds a central position due to its diverse range of oilseed crops and vegetables. Within this genus, rapeseed (*Brassica napus*, *Brassica carinata*) and mustard (*Brassica juncea*, *Brassica campestris*) represent two major cultivated groups and collectively serve as the world's third most crucial source of edible oil after cotton and soybean because of their adaptability and extensive production areas (1). Mustard, although primarily self-pollinating, exhibits a considerable proportion of natural cross-pollination under field conditions and provides seed containing nearly 42% oil alongside edible greens popularly consumed as mustard leaves (2,3). Canola, derived from oilseed rape cultivars containing <2% erucic acid and <30 µmol/g glucosinolates, has gained global importance due to its favourable fatty acid profile consisting predominantly of monounsaturated fats, moderate polyunsaturated fats, minimal saturated fats, and trace erucic acid (4,5). The improved oxidative stability and shelf life of these oils further enhance their culinary and industrial relevance. In Pakistan, *Brassica campestris*, *B. juncea*, *Eruca sativa*, and *B. napus* have long been cultivated, with *B. napus* (canola type) considered economically superior owing to its desirable omega-3 and omega-6 composition, low glucosinolate levels, low erucic acid content, and amphidiploid genetic constitution derived from *B. rapa* (A genome) and *B. oleracea* (C genome) (6). Despite its long tradition of cultivation, Pakistan continues to face a widening gap between edible oil production and consumption. The country's edible oil import bill is second only to petroleum, with rapeseed and mustard occupying 222 thousand hectares and producing 488 thousand tonnes in 2020–21 (7). During FY-2023, 2.681 million tonnes of edible oil—valued at 826.482 billion PKR—were imported, whereas only 0.496 million tonnes were produced locally, reflecting a continued reliance on imports despite favourable domestic potential (Pakistan Bureau of Statistics). Enhancing national self-sufficiency requires expanding cultivation under diverse agro-ecological conditions and developing genetically improved Brassica cultivars with superior agronomic and oil quality attributes (8).

Genetic variability underpins successful crop improvement, as it determines the extent to which populations can adapt, evolve, and respond to selection pressures (9). Traits exhibiting high genetic variation, broad-sense heritability, and substantial genetic advance are particularly valuable for selection, especially when targeting complex quantitative traits such as seed yield (10,11). Parameters including genotypic and phenotypic coefficients of variation (GCV, PCV), heritability estimates, and expected genetic advance aid breeders in identifying traits with the greatest potential for enhancement (12,13). Morphological characteristics such as plant height, number of siliques per plant, and seed number have historically served as reliable indicators for selecting superior genotypes. However, modern breeding priorities emphasize the simultaneous improvement of oil quality—specifically higher monounsaturated fats, reduced polyunsaturated fractions, and minimal saturated fatty acids—to meet evolving nutritional and industrial standards (14). Thus, developing Brassica cultivars that combine high yield potential with premium oil quality remains a pivotal goal. Given Pakistan's increasing edible oil demand and pressing need for self-reliance, the present study aims to evaluate genetic variability, heritability, and trait associations in Brassica genotypes to identify key parameters that can guide effective selection strategies and support the development of improved, high-yielding oilseed cultivars suitable for local agro-climatic conditions.

