

DEVELOPMENT OF *BRASSICA NAPUS* L. LINES FOR HIGH YIELD AND ESSENTIAL FATTY ACIDS

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Abstract Heterosis and combining ability were calculated for 13 different morphological, yield, and quality-related traits of 18 F1 *Brassica napus* hybrids developed through line × tester mating design using 6 lines and 3 testers along with their parents. Line × tester ANOVA revealed highly significant results among all characters except for primary branches per plant. ZM-R-2 (tester) was identified as a good general combiner for days to 50% flowering, days to 50% siliqua formation, linolenic acid (%), number of secondary branches, siliquae per plant, and seeds per siliqua, while ZM-R-6 for 100-seed weight, protein, and oleic acid percentage. ZM-R-11 × ZM-R-2 was identified as a good specific combiner for days to flowering, 50% flowering, and 50% siliqua, while ZM-M-6 × ZM-R-2, ZN-M-9 × ZM-R-6, ZN-R-8 × ZM-R-6, Shiralee × ZM-R-2 were good specific combiners for seeds per siliqua, 100-seed weight, oil, and protein percentage, respectively. Highly significant and desired mid-parent heterosis was observed for ZM-R-7 × ZM-R-2 for different morphological and quality traits like linolenic acid content and Shiralee × ZM-R-2 for oil and protein content. Positive mid parental heterosis for oleic acid, linoleic acid, and seed yield plant-1 were observed in ZM-R-7 × ZM-R-10 and ZN-M-9 × ZM-R-10. These hybrids can be utilized as genetic material for future breeding programs.

Keywords: Line × tester; mid parental heterosis; combining ability; quality traits; industrial development

Introduction

Edible oil is an important food commodity worldwide and a source of essential fatty acids. For the past few decades, Pakistan has been importing a huge amount of edible oil at the cost of precious foreign exchange. Only 20-25% of needs are met through domestic production, and the remaining 75% are satisfied through imports (Shehzad, Sadaqat, *et al.* 2015). Increasing human population day by day cause major increase in oil consumption, so there is a dire need to enhance edible oil production at a national level to meet the growing needs. In 2021 (July-March), 2.91 million tons of edible oil valued at 574.19 billion (US\$ 3.41 billion) was imported, while local production was recorded to be 0.37 million tons (Government of Pakistan 2021). There is a huge gap between the production and consumption of edible oil in Pakistan which is expected to go higher in the future until necessary action is taken to overcome this shortfall.

There is about a 3-4% increase in the import bill of edible oil each year (Khatri *et al.* 2005). This exhibits

the gap between production and consumption, which must be fulfilled by producing advanced local cultivars and lines with higher yield and quality traits. Rapeseed-mustard is an important group of oilseed crops belonging to the *Brassicaceae* family and has a vital position in world agriculture (Mayurakshee Mahanta and Purna Kanta Barua 2020). *Brassica* crops have gained high dietary importance as vegetables, condiments, and edible oil. They are cultivated in over 53 world countries and are considered one of the oldest and earliest crop species domesticated by men (Kumar *et al.* 2014). *Brassica* oil has genetic variability in its fatty acid composition. Pakistan produces a reasonable amount of rapeseed-mustard oil. Still, the indigenous cultivar's oil cannot be used in manufacturing vegetable ghee and oil due to their high erucic acid and glucosinolates content (Ahmad *et al.* 2013). That's why breeding efforts are made to develop genetic stock with improved seed yield and oil quality compared to local cultivars. This goal can be achieved by utilizing the full genetic

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potential of the respective crop, as the success of any breeding program depends on the availability of diverse genetic material with novel characteristics (Gul *et al.* 2018). Oil quality depends upon both nutritional and functional aspects, which are associated with the profile of the fatty acids present in it. Breeding efforts are being made to reduce anti-nutritive components and improve meal quality (Alrosan *et al.* 2022). The main objective of oilseed breeders is to develop *Brassica* cultivars with high seed oil content and superior fatty acid profile.

For the past few years, oil yield and its quality have become an important field of study and research worldwide (Ahmad *et al.* 2022). The oil content with desired fatty acid composition in oilseed *Brassic*as is a chief selection character to plant breeders. This study was carried out to develop the *Brassica napus* genotypes for high seed yield and better oil quality parameters, with more than 40% monounsaturated fatty acids (oleic acid) and 6-11% polyunsaturated fatty acids (Phuah *et al.* 2022). An ideal edible oil needs to meet our daily nutritional demands without causing any harm to human as well as animal health.

Materials and Methods

Genetic Material

The experimental material was obtained from the Department of Plant Breeding and Genetics, University of Agriculture, Faisalabad, Pakistan. It consisted of 9 *Brassica napus* genotypes viz. ZM-R-7, ZN-M-9, ZM-M-6, ZN-R-8, Shiralee, ZN-M-11, ZM-R-6, ZM-R-2, and ZM-R-10. Among these nine genotypes, 6 were used as lines (ZM-R-7, ZN-M-9, ZM-M-6, ZN-R-8, Shiralee, and ZN-M-11) 3 lines as testers (ZM-R-6, ZM-R-2, and ZM-R-10) for line × tester mating design. These nine *Brassica napus* genotypes were crossed in line × tester mating design through controlled pollination. Their seeds were harvested and stored to grow in the next season for evaluation and traits analysis.

Stored F₀ seeds were sown in the research area of the Department of Plant Breeding and Genetics, the University of Agriculture, Faisalabad, and their parents in a Randomized Complete Block Design (RCBD) with three replications. Data on morphological, phenological, seed yield, and quality parameters were collected.

Observed Traits

Data regarding 13 different yield and quality-related parameters were collected, which include plant Height (PH), primary and secondary branches (PB and SB), days to flowering (DF), days to 50% flowering (DF50%), days to 50% siliqua (DS50%), siliqua plant⁻¹ (SPP), seeds per siliqua (SPS), 100 seed weight (HSW), yield plant⁻¹ (YPP), oil content % (OC), protein content % (PC), and fatty acid profile which include major unsaturated and essential fatty acids like oleic acid content % (OA), linoleic acid content % and linolenic acid content % were

measured using multipurpose analyzer (MPA) by using a random sample of 3-4 g seeds of *Brassica napus* through MPA (Multi-Purpose FT-NIR Analyser) it allows the analysis of liquids, solids powders, and tablets in transmittance as well as diffuse reflectance. The MPA spectrometer is mainly operated using a data system (PC, notebook, etc.) and setting the measurement parameters using the spectroscopy software OPUS.

Biometrical Approaches

Analysis of Variance

All the collected data were subjected to an analysis of variance (ANOVA) given by (Steel *et al.* 1997) to determine the variability present within the genotypes Table 1.

Table 1. Mean square values from analysis of variance for various yield and related traits.

Sr. number	Name of characters	MSS
1	Days to flowering	207.32**
2	Days to 50 % flowering	242.04**
3	Days to 50% siliquae formation	230.05**
4	Number of primary branches	1.46
5	Number of secondary branches	7.63**
6	Plant height	349.49**
7	Number of siliquae per plant	33554.69**
8	Number of seeds per siliqua	11.44*
9	Seed yield per plant	9.28*
10	100-seed weight	0.0039**
11	Oil %	37.48**
12	Protein %	32.73**
13	Oleic acid %	105.74**
14	Linolenic acid %	10.57**
15	Linoleic acid %	13.65**

* = Significant at 0.05 probability level; ** = Significant at 0.01 probability level; DF = Degree of freedom, MSS = Mean sum of square, Genotypes Df = 26, Error Df = 52, Replication Df = 2

Results and Discussion

Analysis of variance presented highly significant results for all studied traits except for the primary branches, which indicated non-significant differences among all the genotypes. (Nasim *et al.* 2014) also reported non-significant results for primary branches per plant. ANOVA for line × tester revealed highly significant results for all the portioned variables except for primary branches per plant in which only female parents exhibited significant value. Nasim *et*

al. (2013), Shrimali et al. (2018), and Rashmi et al. (2018) also reported non-significant results for the number of primary branches per plant (Table S1). GCA and SCA results of different phenological, morphological, and biochemical parameters are highlighted and mentioned in Tables S2, and S3.

Combining Ability Effects

Combining ability is an important statistical procedure through which parents and their desired combinations are selected for desired traits and their manipulation in future breeding programs to get hybrids and varieties with better economic values. The GCA and SCA effects help determine the role of additive and non-additive gene actions in the expression and inheritance of a particular trait and formulating a breeding procedure accordingly.

Phenological Parameters

Early flowering is a needed character that indicates early siliqua formation and, ultimately, maturity. Fewer days to flowering are required for early crop establishment (Huang and Laosuwan 2010). In this experiment, ZM-M-6 exhibited highly significant GCA effects for days to flower initiation in a negative direction (-2.04**) similarly among testers highly significant GCA in a negative direction was found in ZM-R-2 (-4.04**). For SCA effects crosses ZN-M-11 × ZM-R-2 (-5.19**), ZN-M-9 × ZM-R-10 (-5.19**), Shiralee × ZM-R-2 (-4.96**) and ZN-R-8 × ZM-R-6 (-4.19**) exhibited highly significant results for days to flowering in a negative direction. Combining ability effects for this trait was also studied by (Huang and Laosuwan 2010, Gami and Chauhan 2013, Adhikari et al. 2017, Dahiya, Tufchi, et al. 2018), and their results were following our findings. Meanwhile, days to 50% flowering are one of the desirable phenological traits which highlight early seed setting. The early flowering genotypes ultimately have more grain filling duration, and the seed will be bold as well. Among lines ZN-R-8 (-2.46**) sowed a highly significant GCA effect in a negative direction, followed by ZM-M-6 and ZM-R-7 (-1.91**), and the only male parent ZM-R-2 (-3.52**) showed highly significant GCA effects for days to 50% flowering in a negative direction. Among crosses, four hybrids ZN-M-11 × ZM-R-2 (-4.70**), ZN-R-8 × ZM-R-6 (-3.93**), ZM-M-6 × ZM-R-10 (-3.48**) and ZN-M-11 × ZM-R-6 (-3.48**) exhibited highly significant SCA effects in a negative direction indicating fewer days to 50% flowering. (Gupta et al. 2010, Gideon J. Synrem et al. 2015, Barupal 2016, Chaurasiya et al. 2018), (Gul et al. 2018) also studied the effects of GCA and SCA on this character. Similarly, fewer days to 50% siliqua formation lead to early maturing and protects the crop from later stages of insect attack and losses. Among lines ZN-R-8 (-2.74**) had a highly significant GCA effect for days to 50% siliqua formation in the negative direction, followed by ZM-R-7 (-2.63**). Among testers (male parents) ZM-R-2

(-3.07**) sowed highly significant GCA in a negative direction. Highly significant SCA in a negative direction was noticed in ZN-M-11 × ZM-R-2 (-5.37**), ZM-M-6 × ZM-R-10 (-3.93**), and ZN-R-8 × ZM-R-6 (-3.48**). This is the character only studied in this experiment. These parental lines can be used in the breeding programs to transmit this character.

