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RESEARCH ARTICLE

Identification and correlation analysis of fatty acid profile and morphological traits in Camelina (*Camelina sativa* L. Crantz)

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Abstract

Oilseeds are the world's second-largest food reserve after cereals. *Camelina sativa*, a member of the Brassicaceae family, is an oilseed crop known for its adaptability to rainfed cultivation, low input requirements and high-quality oil. This study aimed to identify key traits influencing seed yield and oil composition, particularly fatty acid profiles, in *Camelina sativa*. The research was conducted at Razi University in Kermanshah, Iran. Using gas chromatography, we analysed the fatty acid profiles of 136 doubled haploid camelina lines, detecting 18 distinct fatty acids categorised into saturated, monounsaturated and polyunsaturated types. Significant variations were observed in the fatty acid composition among the lines. Yield components such as NPP, BY, SHW and PSW showed strong positive correlations with grain yield (GY) ($r \leq 0.813$), while traits like SL, SP, SA and WS exhibited weak negative correlations. A notable negative correlation was found between oil and protein content ($r = -0.769$, $P < 0.01$), ranging from 32.72 % to 38.55 % and 25.66 % to 30.45 %. Linolenic acid levels ranged from 28.95 % to 34.90 % and linoleic acid levels varied between 16.07 % and 22.24 %. These findings highlight the potential of *Camelina sativa* as a sustainable oilseed crop for marginal environments. Identifying traits directly related to seed yield is a critical aspect of plant breeding, as it enables the development of improved varieties with higher productivity. Future research should optimise breeding strategies to enhance yield and oil quality while exploring its potential in biofuel and nutritional applications.

Keywords

brassicaceae; correlation; camelina; fatty acid; oil seeds; protein

Introduction

Camelina (*Camelina sativa* L. Crantz), commonly known as false flax or gold-of-pleasure, is a member of the Brassicaceae family and is recognised as an essential oilseed crop (1). Camelina is a low-input crop with high nutrient efficiency, making it suitable for cultivation on marginal lands without nitrogen fertilisation. This crop is well-adapted to temperate climates due to its short growing season. Camelina seedlings exhibit frost resistance and can germinate at low temperatures (2). Under drought-stress conditions, camelina demonstrates remarkable resilience, making it more suitable for low-rainfall regions than other oilseed crops (3).

Camelina seeds have industrial and nutritional applications due to their high oil content and unique properties. The seeds contain 38 to 43 % oil and 27 to 32 % protein (4). Camelina oil comprises more than 50 % polyunsaturated fatty acids (5). The oil is highly stable and resistant to oxidation and rancidity, owing to its high content of natural antioxidants, such as tocopherols (6). With a vitamin E content of approximately 110 mg/100 g, Camelina oil is suitable for use as cooking oil. The crop is also being extensively researched due to its exceptionally high levels of linolenic acid (up to 45 %), which is uncommon in most vegetable oils (7). Seed quality traits are crucial for the marketing and processing of camelina in competition with other oilseeds. As a result, part of the focus in Camelina breeding programs has been on selecting seeds that meet specific quality criteria, such as thousand-seed weight, oil content, or modified fatty acid composition (8). However, the practice of selecting seeds with specific quality characteristics has been limited. The genetic differences among Camelina genotypes are not the sole cause of the wide variation in seed quality parameters reported in previous studies. Environmental conditions may also play a significant role in the difficulty of comparing specific results (9).

In the current investigation, doubled haploid (DH) lines were utilised. Using doubled haploid populations in plant breeding programs and genetic research offers the advantage of producing entirely homozygous individuals. A doubled haploid line represents a specific combination of traits from both parents in the initial cross. Doubled haploids allow for the direct fixation of recombinant gametes' genes as fertile homozygous lines. The benefits of doubled haploids in breeding include their deployability, fertility and homozygosity for all traits, which ensures proper breeding in subsequent generations.

In contrast, traditional breeding methods often face the challenge of dominant alleles masking recessive alleles in heterozygous diploid lines. However, recessive genes can be expressed in doubled haploids due to their complete homozygosity at all gene loci. This enhances the accuracy and reliability of selection in breeding programs (10).