METHODS

The experiment was conducted during the spring season of 2023–2024 under field conditions at the Department of Plant Breeding and Genetics, University of Agriculture, Faisalabad, following a randomized complete block design (RCBD) with three replications. Ten *Brassica* genotypes—Rachna Canola, Sandal Canola, NIFA-Gold, Durr-e-NIFA, Dunkled, Abassine-95, RBN-63, RBN-61, Super Canola and RBN-72—were evaluated for morphological and quality-related traits. These genotypes were considered the study's treatments to assess genetic variability. The land was prepared using conventional tillage, after which sowing was performed manually using a hand drill. Di-ammonium phosphate (50 kg) was applied as a basal fertilizer, and urea was administered 20–25 days after sowing with the first irrigation. Seeds were sown on ridges with a line-to-line distance of 1.5 feet and plant-to-plant spacing of 4 inches. Although inclusion and exclusion criteria were not explicitly defined, all selected genotypes represented commonly cultivated Brassica materials appropriate for field evaluation. Ethical approval and informed consent were not applicable to this agronomic field study; however, institutional permission for field experimentation was secured as per standard academic protocol. Morphological observations included plant height, shoot diameter, days to 50% flowering, days to maturity, main raceme length, number of siliques on the main branch, number of seeds per silique, silique length, thousand-seed weight, and seed yield. Standard procedures were followed, whereby flowering and maturity days were recorded from sowing until 50% bloom and full maturity, respectively. Siliques on the main branch were counted manually from five randomly selected plants per replication, and their means were computed. Thousand-seed weight was obtained by counting and weighing 1000 seeds for each genotype, while seed yield per replication was calculated using the formula: Seed yield = (A × B × C) / 10000, where A represented the number of siliques per m², B the number of seeds per silique, and C the 100-seed weight. Quality parameters including oil content, protein percentage, erucic acid, and glucosinolate concentration (µmol/g) were quantified using Near-Infrared (NIR) spectroscopy at the Ayub Research Oil Technology Laboratory, Oilseed Research Institute, Faisalabad. Statistical analyses were performed using R software. A fixed-effects ANOVA model under RCBD was applied to determine the significance of differences among genotypes, while random-effects ANOVA was used to compute means, medians, and standard deviations. Least Significant Difference (LSD) tests were conducted for pairwise comparison of mean values. Correlation coefficients were calculated following Kwon and Torrie (1964) to assess associations among traits. Genotypic and phenotypic coefficients of variation (GCV and PCV) were estimated according to Burton and DeVane (1952). Broad-sense heritability was derived using the method of Singh and Ceccarelli (1996), and genetic advance (GA) along with genetic advance as a percentage of the mean (GAM) was computed following Allard (1960). Thresholds for interpreting PCV, GCV, heritability, and GA categories were adopted from established classifications (Burton, 1953; Stansfield, 1986; Allard, 1960).

RESULTS

Analysis of variance revealed highly significant differences ($p < 0.05–0.001$) among the ten Brassica napus genotypes for all measured morphological and quality traits, indicating the presence of substantial genetic variability suitable for effective selection. The fourteen assessed parameters showed considerable ranges in their minimum and maximum values, and coefficients of variation remained generally low to moderate across traits. Plant height ranged from 173 to 199.2 cm with a mean of 186.2 cm and a CV of 1.55%, whereas shoot diameter varied from 4.3 to 6.36 cm with a CV of 5.95%. Main raceme length ranged between 91.4 and 129.8 cm (CV 4.86%), while silique length ranged from 8.14 to 11.78 cm (CV 4.08%). The number of seeds per silique ranged between 19 and 27.6 (CV 7.97%), and the number of siliques on the main branch ranged from 48 to 78.2 (CV 7.17%). Days to 50% flowering ranged between 78 and 92 days (CV 1.58%), whereas days to maturity ranged from 154 to 171 days (CV 0.7%). Thousand-seed weight ranged from 4.44 to 5.44 g (CV 1.99%), and seed yield showed a broad range from 1252 to 1764 kg/ha (CV 4.75%). Quality parameters exhibited narrower CV values, with oil content ranging from 39.41% to 45% (CV 0.93%), protein content from 19.25% to 23.5% (CV 0.57%), erucic acid from 0.42% to 3.44% (CV 1.3%), and glucosinolates from 119 to 178 µmol/g (CV 0.64%).

Morphological Traits

Highly significant differences were observed for all morphological traits. Plant height showed a broad range of 175.6–199 cm, while shoot diameter varied between 4.9 and 5.97 cm. Days to 50% flowering ranged from 79 to 90 days, and days to maturity ranged from 156 to 168 days. Main raceme length ranged from 95.2 to 124.2 cm, and the number of siliques per main branch varied between 56 and 76. The number of seeds per silique ranged from 20 to 26, whereas silique length ranged from 8.7 to 11.1 cm. Thousand-seed weight varied from 4.50 to 5.28 g, and seed yield ranged from 1316 to 1730 kg/ha. All morphological traits displayed significant genetic variability, confirming their reliability for selection in breeding programs.