Morphological Parameters

There is a positive relationship between the number of primary branches and seed yield, while it has a non-significant relationship with 1000-seed weight During this study, only female parent ZM-R-7 (0.91**) had a highly significant GCA effect. No, hybrid was found with significant results for this character, and line ZM-M-6 (-0.76*) exhibited significant GCA in a negative direction. The rest of the female lines and male tester exhibited non-significant results for GCA effects. Non-significant SCA effects were observed for this trait among all hybrids. (Dahiya et al., 2018a; Khan and Hassan 2019; Shah et al. 2021; Ahmad et al. 2022) also studied the general and specific combining ability effects for primary branches per plant. There is a highly significant and positive relationship between the number of secondary branches with oleic acid and linolenic acid proportion and siliquae per plant ultimately increasing yield. Results exhibited that female parent ZM-R-7 (1.28**) was found highly significant for the GCA effect in a positive direction. Among testers ZM-R-2 (1.44**) showed highly significant GCA in a positive direction. Hybrid ZM-R-7 × ZM-R-2 (2.67**) was found with highly significant GCA effects, while three hybrids ZN-M-11 × ZM-R10 (1.50*), ZN-M-11 × ZM-R-2 (1.39*) and Shiralee × ZM-R-6 (1.39*) exhibited significant SCA for several secondary branches per plant. The results are following the findings of (Ahsan et al. 2013, Singh et al. 2016, Chaurasiya et al. 2018, Khan and Hassan 2019).

Yield Parameters

Siliquae per plant has a positive impact on yield, more the number of siliquae will be the yield of the respective genotype. Therefore, a positive and significant GCA effect is required for this trait. Female parents ZM-R-7 (90.94**) along with ZM-M-6 (42.39**) and ZN-M-9 (38.83**) have significant positive values. Among testers ZM-R-2 (41.56**) exhibited highly significant GCA, followed by ZM-R-10 (36.33**). Hybrids with highly significant SCA were noticed in ZM-M-6 × ZM-R-10 (124.78**), ZN-M-9 × ZM-R-6 (114.89**) and ZN-R-8 × ZM-R-2 (111.33**), in a positive direction. The results were following the findings of (Gami and Chauhan 2013, Gautam and Chauhan 2016, Shah et al. 2021). Moreover, plant height is an important character related to *Brassica* species. It has a positive relation with siliquae per plant and is an important factor for increasing plant yield but yield losses have been observed due to logging. Therefore, a medium plant

height is required to minimize losses and robust plant stand against abiotic factors to maximize yield. Significant negative GCA was observed in female parents Shiralee (-10.89**) and ZN-M-11 (-8.05**) can be used as good general combiners for reduced plant height. Only ZM-R-10 (1.85**) among testers indicated highly significant GCA for this trait while the rest of the male parents expressed non-significant results. In the case of specific combining ability ZM-R-7 × ZM-R-6, ZN-M-11 × ZM-R-2 and ZN-R-8 × ZM-R-10 exhibited highly significant SCA effects in a negative direction (-9.35**, -4.97** and -3.47**) respectively. The remaining hybrids showed non-significant results. (Mohammed 2011, Synrem *et al.* 2014, Rameeh 2020) found similar results. Seeds per siliqua has a direct effect on plant yield a greater number of seeds in siliqua will ensure more yield it also has a positive relation with the number of secondary branches and primary branches per plant (Tariq *et al.*, 2020). Among female parents, only ZM-M-6 exhibited highly significant GCA effects in a positive direction (1.70**) and only 1 tester (ZM-R-2) positively exhibited significant GCA effect and indicated a good general combiner for seeds per siliqua. Regarding SCA effects (Table S3, S4), only 2 crosses ZN-M-9 × ZM-R-6 and ZM-M-6 × ZM-R-2 exhibited highly significant SCA effects (2.30**) among 18 crosses. (V. Rameeh 2003, Kumar *et al.* 2014, Muhammad *et al.* 2014, Synrem *et al.* 2015, Mahanta and Barua 2020) also investigated the SCA and GCA effects for this trait and found familiar results. A positive GCA effect in parental lines for 100-seed weight is desirable. ZN-M-9 line showed highly significant GCA in a positive direction (0.03**) for 100-seed weight. Among testers ZM-R-6 (0.02**) demonstrated highly significant results for GCA in positive. Crosses ZN-M-9 × ZM-R-6 (0.05**) showed a highly significant SCA effect in a positive direction. 3 crosses ZM-M-6 × ZM-R-2 (0.03*), ZN-R-8 × ZM-R-6 (0.03*), and Shiralee × ZM-R-2 (0.03*) exhibited significant positive SCA. Non-significant results were noticed in the rest of the crosses. (Gupta *et al.* 2010, Nasim *et al.* 2014, Ullah *et al.* 2015) reported positive GCA and SCA effects for 100-seed weight. Seed yield per plant is an ultimate objective in any breeding program. The only female parent Shiralee (1.07*) possesses a significant GCA effect in a positive direction while non-significant results were noticed among all the testers for yield per plant indicating that multiple genes control seed yield. Results indicated non-significant SCA effects in all hybrids except ZN-M-9 × ZM-R-2 (-1.87*) and ZN-R-8 × ZM-R-6 (-2.14*) which expressed significant SCA effects in a negative direction. The result of this trait is in conformity with the findings of the following authors. (Gul *et al.* 2018, Bharti *et al.* 2018, Dahiya *et al.* 2018, Saikia *et al.* 2019).

Quality Parameters

Oil content is a trait governed by many genes. High oil content is a required parameter the combining ability effects indicate the parental lines have significant values and can be used to improve oil quality and quantity. In Table S2 highly significant positive GCA effects were observed in two females Shiralee (3.42**) and ZM-R-7 (1.63**). Among testers, non-significant results were noticed. In the case of SCA effects Table, S3 showed that only two crosses ZN-R-8 × ZM-R-6 (4.09**), Shiralee × ZM-R-2 (2.57**) with highly significant positive SCA effects while ZM-R-7 × ZM-R-10 (2.45*), Shiralee × ZM-R-10 (2.11*) showed significant SCA effects and rest of the hybrids expressed non-significant SCA results. The result obtained through this experiment was strengthened by the findings of (Azizinia 2012, Meena *et al.* 2015, Mohan *et al.* 2017, Ahmad *et al.* 2022). Protein is also an essential biochemical for all living things and contributes to the growth and development of humans and animals. Results showed a highly significant difference in protein content among the following genotypes. Two lines ZN-R-8 (0.98**) and ZM-M-9 (0.86**) expressed highly significant GCA effects in a positive direction. Two out of three testers exhibited highly significant GCA effects in the positive direction of ZM-R-6 (0.68**) and ZM-R-2 (0.53**). On the other hand, 8 out of 18 crosses exhibited highly significant SCA effects for protein percentage in which two crosses Shiralee × ZM-R-2 (6.73**) and ZN-R-8 × ZM-R-2 (5.13**) showed the highest SCA effects in a positive direction. The results of this experiment were in line with previous findings (Cuthbert *et al.* 2011, Akabari *et al.* 2017, Dahiya *et al.* 2018). Among essential fatty acids, oleic acid is also required in high proportion in edible oils. Genotypes with high oleic acid content are suitable to use in developing hybrids if the parents exhibit high additive gene action. Highly significant positive GCA effects were observed in two lines ZM-M-6 (3.66**) and ZN-M-9 (0.86**), respectively. Testers ZM-R-6 (2.03**) and ZM-R-2 (1.58**) exhibited highly significant GCA effects. Highly significant SCA was expressed in 6 out of 18 crosses and the highest value in the positive direction was observed in ZM-R-7 × ZM-R-10 (10.72**) followed by Shiralee × ZM-R-6 (6.35**) and ZN-R-8 × ZM-R-6 (5.27**) only one hybrid showed significant SCA ZN-M-11 × ZM-R-6 (0.91*). (Turi *et al.* 2010, Shehzad *et al.* 2015, Sohail 2018) also reported results with partial and complete agreement. In contrast, linolenic acid is a polyunsaturated fatty acid. But its low content is desirable in edible oil because its high content disturbs oil stability while storage and frying. Female parent ZM-R-7 (1.21**) exhibited a highly significant positive GCA effect. Two male parents depicted highly significant GCA effects ZM-R-2 (0.82**) and ZM-R-6 (0.25**). Regarding specific combining ability effects, 8 crosses exhibited highly

significant positive SCA. The highest values were observed in ZN-M-11 \times ZM-R-10 (2.93**) followed by ZM-M-6 \times ZM-R-6 (2.89**) and Shiralee \times ZM-R-2 (2.28**). Significant positive GCA and SCA effects for linolenic acid content were studied by (Vaghela *et al.* 2011, Nasim and Farhatullah 2013, Ahmad *et al.* 2022). Whereas, higher linoleic acid contents are required in rapeseed to increase its usefulness as raw material for margarine manufacturing. The ratio between linolenic acid (18:3) and linoleic acid (18:2) which determined the nutrition value of rapeseed oil is (1:2). Female parents were noticed with highly significant GCA effects which included ZN-M-9 (1.29**), ZM-R-7 (1.05**) and ZM-M-6 (0.48**). Among testers highly significant GCA was observed in male parent ZM-R-10 (0.48**). Highest SCA value in positive direction were observed in three parental combinations ZM-M-6 \times ZM-R-6 (3.11**), Shiralee \times ZM-R-10 (2.31**) and ZN-M-9 \times ZM-R-2 (2.24**). Significantly positive combining ability effects related to linoleic acid percentage was also studied by (Shrimali *et al.* 2018, Sohail 2018, Ahmad *et al.* 2022).

Mid-Parent and Better Parent Heterosis

Heterosis for mid and better parent was calculated for all the traits under study and its results are given in Table S4.

Phenological Traits

Heterosis and heterobeltiosis play an important role in the hybrid breeding program. Negative mid-parent heterosis for days to flowering ranged from -19.59** to -6.36** and the highest BP heterosis was found from -26.40** to -3.58**. ZN-M-9 \times ZM-R-10 expressed the highest, negative mid-parent heterosis (-19.59**) for days to flowering and ZM-M-6 \times ZM-R-2 cross showed the lowest negative mid-parent heterosis (-6.36**). Negative heterosis means fewer days taken to flower initiation. Whereas, all crosses showed negative better parent heterosis, which could be exploited for the development of early maturing varieties. ZM-R-7 \times ZM-R-2 cross showed the highest negative better parent heterosis (-26.40**) and the Shiralee \times ZM-R-6 hybrid showed the lowest negative better parent heterosis (-3.58**). (Bharti *et al.* n.d., Dar *et al.* 2011, Muhammad *et al.* 2014, Ali *et al.* 2015)) also found similar results.