Camelina oil offers numerous benefits but also faces several challenges, including poor performance, limited availability and an uncertain market (11). To address these issues, it is essential to enhance research efforts and increase awareness of Camelina oil (9). This study monitored sets of doubled haploid lines concerning their agronomic characteristics and biochemical parameters. The following objectives were considered in this research: assessing 136 camelina doubled haploid lines for physiological traits and fatty acid levels, analysing the relationship between physiological traits and fatty acid profiles, identifying key breeding traits to enhance the fatty acid profile and determining and investigating the levels of fatty acids in camelina seed oil.

Materials and Methods

Experimental area, design and plant genetic materials

Based on previous research conducted, the 136 Camelina doubled haploid lines used in this study were obtained (12). These lines were investigated using a randomised complete block design (RCBD) with three replications under rainfed conditions at the Razi University Experimental Field in Kermanshah, Iran. The experimental site is located at a longitude of 47°10′ and a latitude of 34° 32′, with an altitude of 1319 m above sea level. The region experiences an average annual rainfall of 410 mm and is characterised by the cool temperate climate of the northern Zagros Mountains. The mean annual temperature ranges between 5.9 °C and 28.6 °C and the soil texture at the site is predominantly silty clay. During the experimental period, the recorded rainfall was 609.5 mm. The seed density was set at 400 seeds per square meter and each genotype was planted in three rows, each with a length of 1 m. Cultivation was carried out during the 2019-2020 cropping season. The experimental farm, characterised by silty clay soil texture, is situated in a region with a moderate and cold climate, receiving 435 mm of rainfall during the cropping season.

Character measurements

At harvest time, yield (Y) was measured based on the seedpods (silicles) collected from five plants. The following morphological and agro-physiological characteristics were measured: grain yield and 1000-seed weight, expressed on an 80 mg/g seed moisture basis (Table 1).

Biochemical analysis

The seed lines were made ready for oil extraction after being cleaned from external contamination. The oil from the seeds was extracted using a cold press oil extraction machine (Ulimac model UM200). The Iranian national standard No. 493 was adhered to during the oil sampling. All experiments were carried out at the Agriculture and Natural Resources Campus of Razi University's laboratory.

Table 1. Investigated traits and abbreviations in this study

Row	Traits	Abbreviation
1	Grain yield or seedpods (silicles) per 5 plant (g/5 plant)	G. Y
2	Number of pod per plant	N. P. P
3	Number of pod per main branch plant	N. P. M. B
4	Number of pod per lateral branch plant	N. P. L. B
5	Biological yield per five plant	B. Y
6	Plant height with roots	P. H
7	Root length	R. L
8	Root weight	R. W
9	Shoots weight	SH. W
10	Pod straw weight	P. S. W
11	Number of seed in pod	N. S. P
12	Number of lateral branch	N. L. B
13	Length of lateral branch	L. L. B
14	Length of main branch	L. M. B
15	Seed length	S. L
16	Seed perimeter	S. P
17	Seed area	S. A
18	Weight of 1000 seeds	W.S.

The identification of fatty acid by gas chromatography (Agilent/HP model 6890) was done. This research utilised a gas chromatography device with a 120-meter capillary tube and Flame Ionization Detector (FID). The sample volume that was injected into the device was one microliter. The injection temperature was 250 °C, the oven temperature was 192 °C and the nitrogen carrier gas was used with a 1 mL/min flow rate. The oil content of the seed samples was determined and reported by comparing their dry weight percentage and estimating the number of fatty acids by comparing their subpeak area with standard samples and comparing it to the total oil percentage (C:12-C:24, Sigma Company). All tests were repeated four times. The results were analysed in a completely random design using SAS V.14.0 and SPSS V.16.0 software and graphs were created using Excel and GraphPad Prism software and correlation coefficients with the software SAS V.14.0.

Results

Morphological and agro-physiological traits

To assess their variance, the analysis of a randomised complete block design, utilising 18 morphological and agro-physiological traits, was conducted across 136 haploid lines. The results revealed that the lines examined in this study exhibited significant differences in all evaluated traits (Table 2). The variation in fatty acid content among doubled haploid Camelina lines will enhance the breeding potential for developing cultivars with specific characteristics. The selection of cultivars for particular applications will be guided by the composition of fatty acids in seed oil (12). Rapeseed, the most prominent member of the Camelina family, has been extensively studied to develop varieties with reduced glucosinolates and erucic acid (13). These efforts led to the creation of the first canola cultivar with low glucosinolate content in 1967. By 1974, a canola variety with low glucosinolates and erucic acid levels was introduced to the market (14).