Quality Traits

Quality-related parameters also exhibited highly significant variation across genotypes. Oil content ranged from 39.6% to 44.8%, protein content from 19.3% to 23.5%, erucic acid from 0.4% to 3.4%, and glucosinolate concentration from 119 to 178 µmol/g. Both erucic acid and glucosinolates exhibited exceptionally high heritability estimates (~99%), while oil and protein contents showed moderate ranges but remained statistically significant, indicating strong genetic influence over these traits.

Correlation and Heat Map Analysis

Correlation analysis showed that several yield-contributing traits—such as plant height, main raceme length, number of siliques per branch, silique length, and thousand-seed weight—were positively associated with seed yield. These traits clustered in the strongly positive (red) correlation regions of the heat map. Negative correlations were observed between oil content and protein content, as well as between oil content and erucic acid, forming distinct blue regions in the heat map. Overall, the correlation pattern indicated that simultaneous improvement in interrelated yield components could effectively enhance seed yield.

Genetic Parameters

The differences between GCV and PCV were narrow for most traits, suggesting minimal environmental influence and high genetic control. The highest GCV and PCV were recorded for erucic acid (49.36% and 49.38%) and glucosinolates (13.92% and 13.93%), indicating high variability. Heritability values ranged from 33.24% for shoot diameter to 99% for erucic acid and glucosinolates. High heritability coupled with high genetic advance as a percentage of mean was noted for seed yield, erucic acid, and glucosinolates, confirming their suitability for improvement through direct selection.

Table 1: Range, Mean and CV (%) of 10 Brassica genotypes for 14 characters

Parameters	Range		Mean	CV (%)
	Min.	Max.		
PH	173	199.2	186.2	1.55
SD	4.3	6.36	5.33	5.95
MRL	91.4	129.8	110.71	4.86
SL	8.14	11.78	9.46	4.08
NOS	19	27.6	23.02	7.97
NSOMB	48	78.2	63.1	7.17
F	78	92	85.66	1.58
MATU	154	171	162.23	0.7
SW	4.44	5.44	5	1.99
SY	1252	1764	1514.86	4.75
OIL	39.41	45	42.14	0.93
PRO	19.25	23.5	20.93	0.57
ERU	0.42	3.44	2.01	1.3
GLU	119	178	139.06	0.64

Min: Minimum value, Max: Maximum value, CV: Coefficient of variation

Table 2: Analysis of Variance (ANOVA) for all traits showing F-calculated values with significance levels indicated as superscripts

Traits/SOV1	Genotype	Replications
Plant height (cm)	19.3595***	0.0237NS
Shoot diameter (cm)	2.6343*	19.3969***
Days to 50% flowering	25.274***	16.558***

Traits/SOV1	**Genotype**	**Replications**
Days to maturity	92.921***	33.833***
Main raceme length (cm)	6.1625***	2.2641NS
No. of siliques on main branch	7.7279***	0.044NS
No. of seeds per silique	3.2442*	4.7624*
Silique length (cm)	10.3075***	2.5488NS
1000 seed weight (g)	17.0355***	0.4521NS
Seed yield (kg/ha)	6.4497***	0.2879NS
Oil content (%)	51.0043***	0.1863NS
Protein content (%)	268.7334***	2.2515NS
Erucic acid (%)	4334.179***	0.6481NS
Glucosinolates (µmol/g)	1406.833***	2.6667NS