For days to 50% flowering ZM-R-7 \times ZM-R-10 hybrid exhibited negative mid-parent heterosis (-19.26**) and ZN-M-9 \times ZM-R-6 (-2.07**) exhibited the lowest negative value. ZM-R-7 \times ZM-R-2 exhibited the highest negative better parent heterosis (-26.40**) and Shiralee \times ZM-R-6 c showed the lowest negative better parent heterosis of (-3.58**) (Sabaghnia *et al.* 2010, Singh *et al.* 2012, Gautam and Chauhan 2016) results were following our findings. Similarly, fewer days to 50% flowering and days to 50% siliqua formation would lead to early maturity. High negative MP heterosis was observed from -18.96** to -4.31**.

ZM-R-7 \times ZM-R-10 showed the highest negative mid-parent heterosis (-18.96**) and ZM-M-6 \times ZM-R-2 cross showed the lowest negative mid-parent heterosis (-4.31**). Similarly, higher negative BP heterosis was recorded in ZM-R-7 \times ZM-R-2 (-26.74**) and Shiralee \times ZM-R-6 showed the lowest negative value (-5.03**).

Morphological Parameters

Positive heterosis for primary branches is required to get more seed yield. All crosses showed non-significant mid-parent heterosis for the number of primary branches except ZM-M-6 \times ZM-R-6 which showed the highest mid-parent heterosis in a negative direction (-36.00**). For better parent heterosis except ZM-M-6 \times ZM-R-6 exhibited negative better parent heterosis (-46.67**) remaining crosses were non-significant. The results partially agreed with (Ahsan *et al.* 2013, Nasim *et al.* 2014, Saikia *et al.* 2019, Mahanta and Barua 2020). Meanwhile, Positive heterosis is desirable for secondary branches due to its direct effect on seed yield. Hybrid with positive significant MP heterosis for secondary branches per plant was ZM-R-7 \times ZM-R-2 (143.48**) and Shiralee \times ZM-R-6 exhibited the lowest value the results conform with (Gupta *et al.* 2010, Bharti *et al.* 2018, Chaurasiya *et al.* 2018). The highest better parent heterosis was observed in ZM-R-7 \times ZM-R-2 (86.67**) (Lal *et al.* 2018, Mahanta and Barua 2020); results aligned with the current study.

Plant height is a highly important trait, the study revealed the highest mid-parent heterosis in the negative direction from -6.24** to -3.04** while 4 out of 18 hybrids showed highly significant better parent heterosis in which ZM-R-7 \times ZM-R-6 has the highest value of (-12.14**) and remaining 6 hybrids showed positive BP heterosis. Studies by Oghan *et al.* 2009, Azizinia 2012, Meena *et al.* 2015, Channa *et al.* 2018 supported our results.

Yield Parameters

Higher numbers of Siliquae are required for achieving desired yield targets. High heterosis either mid-parent or better parent is required in a positive direction. ZN-M-9 \times ZM-R-6 indicated the highest mid-parent heterosis (72.11**) and ZN-R-8 \times ZM-R-2 with the lowest positive mid-parent heterosis (38.80*). ZN-M-9 \times ZM-R-6 hybrid showed the highest positive better parent heterosis (64.55*) and ZM-M-6 \times ZM-R-10 presented the lowest positive better parent heterosis (37.93*). Significant results over mid and better parent were also studied by (Singh *et al.* 2012) and (Synrem *et al.* 2015) on number of siliqua on main plant branch. (Muhammad Zahir Ahsan *et al.* 2013, Lal *et al.* 2018, Shrimali *et al.* 2018) also found significant heterosis over a mid and superior parent. Meanwhile, for seeds per siliqua, all 9 hybrids showed non-significant mid-parent heterosis except ZM-M-6 \times ZM-R-2, Shiralee \times ZM-R-10, ZN-M-11 \times ZM-R-6, and ZN-M-11 \times ZM-R-10. ZM-M-6 \times ZM-R-2

showed the highest positive mid-parent heterosis and ZN-M-11 × ZM-R-10 showed the lowest mid-parent heterosis. Most crosses expressed non-significant better parent heterosis. ZM-M-6 × ZM-R-2 cross showed the highest positive better parent heterosis among crosses and the remaining significant crosses showed negative better parent heterosis. Similar findings were given by (Dholu *et al.* 2014, Synrem *et al.* 2015, Rashmi *et al.* 2018). Estimating mid-parent heterosis and better parent heterosis for 100-seed weight showed the range of (-2.12 to 22.07) and (-0.00 to 15.91) respectively. ZN-M-9 × ZM-R-6 cross had the highest positive mid-parent heterosis (22.07**) and ZM-M-6 × ZM-R-10 had the lowest positive mid-parent heterosis (5.97*). ZN-M-9 × ZM-R-6 hybrid showed the highest positive better parent heterosis (15.91**) and the ZN-M-11 × ZM-R-10 cross reflected the lowest positive better parent heterosis. (Aher *et al.* 2009, Nasim *et al.* 2014, Ullah *et al.* 2015) also found results similar to our findings. High seed yield is the prime objective of any breeding program. High positive heterosis is required among crosses. ZN-M-9 × ZM-R-10 presented the highest positive mid-parent heterosis (79.81**) and ZM-R-7 × ZM-R-6 cross showed the lowest mid-parent heterosis (26.28*) for this parameter. Estimation of better parent heterosis indicated that ZN-M-9 × ZM-R-10 had a high potential for seed yield per plant as indicated by high positive better parent heterosis (69.14**) among all crosses. ZN-M-11 × ZM-R-10 cross presented the lowest better parent heterosis (Ali *et al.*, 2013; Ali *et al.*, 2014; Ali *et al.*, 2016; Vaghela *et al.* 2011, Lal *et al.* 2013, Chaurasiya *et al.* 2018, Dahiya *et al.* 2018, Surin *et al.* 2018) findings were in agreement with our results.

Quality Parameters

Shiralee × ZM-R-10 cross showed the highest positive heterosis for oil content and ZN-R-8 × ZM-R-10 cross showed the lowest heterosis over mid-parent. Shiralee × ZM-R-2 showed the highest heterosis over a better parent and ZM-R-7 × ZM-R-10 exhibited the lowest better parent heterosis. (Wang *et al.* 2009, Iqbal *et al.* 2014, Rashmi *et al.* 2018, Saikia *et al.* 2019) studied significant mid and superior parent heterosis against oil content. Higher seed yield and higher oil content is the prime objective of this study; therefore, crosses with positive heterosis were selected. In the same way, positive high heterosis is required for protein content. ZN-R-8 × ZM-R-2 cross showed the highest mid-parent heterosis (30.48**) for protein content and ZN-M-9 × ZM-R-2 cross showed the lowest mid-parent heterosis (6.78**) for protein content. ZN-R-8 × ZM-R-2 positively showed the highest better parent heterosis, and Shiralee × ZM-R-10 showed the lowest better parent heterosis (-21.37**). (Dholu *et al.* 2014, Ahmad *et al.* 2022) found significant outcomes for protein content over mid and better parent and were in line with our findings, resulting in highest mid and

better parent heterosis in ZN-R-8 × ZM-R-2. Likewise, oleic acid is one of the essential fatty acids in edible oil and its high content is desirable as it is monounsaturated fatty acid. ZM-R-7 × ZM-R-10 cross showed highest positive mid parent heterosis for oleic acid and Shiralee × ZM-R-6 cross showed lowest mid parent heterosis. ZM-R-7 × ZM-R-10 cross showed highest better parent heterosis and ZN-M-11 × ZM-R-2 cross showed lowest better parent heterosis. (Muhammad *et al.* 2014, Gami and Chauhan 2014, Shehzad *et al.* 2015, Ahmad *et al.* 2022) also investigated the significant mid and better parent heterotic effects similar to our results.

Linolenic acid is also required by the human body but in a lower amount therefore, its decreased content in edible oil is preferred. For this purpose, negative heterosis is desired. ZN-R-8 × ZM-R-6 showed the highest mid-parent heterosis and ZN-M-9 × ZM-R-6 with lowest heterotic effect. ZN-R-8 × ZM-R-6 was the hybrid manifesting the highest better parent heterosis and ZN-M-9 × ZM-R-2 with lowest better parent heterosis. The findings of (Vaghela *et al.* 2011, Ali *et al.* 2015, Surin *et al.* 2018, Shrimali *et al.* 2018) were following ours. Linoleic acid is also a polyunsaturated fatty acid its moderate quantity prevents the oil degradation process through rancidity. Among 18 hybrids ZN-M-9 × ZM-R-10 showed the highest mid-parent heterosis (35.39**) and ZM-M-6 × ZM-R-6 with the lowest mid-parent heterosis (7.41**) in a positive direction. ZN-M-9 × ZM-R-10 cross showed the highest better parent heterosis (23.10**) and ZN-R-8 × ZM-R-10 showed the lowest better parent heterosis in a positive direction. (Khan *et al.* 2008, Vaghela *et al.* 2011, Iqbal *et al.* 2014, Shehzad *et al.* 2015) studied significant positive mid and better parent heterosis for linoleic acid.

Conclusions

At the end of the experiment ZM-R-7 × ZM-R-2 was observed as a potential hybrid with highly significant Mid Parent heterosis for days to Flower Initiation, Days to 50% Flowering, and Days to 50% siliqua in a negative direction while positive Mid Parent heterosis for Secondary branches per plant, siliquae per plant and linolenic acid percentage. Shiralee × ZM-R-2 is recommended as a potential hybrid for oil (24.87**) and protein (19.27**) containing highly significant Mid Parent as well Better Parent heterosis. Hybrids ZM-R-7 × ZM-R-10 exhibited potential MP heterosis for oleic acid % (46.81**) and ZN-M-9 × ZM-R-10 for linoleic acid and seed yield per plant (79.81**). This material can be used in future breeding programs to get economical results.

Conflicts of Interest

The authors declare no conflict of interest.

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Development of *Brassica napus* L. Lines for High Yield and Essential Fatty Acids

Table S1. Line × tester ANOVA for various parameters in *Brassica napus* L.