Oil content and fatty acid profiles in camelina

The oil content in Camelina seeds ranged from 32.72 % to 38.55 %, while the protein content was estimated to be between 25.66 % and 30.45 %. The highest oil content in camelina seeds was observed in line 45, with an average of 38.55 %. The highest protein content was observed in line number 133, with a value of 31.73 %. Hydraulic pressing is the most widely used method for extracting oil from seeds. This study employed the press method to understand the oil extraction process better. The identification and quantification of 18 different fatty acids were performed in the oil extracted from the seeds of doubled haploid Camelina lines (Table 3). Camelina oil contains saturated fatty acids (SFAs), monounsaturated fatty acids (MUFAs) and polyunsaturated fatty acids (PUFAs) as its primary fatty acid components. Specifically, camelina oil predominantly comprises MUFAs and PUFAs (12). The majority of vegetable oil produced worldwide is consumed by humans. In recent years, an increasing proportion of this valuable agricultural commodity has been utilised to produce biodiesel and other industrial products (9). The growing demand for vegetable oils has driven the discovery of new sources of vegetable fats, particularly for non-food applications. Doubled haploid lines of camelina are valuable for investigating genetic and biological parameters and monitoring the fatty acid biosynthesis

Table 3. Fatty acids in camelina seed oil

Row	Type of fatty acid	C:D*
1	Lauric acid	C12:0
2	Myristic acid	C14:0
3	Palmitic acid	C16:0
4	Palmitoleic acid	C16:1
5	Citric acid	C18:0
6	Oleic acid	C18:1
7	Linoleic acid	C18:2
8	Linolenic acid	C18:3
9	Eicosanoic acid	C20:0
10	Eicosenoic acid	C20:1
11	Eicosadienoic acid	C20:2
12	Eicosatrienoic acid	C20:3
13	Behenic acid	C22:0
14	Erucic acid	C22:1
15	Docosadienoic acid	C22:2
16	Docosatrienoic acid	C22:3
17	Lignoceric acid	C24:0
18	Nervonic acid	C24:1

* :"C" stands for Carbon; "D" stands for Doubled bond; "C: D "is the ratio of the total amount of Carbon atoms of the fatty acid concerning the number of doubled (*unsaturated*) bonds in it.

Table 2. Analysing the variation of traits studied

S.O.V.	DF	Mean squares					
		GY	NPP	NPMB	NPLB	BY	PH
Replication	2	19.39	137.19	67.33	12.76	747.49	8.75
Genotype	135	51.27**	766.68**	326.76**	133.85*	725.47**	40.40**
Error	270	6.08	57.29	23.96	17.65	80.23	13.089
C.V. (%)	-	31.22	11.47	10.79	20.39	27.25	7.56
S.O.V	DF	Mean squares					
		RL	RW	SHW	PSW	NSP	NLB
Replication	2	17.59*	1.65	61.70	1.35	41.91	2.91
Genotype	135	16.29**	4.62**	135.55**	24.23**	39.34**	7.36**
Error	270	4.10	0.63	18.51	2.90	10.49	1.19
C.V. (%)	-	17.78	30.98	28.99	27.49	29.26	18.21
S.O.V	DF	Mean squares					
		LLB	LMB	SL	SP	SA	WS
Replication	2	6.41	2.74	0.004	0.016	0.019	0.004
Genotype	135	179.85**	90.64**	0.07**	0.49**	0.20**	0.083**
Error	270	16.76	8.71	4.10	8.71	0.03	0.001
C.V. (%)	-	15.33	11.84	6.28	5.34	11.71	3.51

* and **: Not significant, significant at the 5 % and 1 % probability levels, respectively.

pathway. The analysis of variance (ANOVA) for fatty acid composition in Camelina lines is presented in Table 4. Table 4 presents the analysis of variance for fatty acid composition among the Camelina lines. Based on the results presented in the table, it can be concluded that there are significant differences in the fatty acid profiles among the lines examined in this study.

The fatty acids in Camelina seed oil

Camelina seed oil contains three main categories of fatty acids: saturated fatty acids, monounsaturated fatty acids and polyunsaturated fatty acids. These groups represent the primary classification of fatty acids present in the oil.

Saturated fatty acids in Camelina seed oil

Camellia seeds contained seven different kinds of saturated fatty acids in their oil combination including Lauric acid (C12:0), Myristic acid (C14:0), Palmitic acid (C16:0), Stearic acid (C18:0), Eicosanoic acid (C20:0), Behenic acid (C22:0) and Lignoseric acid (C24:0), (Fig. 1).