Sources of Variation

* Significance at α = 0.05

** Significance at α = 0.01

*** Significance at α = 0.001

NS non-significant

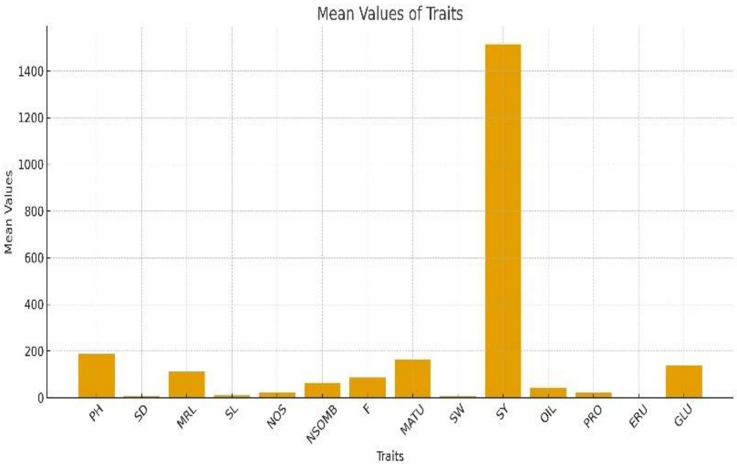


Figure 1Mean values of Traits

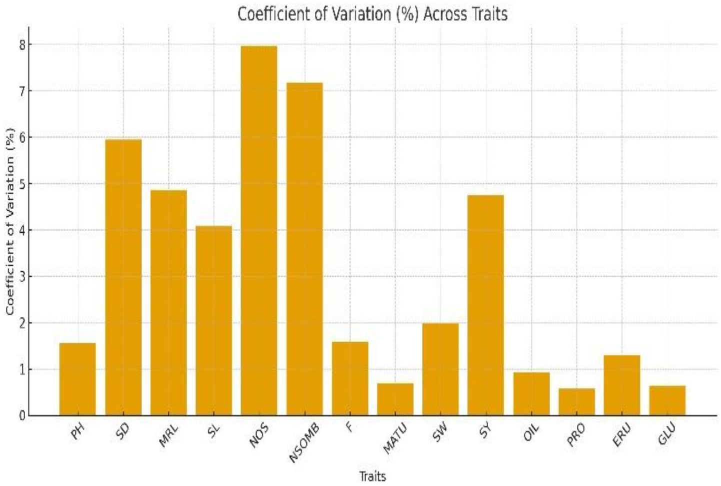
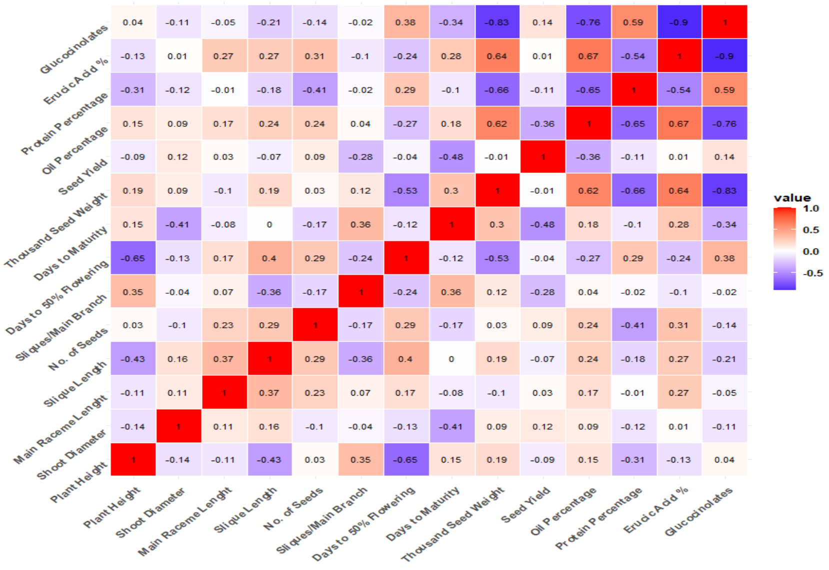