* = Significant at 0.05 probability level

** = Significant at 0.01 probability level

DFI = Days to flower initiation, **D50%F** = Days taken to 50% flowering, **OC** = Oil Percentage, **D50%S** = Days to 50% siliqua formation, **PB** = Number of secondary branches per plant, **PC** = Protein Percentage, **SB** = Number of secondary branches per plant, **S/P** = Number of siliquae per plant, **OA** = Oleic acid Percentage, **PH** = Plant height, **S/S** = Number of seeds per siliqua, **LA** = Linolenic acid Percentage, **Y/P** = Seed yield per plant, **HSW** = 100-seed weight, **LleicA** = Linoleic acid Percentage

SOV	D.F	DFI	DFI	DFI	DFI	DFI	DFI	DFI	DFI	DFI	DFI	DFI	DFI	DFI	DFI	DFI
Rep	2	4.98	2.82	1.35	0.51	0.44	0.90	0.82	5.13**	3.64*	0.98	1.18	0.43	3.37*	0.46	0.1608
Trt	26	90.47**	148.44*	154.73**	1.42	7.48**	93.67**	7.40**	4.54*	4.67*	9.45**	20.23**	57.53**	163.71**	67.81**	800.88**
Parents	8	150.73**	257.11*	251.17**	1.14	2.10	132.92**	4.28*	4.80*	2.43*	6.97**	24.22**	41.22**	167.07**	43.28**	666.78**
Crosses	17	30.31**	44.18**	48.29**	1.64	10.15**	67.94**	8.51**	4.27*	3.55*	9.37**	18.19**	68.02**	150.03**	81.20**	883.20**
P vs C	1	631.02**	1051.39**	1192.86**	0.006	5.08**	217.04**	13.56**	7.15**	41.81**	30.80**	23.11**	9.71**	369.33**	36.65**	474.18**
Lines	5	9.05*	40.21**	54.90**	2.90*	4.38*	184.37**	9.59**	4.14*	3.95*	7.11**	34.58**	10.95**	85.52**	24.90**	684.87**
Testers	2	101.46**	102.80*	85.84**	1.09	27.80**	12.35**	18.10**	6.74**	3.70*	13.71**	1.28	34.92**	274.09**	107.87**	193.82**
L × T	10	26.71**	34.44**	37.47**	1.11	9.50**	20.84**	6.05**	3.84*	3.31*	9.63**	13.37**	103.17**	157.47**	104.01**	1120.24**

* = Significant at 0.05 probability level; ** = Significant at 0.01 probability level; SOV = Source of variation; DF = Degree of freedom

Table S2. Estimation of general combining ability effects of various traits in *Brassica napus* L.

* = Significant at 0.05 probability level

** = Significant at 0.01 probability level

DFI = Days to flower initiation, **D50%F** = Days taken to 50% flowering, **OC** = Oil Percentage, **D50%S** = Days to 50% siliqua formation, **PB** = Number of secondary branches per plant, **PC** = Protein Percentage, **SB** = Number of secondary branches per plant, **S/P** = Number of siliquae per plant, **OA** = Oleic acid Percentage, **PH** = Plant height, **S/S** = Number of seeds per siliqua, **LA** = Linolenic acid Percentage, **Y/P** = Seed yield per plant, **HSW** = 100-seed weight, **LleicA** = Linoleic acid Percentage

Traits	DFI	D50%F	D50%S	PB	SB	S/P	PH	S/S	Y/P	HSW	OC	PC	OA	LA	LleicA
Lines															
ZM-R-7	0.19ns	-1.91**	-2.63**	0.91**	1.28**	90.94**	6.19**	-0.07ns	0.44ns	-0.03**	1.63**	0.19ns	-0.08ns	1.21**	1.05**
ZN-M-9	1.41**	3.65**	3.59**	-0.43ns	-0.28ns	38.83**	12.40**	-1.41**	0.38ns	0.03**	1.01ns	0.81**	0.86**	0.09ns	1.29**
ZM-M-6	-2.04**	-1.91**	-1.85**	-0.76*	-0.17ns	42.39**	2.02**	1.70**	-1.37**	-0.01ns	-4.22**	-0.16ns	3.66**	-0.68**	0.48**
ZN-R-8	-1.59**	-2.46**	-2.74**	0.13ns	-0.17ns	-58.06**	-1.66*	-0.07ns	-0.92ns	0.01ns	-1.52**	0.98**	-3.92**	0.03ns	-0.94**
Shiralee	0.41ns	-0.46ns	-0.19ns	-0.09ns	0.17ns	-20.39ns	-10.89**	0.59ns	1.07*	-0.00ns	3.42**	-0.64**	-0.97**	-0.31**	-0.30**
ZN-M-11	1.63**	3.09**	3.81**	0.24ns	-0.83*	-93.72**	-8.05**	-0.74ns	0.40ns	0.00ns	-0.33ns	-1.17**	0.44ns	-0.35**	-1.59**
Testers															
ZM-R-6	2.85**	1.93**	1.59**	-0.26ns	-0.83**	-77.89**	-0.99ns	-1.07**	0.10ns	0.02**	0.42ns	0.68**	2.03**	0.25**	-0.13**
ZM-R-2	-4.04**	-3.52**	-3.07**	0.24ns	1.44**	41.56**	-0.86ns	0.81*	0.58ns	-0.02**	-0.19ns	0.53**	1.58**	0.82**	-0.35**
ZM-R-10	1.19**	1.59**	1.48**	0.02ns	-0.61*	36.33**	1.85**	0.26ns	-0.68ns	0.00ns	-0.23ns	-1.21**	-3.61**	-1.07**	0.48**

Table S3. Estimation of Specific combing ability effects of various *Brassica napus* L. traits

* = Significant at 0.05 probability level, ** = Significant at 0.01 probability level, **DFI** = Days to flower initiation, **D50%F** = Days taken to 50% flowering, **D50%S** = Days to 50% siliqua formation, **PB** = Number of secondary branches per plant, **SB** = Number of secondary branches per plant, **S/P** = Number of siliquae per plant, **S/S** = Number of seeds per siliqua, **Y/P** = Seed yield per plant, **HSW** = 100-seed weight, **OC** = Oil Percentage, **PC** = Protein Percentage, **OA** = Oleic acid Percentage, **PH** = Plant height, **LA** = Linolenic acid Percentage, **LleicA** = Linoleic acid Percentage

Crosses	DFI	D50%F	D50%S	PB	SB	PH	S/P	S/S	Y/P	HSW	OC	PC	OA	LA	LleicA
ZM-R-7 × ZM-R-6	-0.96ns	2.85**	2.41**	-0.52ns	-0.72ns	-9.35**	30.11ns	0.96ns	0.03ns	0.02ns	-1.06ns	2.34**	-7.80**	-1.16**	-1.32**
ZM-R-7 × ZM-R-2	0.93ns	-1.37ns	-0.93ns	0.65ns	2.67**	3.33**	27.00ns	-0.59ns	0.35ns	-0.04**	-1.39ns	-4.65**	-2.92**	1.01**	1.33**
ZM-R-7 × ZM-R-10	0.04ns	-1.48ns	-1.48*	-0.13ns	-1.94**	6.02**	-57.11*	-0.37ns	-0.39ns	0.02ns	2.45*	2.31**	10.72**	0.15ns	-0.01ns

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ZN-M-9 × ZM-R-6	-0.19ns	0.63ns	-0.81ns	0.48ns	-0.17ns	2.19ns	114.89**	2.30**	0.81ns	0.05**	0.84ns	3.33**	-1.78**	-	-2.93**
ZN-M-9 × ZM-R-2	5.37**	1.74*	1.19ns	-0.69ns	-0.44ns	4.18**	-95.56**	0.07ns	-1.87*	-	1.38ns	-1.35**	3.62**	0.72**	2.24**
ZN-M-9 × ZM-R-10	-5.19**	-2.37**	-0.37ns	0.20ns	0.61ns	-	-19.33ns	-2.37**	1.06ns	-	-2.22*	-1.97**	-1.84**	0.28*	0.69**
ZM-M-6 × ZM-R-6	-0.74ns	0.52ns	0.63ns	-0.52ns	-0.94ns	0.69ns	-	-	1.52ns	-	-	1.46**	-2.99**	2.89**	3.11**
ZM-M-6 × ZM-R-2	3.81	2.96**	3.30**	-0.02ns	-0.22ns	0.16ns	-23.44ns	2.30**	-	0.03*	-	-2.14**	3.53**	-	-0.68**
ZM-M-6 × ZM-R-10	-3.07**	-3.48**	-3.93**	0.54ns	1.17ns	-	124.78**	-	-	-	1.56ns	0.68ns	-	0.64**	-2.43**
ZN-R-8 × ZM-R-6	-4.19**	-3.93**	-3.48**	-0.41ns	-0.94ns	3.10*	-15.89ns	-	-2.14*	0.03*	4.09**	-4.85**	5.27**	1.44**	-1.59**
ZN-R-8 × ZM-R-2	0.04ns	3.19**	3.19**	0.43ns	1.11ns	0.37ns	111.33**	-	0.97ns	-	-	5.13**	-3.05**	0.54**	-0.16*
ZN-R-8 × ZM-R-10	4.15**	0.74ns	0.30ns	-0.02ns	-0.17ns	-3.47**	-95.44**	0.63ns	1.17ns	-	-3.00**	-	-2.22**	-	1.75**
Shiralee × ZM-R-6	4.48**	3.41**	3.96**	0.81ns	1.39*	5.00**	-32.56ns	-	0.98ns	-0.04**	-4.67**	-3.68**	6.35**	-0.26*	0.69**
Shiralee × ZM-R-2	-4.96**	-1.81*	-1.37ns	-0.02ns	-0.22ns	-3.06*	56.67*	-	0.34ns	0.03*	2.57**	6.73**	-2.87**	2.28**	-3.01**
Shiralee × ZM-R-10	0.48ns	-1.59*	-2.59**	0.80ns	1.17ns	-	-24.11ns	1.30ns	-	0.02ns	2.11*	-3.05**	-3.48**	-	2.31**
ZN-M-11 × ZM-R-6	1.59*	-3.48**	-2.70**	0.15ns	1.39*	2.75*	4.78ns	-2.04*	-	-0.03*	1.01ns	1.41**	0.95*	-	2.03**
ZN-M-11 × ZM-R-2	-5.19**	-4.70**	-5.37**	-	-2.89**	-4.97**	-76.00**	0.07ns	0.28ns	0.02ns	-	-3.72**	1.68**	-	0.28**
ZN-M-11 × ZM-R-10	3.59**	8.19**	8.07**	0.20ns	1.50*	2.33ns	71.22**	1.96*	0.91ns	0.01ns	-	2.31**	-2.64**	2.93**	-2.31**

Table S4. Estimation of Mid parent and Better parent Heterosis for various *Brassica napus* L. traits

* = Significant at 0.05 probability level, ** = Significant at 0.01 probability level

DFI = Days to flower initiation, **D50%F** = Days taken to 50% flowering, **D50%S** = Days to 50% siliqua formation

MPH = Mid parent heterosis; **BPH** = Better parent heterosis

Traits	DFI		D50%F		D50%S		Primary Branches		Secondary Branches		Plant Height		Siliques/Plant	
	MPH	BPH	MPH	BPH	MPH	BPH	MPH	BPH	MPH	BPH	MPH	BPH	MPH	BPH
ZM-R-7 × ZM-R-6	-7.35**	-21.74**	-7.48**	-20.80**	-8.04**	-20.57**	4.00ns	-13.33ns	29.41ns	22.22ns	-6.24**	-12.14**	58.59*	53.42*

[Citation Awan, A.R., Abbas, M., Hassan, A., Sardar, S., Akbar, S., Naeem, A., Qadir, M.S., Ali, W., Nawaz, M.S., Aqeel, M., Mahmood, N. (2023). Development of *brassica napus* L. lines for high yield and essential fatty acids. *Biol. Clin. Sci. Res. J.*, 2023:329. doi: <https://doi.org/10.54112/bcsrj.v2023i1.329>]