Monounsaturated fatty acids in Camelina seed oil

Camelina seed oil has five types of monounsaturated fatty acids (MUFAs), which include Palmitoleic acid (C16:1), Oleic acid (C18:1), Eicosenoic acid (C20:1), Erucic acid (C22:1) and Nervonic acid (C24:1) (Fig. 2).

In plants, membrane fluidity is regulated by the ratio of unsaturated fatty acids to sterols. This fluidity is critical in maintaining membrane stability under abiotic stress conditions. Various breeding techniques successfully increased the proportion of unsaturated fatty acids in tobacco plants. These efforts led to the development of cold -resistant tobacco cultivars (14). To enhance the resistance of Camelina plants to abiotic stresses, it is essential to identify optimal ratios of unsaturated fatty acids and their specific functional roles (15).

Polyunsaturated fatty acids in Camelina seed oil

There are 6 types of polyunsaturated fatty acids (PUFAs) of fatty acids in camellia seed oil: Linoleic acid (C18:2), Linolenic acid (C18:3), Eicosadienoic acid (C20:2), Eicosatrienoic acid (C20:3), Docasadienoic acid (C22:2) and Docasatetraenoic acid (C22:3), which average amount Each of these 6 types of fatty acids was measured in 137

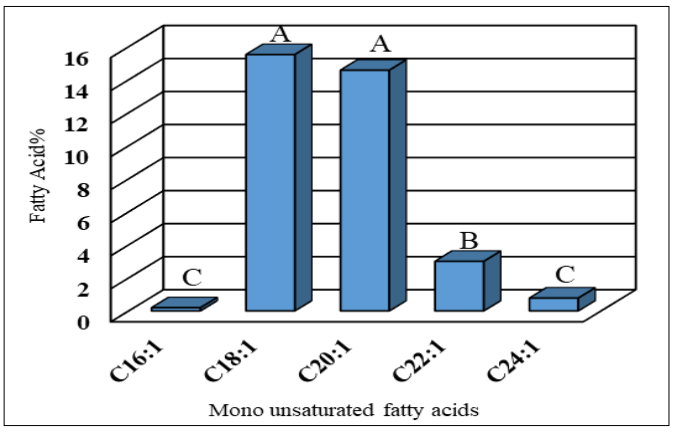

Fig. 2. Mean comparison of monounsaturated fatty acids in Camelina seed oil.

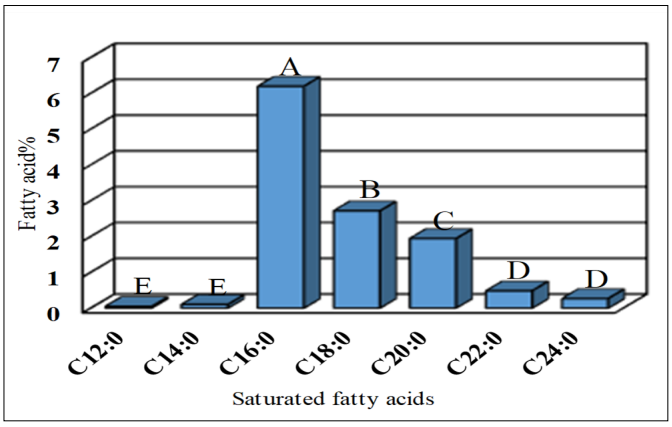

Fig. 1. Camelina seed oil saturated fatty acids and their mean comparison.

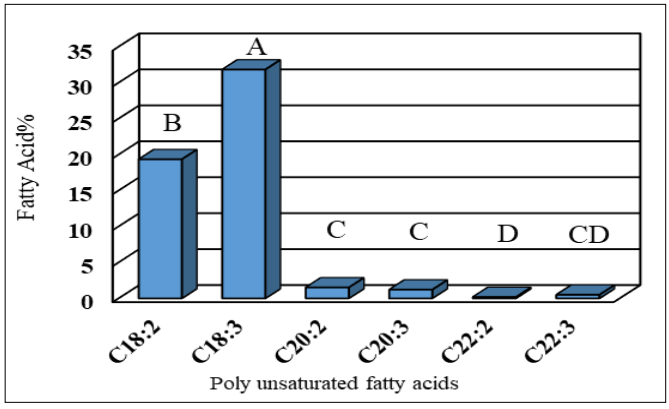

Fig. 3. Polyunsaturated fatty acids in camelina seed oil.