Figure 1 Coefficients of Variation (%) Across Traits



	Plant Height	Shoot Diameter	Main Raceme Lenght	Silique Length	No. of Seeds	Siliques/Main Branch	Days to 50% Flowering	Days to Maturity	Thousand Seed Weight	Seed Yield	Oil Percentage	Protein Percentage	Erucic Acid %	Glucocinolates
Glucocinolates	0.04	-0.11	-0.05	-0.21	-0.14	-0.02	0.38	-0.34	-0.83	0.14	-0.76	0.59	-0.9	1
Erucic Acid %	-0.13	0.01	0.27	0.27	0.31	-0.1	-0.24	0.28	0.64	0.01	0.67	-0.54	1	-0.9
Protein Percentage	-0.31	-0.12	-0.01	-0.18	-0.41	-0.02	0.29	-0.1	-0.66	-0.11	-0.65	1	-0.54	0.59
Oil Percentage	0.15	0.09	0.17	0.24	0.24	0.04	-0.27	0.18	0.62	-0.36	1	-0.65	0.67	-0.76
Seed Yield	-0.09	0.12	0.03	-0.07	0.09	-0.28	-0.04	-0.48	-0.01	1	-0.36	-0.11	0.01	0.14
Thousand Seed Weight	0.19	0.09	-0.1	0.19	0.03	0.12	-0.53	0.3	1	-0.01	0.62	-0.66	0.64	-0.83
Days to Maturity	0.15	-0.41	-0.08	0	-0.17	0.36	-0.12	1	0.3	-0.48	0.18	-0.1	0.28	-0.34
Days to 50% Flowering	-0.65	-0.13	0.17	0.4	0.29	-0.24	1	-0.12	-0.53	-0.04	-0.27	0.29	-0.24	0.38
Siliques/Main Branch	0.35	-0.04	0.07	-0.36	-0.17	1	-0.24	0.36	0.12	-0.28	0.04	-0.02	-0.1	-0.02
No. of Seeds	0.03	-0.1	0.23	0.29	1	-0.17	0.29	-0.17	0.03	0.09	0.24	-0.41	0.31	-0.14
Silique Length	-0.43	0.16	0.37	1	0.29	-0.36	0.4	0	0.19	-0.07	0.24	-0.18	0.27	-0.21
Main Raceme Lenght	-0.11	0.11	1	0.37	0.23	0.07	0.17	-0.08	-0.1	0.03	0.17	-0.01	0.27	-0.05
Shoot Diameter	-0.14	1	0.11	0.16	-0.1	-0.04	-0.13	-0.41	0.09	0.12	0.09	-0.12	0.01	-0.11
Plant Height	1	-0.14	-0.11	-0.43	0.03	0.35	-0.65	0.15	0.19	-0.09	0.15	-0.31	-0.13	0.04

Heat Map for Correlation Among All the Traits

Figure 2 Heat Map for Correlation Among All the Traits

DISCUSSION

The results of this study demonstrated substantial and consistent variability among *Brassica napus* genotypes for all evaluated morphological and quality traits, reinforcing the premise that the available germplasm possesses adequate genetic diversity for meaningful selection. The presence of highly significant genotypic differences across traits such as plant height, siliques per main branch, seed weight, oil content, and erucic acid supports earlier reports in which broad intra-specific variability was considered central to improving both seed yield and oil quality in rapeseed breeding programs (15). The observed ranges in plant height, raceme length and siliques per branch agree with findings from previous agronomic evaluations, where these traits were repeatedly identified as strong contributors to yield potential under variable environmental conditions. The high heritability estimates for flowering time, maturity duration and raceme characteristics further validate their genetic stability and suitability for direct selection, consistent with earlier studies indicating that such phenological traits are often governed by additive gene action and are less influenced by environmental fluctuations (16,17). The performance variation recorded among genotypes for quality parameters highlights the genetic distinction within the breeding material. The low erucic acid values, ranging between 0.4% and 3.4%, and glucosinolate concentrations between 119 and 178 µmol/g, align with the recognized global breeding objective of developing healthier, canola-grade cultivars. The extremely high heritability estimates for erucic acid and glucosinolates reflect strong genetic control and minimal environmental influence, enabling efficient progress through selection. Such values mirror the findings of previous biochemical screening studies where oil quality traits were consistently shown to be genetically stable and heritable (18,19). Genotypes exhibiting desirable oil content and protein percentage strengthen the argument for their inclusion in advanced breeding cycles targeting improved nutritional profiles.