ZM-R-7 × ZM-R-2	-13.97**	-26.40**	-17.03**	-28.53**	-16.79**	-26.74**	24.14ns	20.00ns	143.48**	86.67**	8.65**	-4.19**	56.65**	25.78ns
ZM-R-7 × ZM-R-10	-18.03**	-22.36**	-19.26**	-24.53**	-18.96**	-23.65**	11.11ns	0.00ns	-23.81ns	-38.46*	2.99**	-0.83ns	25.10ns	0.38ns
ZN-M-9 × ZM-R-6	-1.90ns	-15.13**	-2.07*	-14.72**	-3.34**	-15.20**	4.35ns	-7.69ns	-11.11ns	-11.11ns	11.51**	10.03**	72.11**	64.55*
ZN-M-9 × ZM-R-2	-4.69**	-16.45	-6.81**	-18.33**	-7.60**	-17.33**	-25.93ns	-28.57ns	16.67ns	-6.67ns	24.13**	17.78**	-5.63ns	-23.52ns
ZN-M-9 × ZM-R-10	-19.59**	-21.71**	-13.41**	-17.50**	-11.27**	-14.93**	-4.00ns	-7.69ns	-0.00ns	-15.38ns	10.38**	5.91**	18.96ns	-3.66ns
ZM-M-6 × ZM-R-6	0.41ns	-8.21**	-0.17ns	-7.64**	-0.33ns	-7.55**	-36.00*	-46.67**	-24.41ns	-33.33ns	4.42**	4.15**	-32.22ns	-37.78ns
ZM-M-6 × ZM-R-2	-3.82**	-10.82**	-3.93**	-10.51**	-4.31**	-9.37**	-24.14ns	-26.67 ns	30.43ns	0.00ns	11.13**	3.87**	16.66ns	-2.16ns
ZM-M-6 × ZM-R-10	-15.83**	-18.75**	-13.44**	-15.03**	-13.48**	-15.12**	-11.11ns	-20.00ns	23.81ns	-0.00ns	2.32*	-0.29ns	64.59**	37.93*
ZN-R-8 × ZM-R-6	-2.47ns	-10.23**	-4.01**	-10.13**	-4.28**	-10.46**	-0.00ns	-8.33ns	-42.86*	-50.00*	1.41ns	-0.86ns	-32.04ns	-33.06ns
ZN-R-8 × ZM-R-2	-7.10**	-13.26**	-2.95**	-8.50**	-4.35**	-8.62**	15.38ns	7.14ns	40.74*	26.67ns	6.15**	-2.63*	38.80*	7.53ns
ZN-R-8 × ZM-R-10	-6.88**	-10.76**	-8.86**	-11.66**	-9.72**	-12.21**	8.33ns	8.33ns	-28.00ns	-30.77ns	-3.96**	-4.52**	-38.51*	-52.39**
Shiralee × ZM-R-6	7.39**	-3.58**	2.89**	-5.90**	3.38**	-5.03**	27.27ns	-16.67	47.37*	40.00ns	3.01**	-1.42ns	-9.95ns	-22.59ns
Shiralee × ZM-R-2	-13.39**	-21.15**	-8.60**	-15.84**	-8.20**	-13.91**	-0.00ns	-7.14ns	28.00ns	6.67ns	3.96**	1.69ns	45.99*	2.73ns
Shiralee × ZM-R-10	-11.11**	-12.50**	-11.42**	-11.96**	-11.73**	-12.50**	-16.67ns	-16.67ns	-39.13*	-46.15*	-3.04**	-9.69**	11.35ns	-21.69ns
ZN-M-11 × ZM-R-6	5.60**	-5.04**	0.17ns	-7.86**	1.95*	-5.44**	30.00ns	30.00ns	29.41ns	22.22ns	6.10**	-1.00ns	-26.94ns	-40.53ns
ZN-M-11 × ZM-R-2	-12.03**	-19.78**	-7.30**	-14.15**	-7.18**	-12.08**	8.33ns	-7.14ns	-56.52**	-66.67**	7.57**	7.11**	-34.21ns	-55.41**
ZN-M-11 × ZM-R-10	-6.36**	-7.99**	1.55ns	0.31ns	2.22**	0.29ns	27.27ns	16.67ns	14.29ns	-7.69ns	4.53**	-4.99**	24.74ns	-15.49ns

Seeds per siliqua		Seed yield plant ⁻¹		100-seed weight		Oil content (%)		Protein content		Oleic acid content		Linolenic acid %		Linoleic acid %	
MPH	BPH	MPH	BPH	MPH	BPH	MPH	BPH	MPH	BPH	MPH	BPH	MPH	BPH	MPH	BPH
-3.03ns	-12.73	26.28*	15.86ns	9.50**	8.24*	-3.20ns	-5.03ns	2.33ns	0.09ns	-5.29**	-18.39**	25.48**	24.71**	-22.75**	-23.14**
1.03ns	-7.55ns	48.72**	25.45ns	-12.00**	-13.53**	1.93ns	-3.68ns	-16.22**	-18.53**	17.04**	10.34**	32.05**	16.19**	1.06ns	-8.52**
5.49ns	2.13ns	25.73ns	1.53ns	11.41**	7.69*	17.41**	5.94*	-0.30ns	-1.55ns	46.81**	46.58**	16.44**	9.25**	8.80**	-11.60**

[Citation Awan, A.R., Abbas, M., Hassan, A., Sardar, S., Akbar, S., Naeem, A., Qadir, M.S., Ali, W., Nawaz, M.S., Aqeel, M., Mahmood, N. (2023). Development of *brassica napus* L. lines for high yield and essential fatty acids. *Biol. Clin. Sci. Res. J.*, 2023:329. doi: <https://doi.org/10.54112/bcsrj.v2023i1.329>]

5.49ns	-12.73ns	62.26**	48.96**	22.07**	15.91**	-0.55ns	-1.92ns	17.84**	5.07**	6.20**	-4.99**	6.35*	-1.99ns	-21.69**	-30.65**
5.62ns	-11.32ns	41.97*	40.85**	-2.12ns	-4.39ns	7.06**	0.66ns	6.78**	-0.32ns	30.17**	27.95**	11.26**	5.14*	24.98**	21.37**
-8.43ns	-19.15*	79.81**	69.14**	7.58*	-0.00ns	2.00ns	-8.41**	-3.54ns	-11.24**	10.96**	6.51**	-0.97ns	-2.26ns	35.39**	23.10**
-3.09ns	-14.55*	39.32**	34.10*	3.51ns	-0.64ns	-10.89**	-17.15**	-5.96**	-8.09**	10.83**	-1.94ns	24.54**	7.09**	7.41**	1.07ns
32.63**	18.87*	35.34*	27.87ns	3.18ns	1.92ns	-8.40**	-8.57**	-13.25**	-19.21**	38.37**	34.34**	-35.52**	-36.84**	-9.74**	-12.99**
14.61ns	8.15ns	-2.79ns	-12.69ns	5.97*	-0.45ns	4.77ns	-0.43ns	-11.13**	-16.02**	22.95**	19.47**	-12.35**	-19.73**	-5.99**	-19.35**
-7.22ns	-18.18*	-20.61ns	-27.92*	14.25**	7.14*	14.63**	-0.17ns	-10.60**	-19.79**	1.52ns	-0.63ns	44.34**	41.02**	-27.83**	-36.00**
3.16ns	-7.55ns	36.40*	14.00ns	-2.82ns	-6.28*	7.32*	0.15ns	30.48**	22.60**	-7.02**	-13.85**	20.36**	3.26ns	-10.42**	-12.88**
14.61ns	8.51ns	26.92ns	1.61ns	3.46ns	-5.00ns	6.95*	4.72ns	2.57ns	-5.01**	-11.97**	-22.62**	-13.23**	-20.78**	25.14**	13.63**
-9.62ns	-14.55*	47.52**	35.86*	5.04ns	3.14ns	-2.20ns	-9.45**	-19.26**	-21.19**	7.99**	7.12**	8.24**	-1.26ns	-9.83**	-18.32**
-3.92ns	-7.55ns	58.26**	33.95*	10.02**	4.99ns	24.87**	24.54**	19.27**	16.21**	-1.67ns	-10.02**	19.57**	14.20**	-28.90**	-29.20**
14.58*	12.24ns	21.89ns	-1.25ns	18.08**	17.57**	29.70**	23.79**	-20.52**	-21.37**	-9.51**	-21.38**	-25.89**	-27.65**	31.96**	17.30**
-23.71**	-32.73**	16.92ns	13.41ns	3.87ns	0.39ns	-3.63ns	-4.77ns	-1.97ns	-7.03**	1.41ns	-0.55ns	-11.49**	-21.90**	-12.51**	-18.04**
3.16ns	-7.55ns	57.63**	39.78*	2.88ns	2.34ns	-0.62ns	-6.73*	-14.92**	-15.24**	12.47**	4.02**	-11.07**	-11.91**	-17.26**	-19.87**
19.10*	12.77ns	55.03**	31.20*	12.89**	6.76*	1.73ns	-8.80**	-1.68ns	-3.55ns	-3.15*	-15.02**	12.83**	6.25*	-21.12**	-32.05**

* = Significant at 0.05 probability level, ** = Significant at 0.01 probability level

MPH = Mid parent heterosis; **BPH** = Better parent heterosis

Fig. 1 Mean comparison of parents and F₁ hybrids for days taken to flower initiation

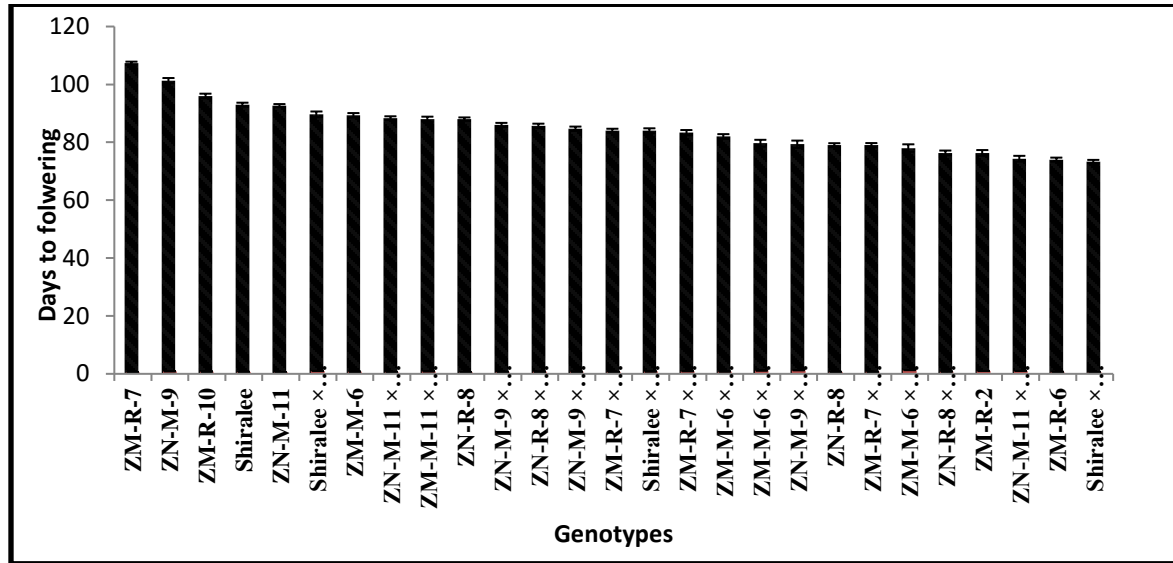


Fig. 2 Mean comparison of parents and F₁ hybrids for days taken to 50 % flowering