Table 4. Analysis of variance for studied traits in *Camelina sativa*

S.O.V.	DF	Mean squares					
		C12:0	C14:0	C16:0	C16:1	C18:0	C18:1
Replication	3	0.591	0.515	14.622	0.473	3.421	119.05
Genotype	136	0.00**	0.00**	0.24**	0.0004*	0.51**	4.75**
Error	408	6.08	0.00	0.0002	0.00002	0.0004	0.005
C.V. (%)	-	0.55	0.42	0.22	2.62	0.79	0.46
S.O.V.	DF	Mean squares					
		C18:2	C18:3	C20:0	C20:1	C20:2	C20:3
Replication	3	111.59	316.44	0.136	67.97	0.030	0.012
Genotype	136	6.94**	5.81**	0.22**	1.15**	0.010**	0.05**
Error	408	0.005	0.004	0.0001	0.0009	0.000	0.000
C.V. (%)	-	0.36	0.20	0.69	0.20	0.60	0.56
S.O.V.	DF	Mean squares					
		C22:0	C22:1	C22:2	C22:3	C24:0	C24:1
Replication	3	0.501	0.691	0.533	0.264	0.461	0.133
Genotype	136	0.007**	0.23**	0.0018**	0.013**	0.005**	0.0016**
Error	408	0.000	0.0001	0.000	0.000	0.000	0.000
C.V. (%)	-	1.878	0.432	0.655	0.689	0.797	0.494

* and **: Not significant, significant at the 5 % and 1 % probability levels, respectively.

Camelina lines (Fig. 3).

PUFAs significantly impact organisms' function and structure more than other fatty acids. The health of humans and other mammals is benefitted by the valuable and essential functions of PUFAs, which are involved in the prevention and treatment of various diseases. Human and mammalian bodies cannot produce precursors of PUFAs. Therefore, having a balanced and adequate amount of these fatty acids in the diet is crucial (16).

The correlation between morphological and physiological traits

Given that one of the purposes of this study is to identify correlations that have the most significant correlation with grain yield, the correlation coefficients between different traits were determined. The linkage between genes, non-allelic interactions and pleiotropy can cause a correlation between two traits. The correlation matrix of the morphological and physiological characteristics examined is shown in Table 5. In this research, the positive correlation of some yield components such as NPP, BY, SHW and PSW revealed the most positive and significant correlation ($X \leq 0.813$) with grain yield (GY). The correlation between grain yield (GY) and traits (NPMB, NPLB, PH, RL, NLB, LLB and LMB) was positive but not as strong as the previous traits. The correlation between the NSP trait and grain yield (GY) was positive but insignificant.

A line × tester analysis in rapeseed (*Brassica napus* L.) revealed a positive and significant relationship between the number of siliques per plant and grain yield (17). Research indicates a strong and positive relationship between the number of seeds in the siliqua and the weight of 1000 seeds in rapeseed (18). In contrast, research disapproved of this hypothesis and stated that the weight of 1000 seeds has a direct and slight positive impact on the yield. The trait of the number of siliques per plant is more effective on seed yield and it stated that this trait explains 40 % of the variation in rape seed yield (19). In the present investigation, the seed yield is directly and significantly correlated with the number of pods per plant (0.824). The research results showed that the trait of siliqua number in canola plants has a positive correlation with yield and siliqua's seed number trait has a negative correlation with yield (20).

The negative and positive correlations in the correlation table require attention when using plant breeding programs and selecting traits. According to Table 5 and Fig. 4, SL (0.078), SP (0.074), SA (0.100) and WS (0.042) are negatively but not significantly correlated with grain yield (GY) and other traits (Fig. 4). It's possible to describe this issue in this manner: Increasing the seed's length, area and perimeter can limit the growth of other lateral seeds in pod-limited environments. In oil Camelina, the amount of oil and protein are two significant traits. The amount of oil and protein in Camelina oil is depicted in Fig. 7. Estimates were made of the minimum and maximum amount of Camelina oil at (32.72 - 38.55 %). The maximum oil content (38.55 %) was found in line number 45, whereas the minimum oil content (32.72 %) was observed in line 137. The protein content in Camelina oil was estimated to range between 25.66 and 30.45 %. The maximum protein content in camelina seed oil (30.45 %) was detected in line number 133, whereas the minimum protein content (25.66 %) was found in line 98. Paying attention to the correlation between these two traits is crucial in the plant breeding program (Fig.5A). Fig. 5 shows the Pearson correlation coefficient between oil and protein content (Figure 5B).