The correlation pattern revealed that yield-enhancing traits such as raceme length, siliques per branch, silique length, and thousand-seed weight were positively associated with seed yield, supporting the consensus that yield is a polygenic trait resulting from the cumulative influence of multiple components. The negative correlation observed between oil content and protein percentage, as well as between oil content and erucic acid, reinforces widely reported inverse genetic relationships among quality traits. These relationships imply that simultaneous enhancement of multiple quality parameters may require careful recombination strategies to mitigate trade-offs inherent in the biochemical pathways governing seed composition (20-22). The heat map interpretations further confirmed that genotypes possessing robust structural traits tended to show superior yield potential, creating a logical foundation for selecting multiple, synergistic traits rather than relying on single-trait improvement. The genetic parameters calculated in this study further strengthen the interpretability of the results. The small differences between GCV and PCV for most traits indicated modest environmental influence, demonstrating that phenotypic expression reliably reflected underlying genotype. Traits such as erucic acid, glucosinolates and seed yield exhibited high genetic advance alongside high heritability, suggesting predominance of additive gene action and strong opportunities for improvement via direct phenotypic selection. This pattern aligns with quantitative genetic principles derived from classical breeding studies, where high heritability coupled with high genetic advance has been consistently associated with effective selection response (23,24).

The study possesses notable strengths, including the use of a well-controlled RCBD experimental design, the evaluation of both morphological and biochemical traits, and the integration of multiple genetic parameters for robust interpretation. The inclusion of ten diverse genotypes further ensured adequate representation of variability. However, certain limitations influence the broader applicability of the findings. The study was conducted at a single location and within a single season, restricting the assessment of genotype × environment interactions that are known to influence yield and oil quality traits in Brassica. The absence of multi-environment testing restricts the extrapolation of stability and adaptability information. Moreover, although heritability estimates were comprehensive, the study did not incorporate molecular marker-based diversity assessment, which could complement phenotypic data and refine selection strategies. Genotype-wise detailed trait means and LSD-based comparisons were not presented, which limits the precision in identifying statistically superior cultivars. Additional analyses such as principal component analysis (PCA) and cluster analysis could further strengthen genotype categorization based on multivariate patterns. Despite these constraints, the findings offer credible evidence that significant genetic variation exists among the evaluated genotypes, supporting their suitability for targeted breeding. The favorable yield attributes observed in genotypes such as Dunkled and the promising quality profiles in RBN-63 and RBN-72 provide a valuable starting point for future hybridization efforts. Subsequent studies should incorporate multi-location trials, evaluation under stress conditions, and integration of genomic tools to validate trait stability and enhance the accuracy of selection (25). The incorporation of gene expression profiling for key oil quality pathways may also deepen understanding of trait inheritance. Overall, the study contributes meaningfully to rapeseed improvement by identifying genetically diverse and agronomically superior candidates for further breeding advancement.

CONCLUSION

The study confirmed substantial genetic variability among the evaluated *Brassica napus* genotypes, demonstrating clear potential for targeted selection and effective breeding. Yield-enhancing traits showed strong positive associations, indicating that improving one component can strengthen overall productivity. Genotypes displaying superior structural attributes and desirable oil and protein profiles emerged as valuable candidates for future hybridization and cultivar development. These findings reinforce the importance of integrating both agronomic and quality traits in rapeseed breeding and provide a strong foundation for advancing high-yielding, nutritionally improved varieties suited to evolving agricultural and market needs.

AUTHOR CONTRIBUTION

Author	**Contribution**
Menahil Riaz	Substantial Contribution to study design, analysis, acquisition of Data Manuscript Writing Has given Final Approval of the version to be published
Muhammad Azam Khan*	Substantial Contribution to study design, acquisition and interpretation of Data Critical Review and Manuscript Writing Has given Final Approval of the version to be published
Umara Sahar Rana	Substantial Contribution to acquisition and interpretation of Data Has given Final Approval of the version to be published
Muhammad Ahsan Khan	Contributed to Data Collection and Analysis

Author	Contribution
	Has given Final Approval of the version to be published
Huma Saleem	Contributed to Data Collection and Analysis Has given Final Approval of the version to be published
Mubashar Nadeem	Substantial Contribution to study design and Data Analysis Has given Final Approval of the version to be published
Muhammad Ibrahim	Contributed to study concept and Data collection Has given Final Approval of the version to be published
Muhammad Qasim	Writing - Review & Editing, Assistance with Data Curation
Shadab Shaukat	Writing - Review & Editing, Assistance with Data Curation
Kashif Rashid	Writing - Review & Editing, Assistance with Data Curation

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