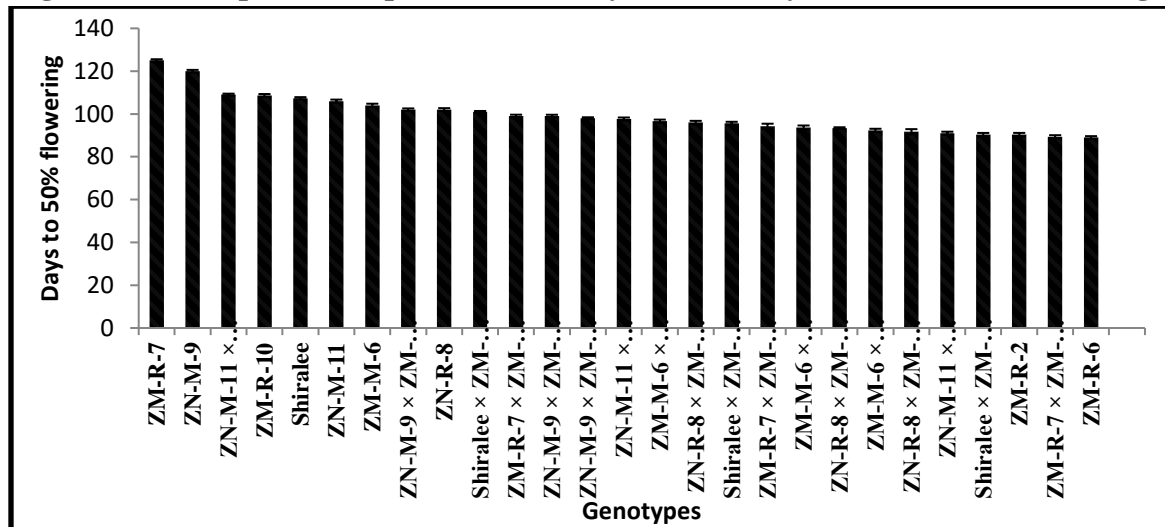
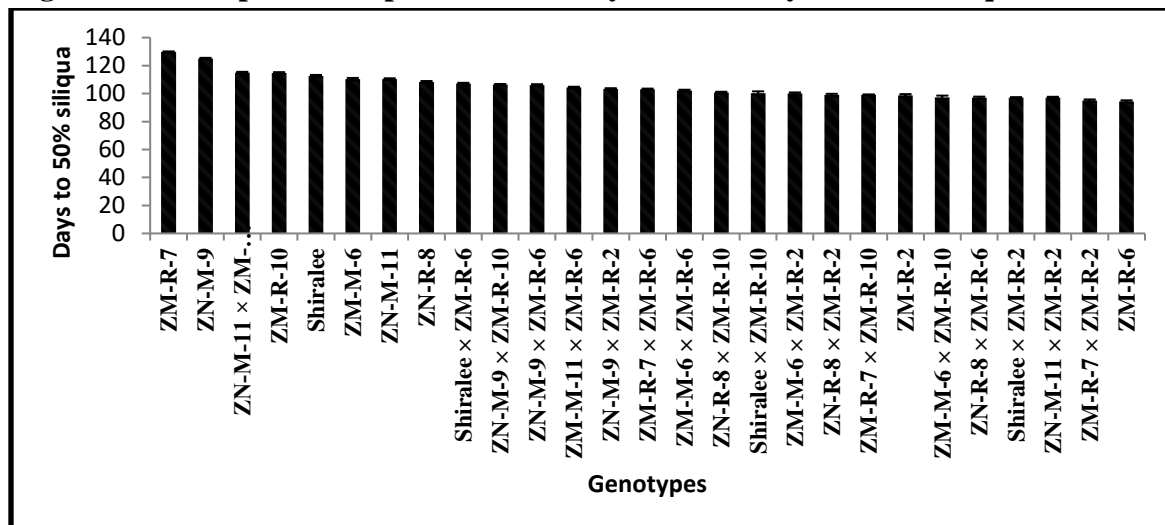


Fig. 3 Mean comparison of parents and F₁ hybrids for days to 50 % siliqua formation



[Citation Awan, A.R., Abbas, M., Hassan, A., Sardar, S., Akbar, S., Naem, A., Qadir, M.S., Ali, W., Nawaz, M.S., Aqeel, M., Mahmood, N. (2023). Development of *brassica napus* l. lines for high yield and essential fatty acids. *Biol. Clin. Sci. Res. J.*, 2023:329. doi: <https://doi.org/10.54112/bcsrj.v2023i1.329>]

Fig. 4 Mean comparison of parents and F₁ hybrids for number of primary branches

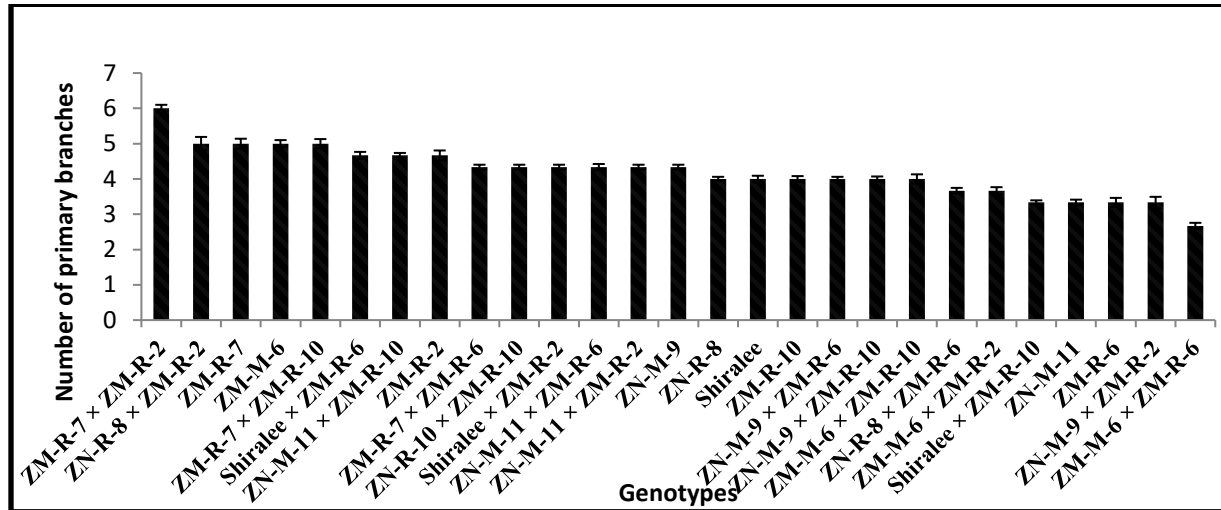


Fig. 5 Mean comparison of parents and F₁ hybrids for number of secondary branches

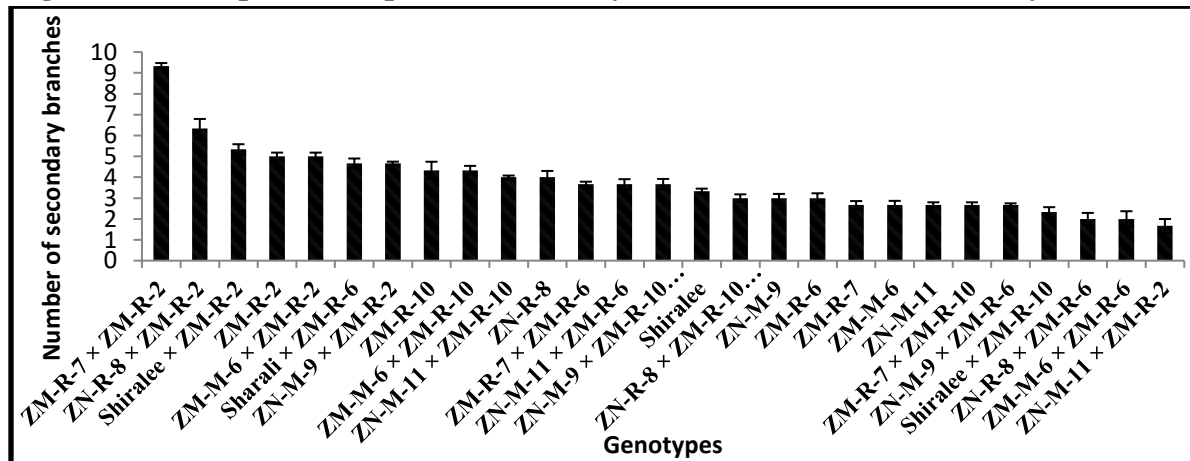
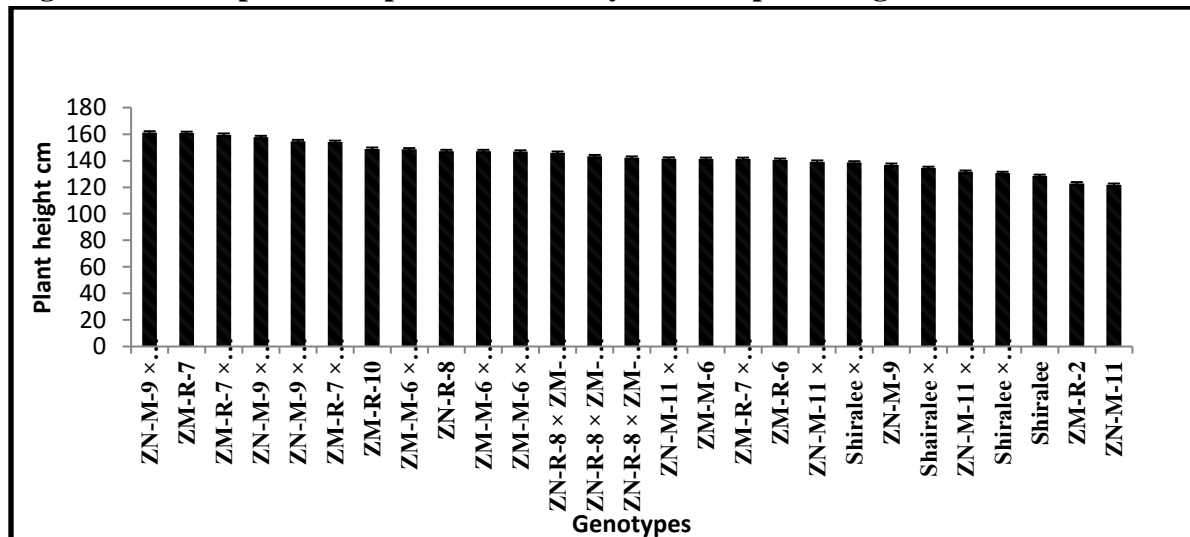


Fig. 6 Mean comparison of parents and F₁ hybrids for plant height



[Citation Awan, A.R., Abbas, M., Hassan, A., Sardar, S., Akbar, S., Naem, A., Qadir, M.S., Ali, W., Nawaz, M.S., Aqeel, M., Mahmood, N. (2023). Development of *brassica napus* l. lines for high yield and essential fatty acids. *Biol. Clin. Sci. Res. J.*, 2023:329. doi: <https://doi.org/10.54112/bcsrj.v2023i1.329>]

Fig. 7 Mean comparison of parents and F₁ hybrids for siliquae per plant

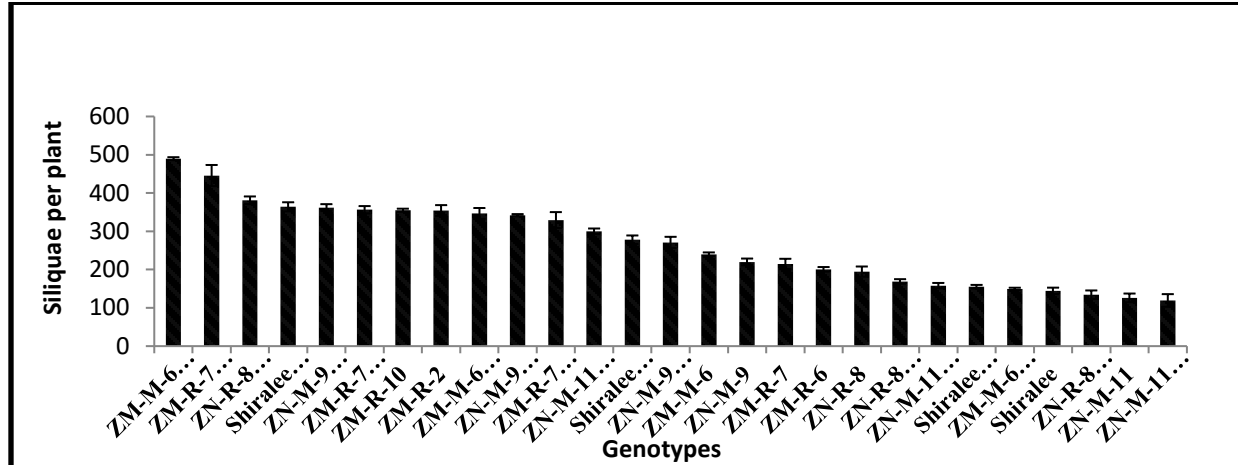


Fig. 8 Mean comparison of parents and F₁ hybrids for seed per siliqua

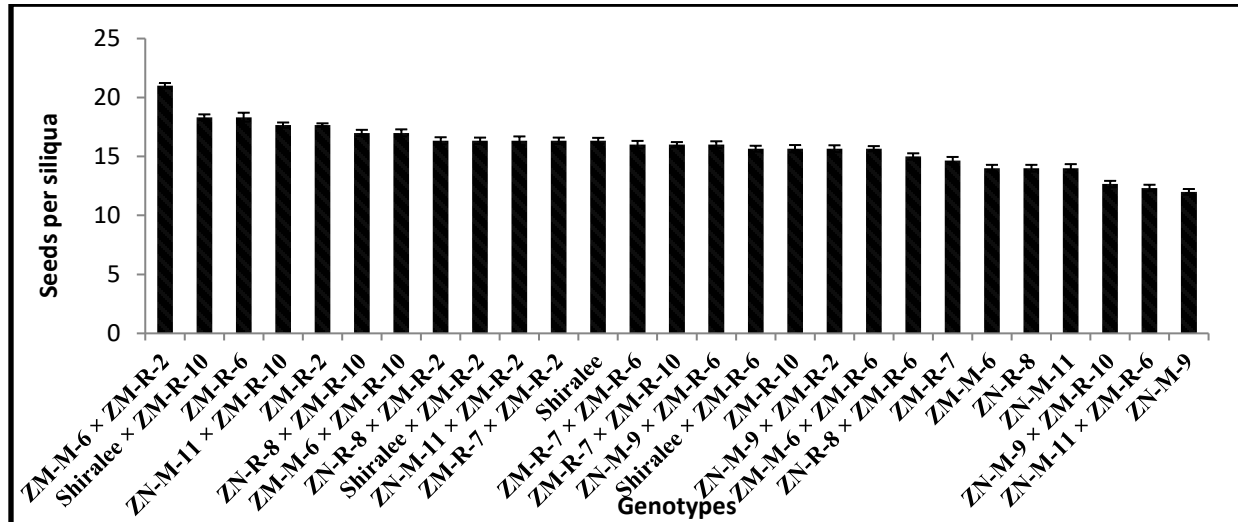
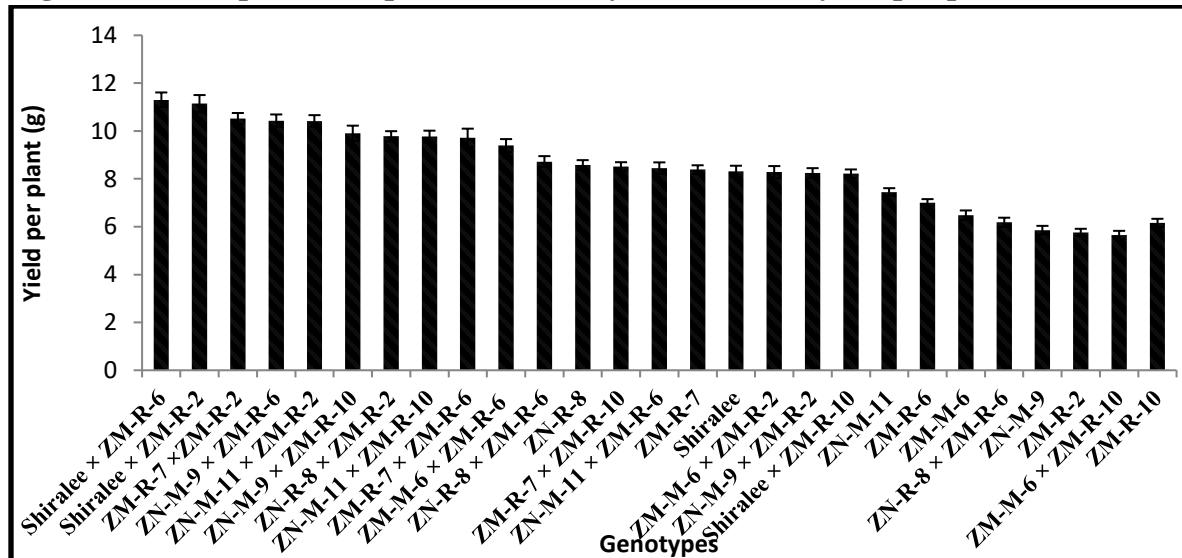


Fig. 9 Mean comparison of parents and F₁ hybrids for seed yield per plant



[Citation Awan, A.R., Abbas, M., Hassan, A., Sardar, S., Akbar, S., Naem, A., Qadir, M.S., Ali, W., Nawaz, M.S., Aqeel, M., Mahmood, N. (2023). Development of *brassica napus* l. lines for high yield and essential fatty acids. *Biol. Clin. Sci. Res. J.*, 2023:329. doi: <https://doi.org/10.54112/bcsrj.v2023i1.329>]

Fig. 10 Mean comparison of parents and F₁ hybrids for 100-seed weight per plant

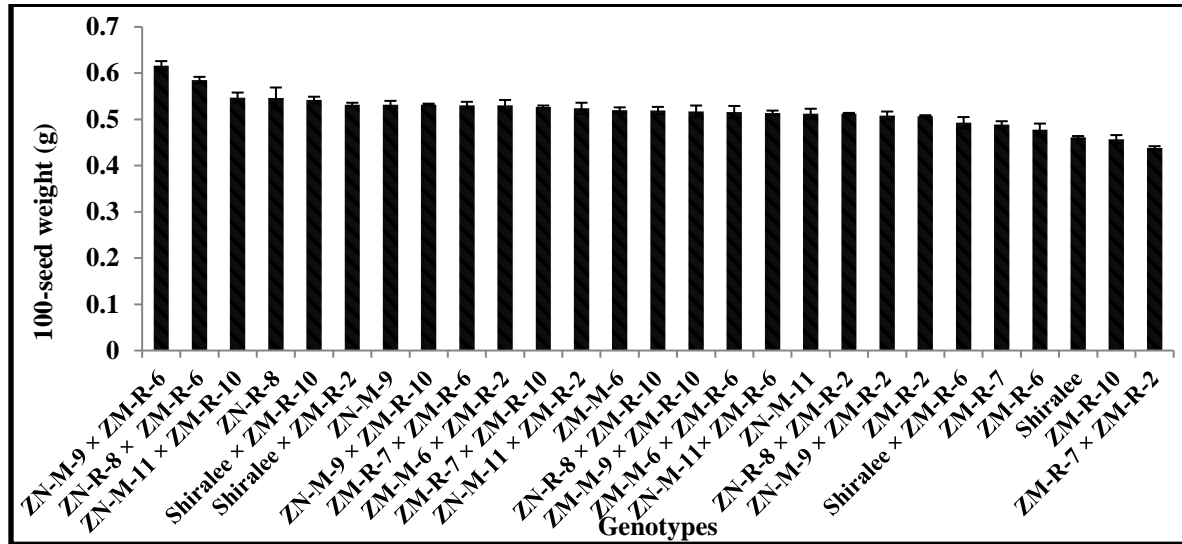


Fig. 11 Mean comparison of parents and F₁ hybrids for oil percentage

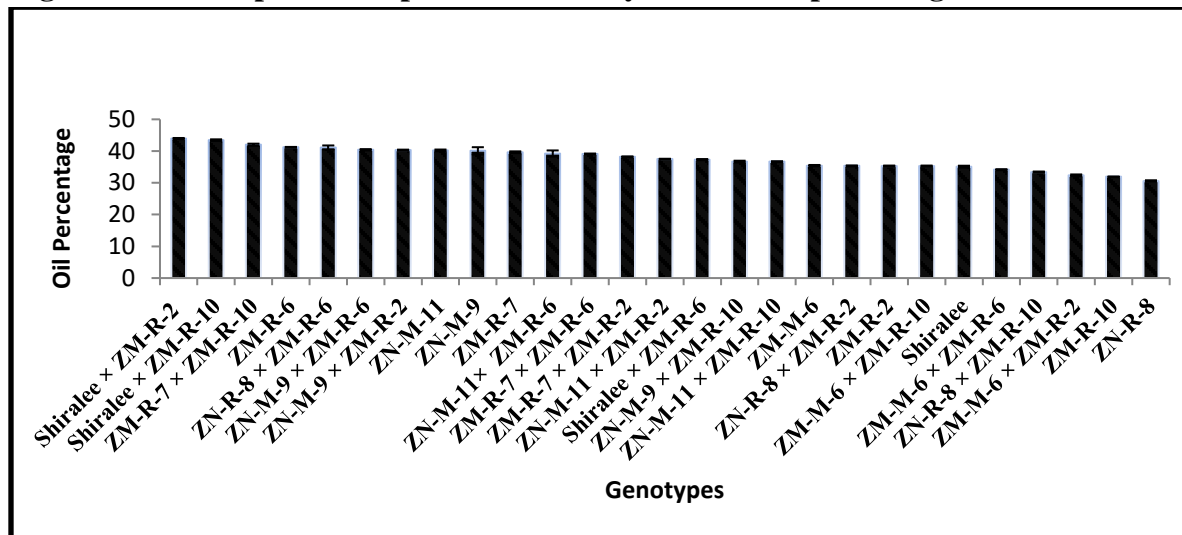
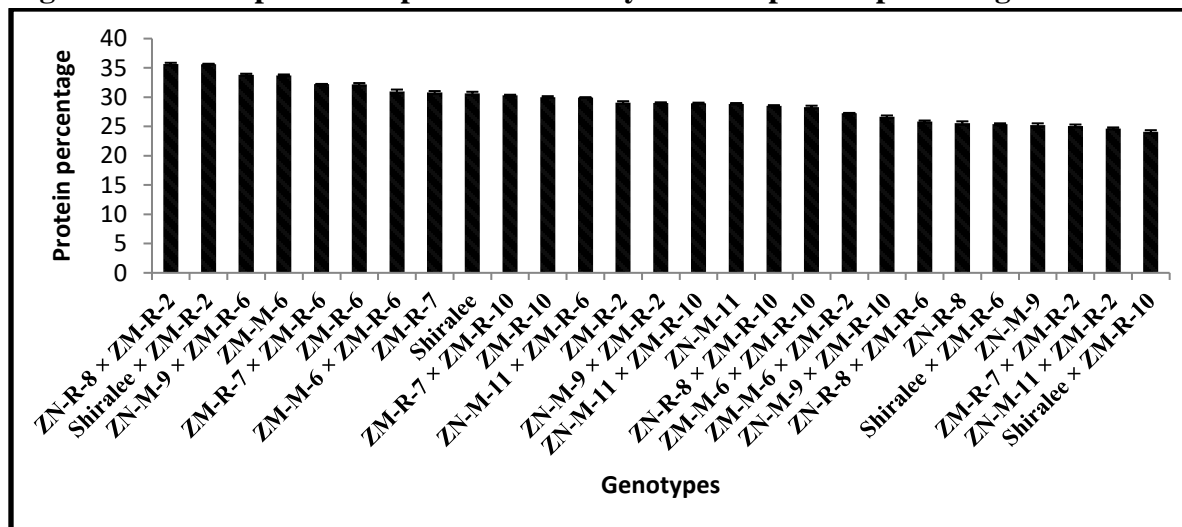


Fig. 12 Mean comparison of parents and F₁ hybrids for protein percentage



[Citation Awan, A.R., Abbas, M., Hassan, A., Sardar, S., Akbar, S., Naem, A., Qadir, M.S., Ali, W., Nawaz, M.S., Aqeel, M., Mahmood, N. (2023). Development of *brassica napus* l. lines for high yield and essential fatty acids. *Biol. Clin. Sci. Res. J.*, 2023:329. doi: <https://doi.org/10.54112/bcsrj.v2023i1.329>]

Fig. 13 Mean comparison of parents and F₁ hybrids for oleic acid percentage

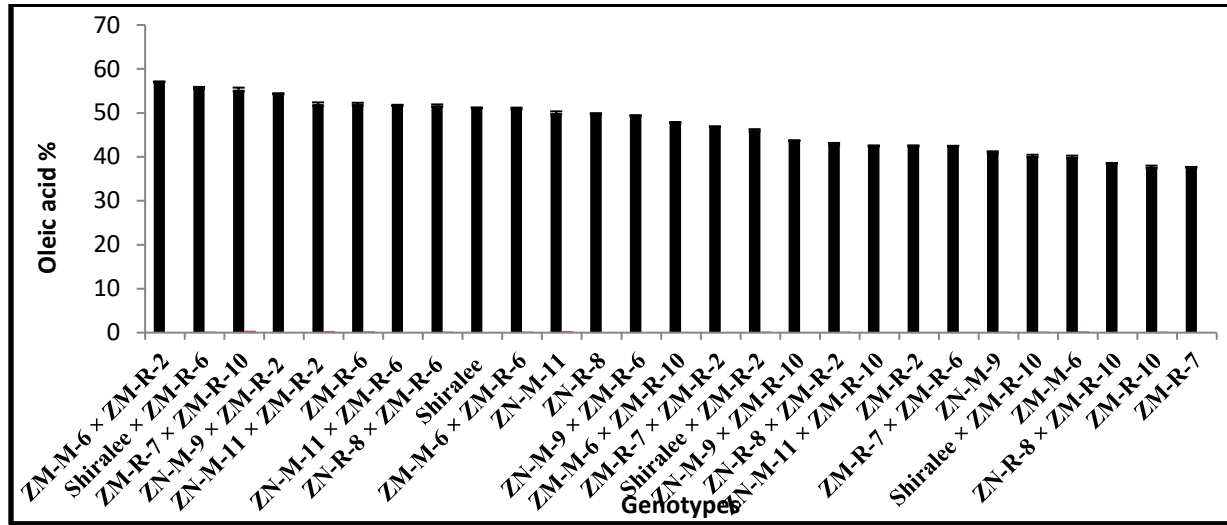


Fig. 14 Mean comparison of parents and F₁ hybrids for linolenic acid percentage

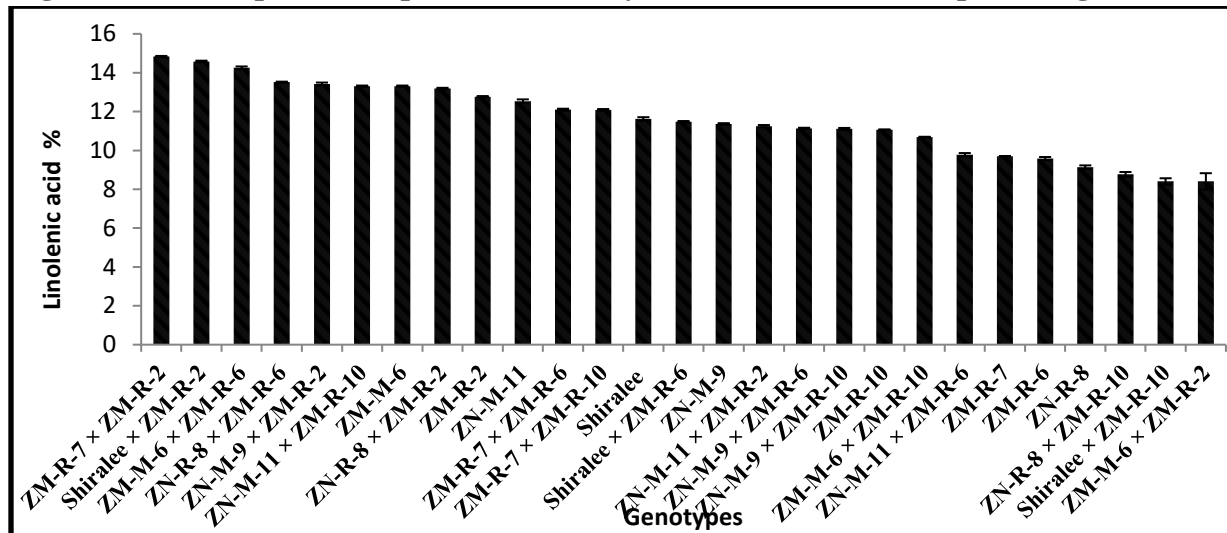
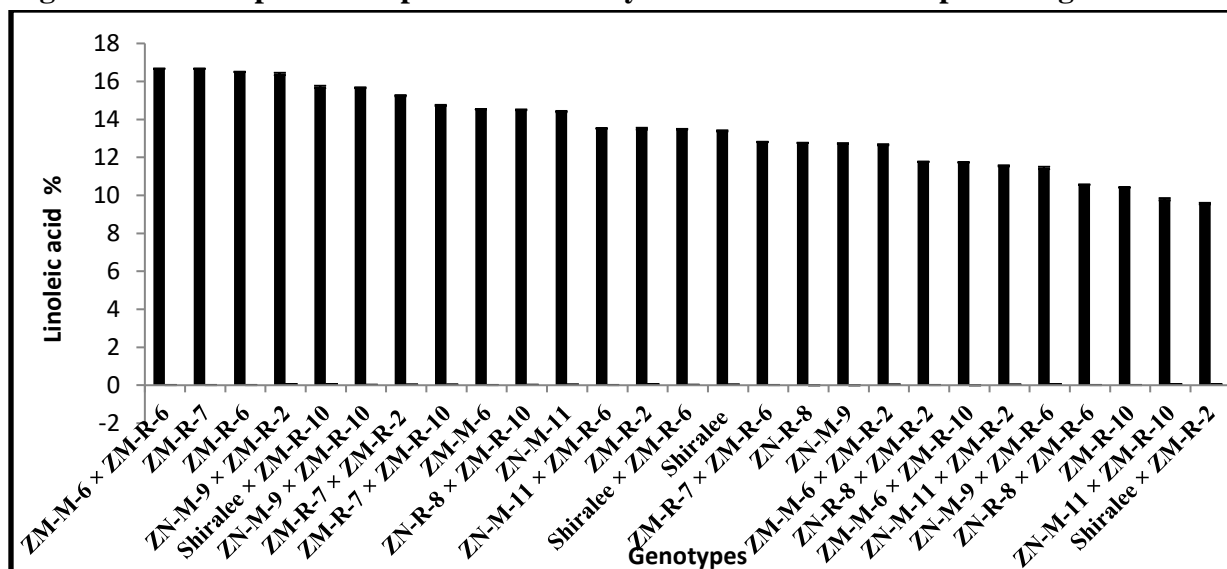


Fig. 15 Mean comparison of parents and F₁ hybrids for linoleic acid percentage



[Citation Awan, A.R., Abbas, M., Hassan, A., Sardar, S., Akbar, S., Naem, A., Qadir, M.S., Ali, W., Nawaz, M.S., Aqeel, M., Mahmood, N. (2023). Development of *brassica napus* l. lines for high yield and essential fatty acids. *Biol. Clin. Sci. Res. J.*, 2023:329. doi: <https://doi.org/10.54112/bcsrj.v2023i1.329>]



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