Linoleic acid and alpha-linolenic acid in the Camelina oil

Due to its diverse fatty acids, Camelina oil can benefit many applications. Essential fatty acids can be found in Camelina oil, such as (linoleic acid 18:2 and Linolenic acid 18:3). Linoleic acid (LA) and alpha-linolenic acid (ALA) are essential fatty acids in the PUFA category. According to many researchers, camelina oil mainly comprises LA and

Table 5. Correlation matrix of the morphological and physiological traits

GY	NPP	NPMB	NPLB	BY	PH	RL	W	SHW	PSW	NSP	NLB	LLB	LMB	SL	SP	SA	WS
1																	
.824**	1																
.772**	.959**	1															
.766**	.896**	.732**	1														
.936**	.849**	.794**	.792**	1													
.686**	.714**	.729**	.570**	.732**	1												
.362**	.427**	.460**	.304**	.403**	.485**	1											
.813**	.794**	.760**	.712**	.833**	.650**	.556**	1										
.892**	.817**	.768**	.762**	.922**	.776**	.388**	.768**	1									
.901**	.774**	.701**	.783**	.916**	.655**	.342**	.797**	.895**	1								
.126	.175*	.185*	.130	.087	.003	.121	.124	.113	.046	1							
.790**	.712**	.640**	.686**	.785**	.564**	.349**	.726**	.798**	.827**	.002	1						
.786**	.761**	.663**	.789**	.826**	.692**	.332**	.708**	.854**	.856**	.032	.777**	1					
.739**	.846**	.818**	.745**	.772**	.797**	.342**	.651**	.812**	.737**	.067	.588**	.795**	1				
-.078	-.123	-.160	-.070	-.055	-.013	-.075	-.025	-.058	.044	-.242**	-.012	.018	-.088	1			
-.074	-.112	-.135	-.068	-.058	.026	-.065	-.035	-.054	.053	-.273**	.000	.025	-.051	.945**	1		
-.100	-.145	-.168	-.084	-.088	.009	-.080	-.052	-.076	.013	-.276**	-.018	.004	-.074	.905**	.969**	**1**	
-.042	-.148	-.191*	-.057	.000	.021	-.093	.007	-.013	.076	-.306**	-.017	.047	-.089	.784**	.780**	.763**	**1**

* and ** Correlation is significant at the 0.05 and 0.01 level, respectively (2-tailed)

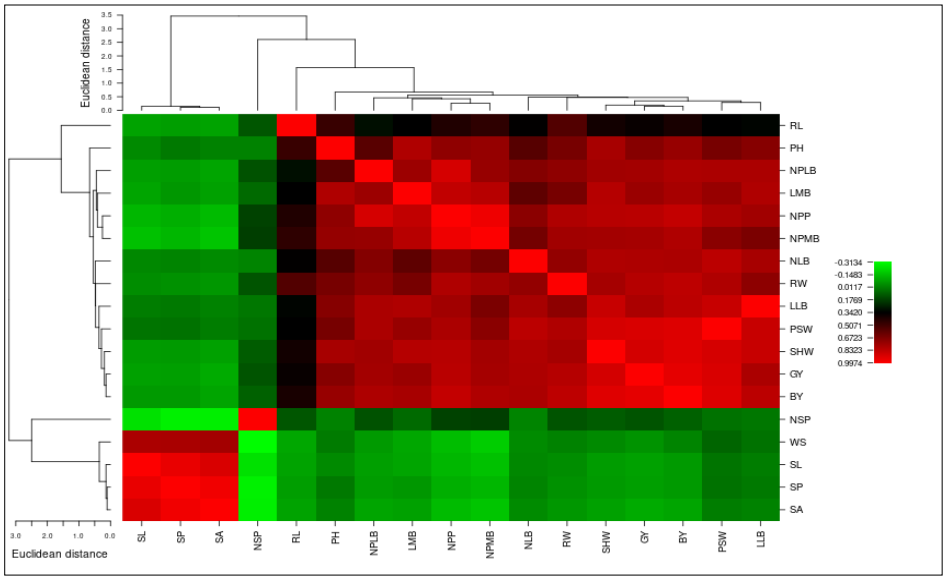

Fig. 4. Heatmap correlations and cluster analysis of the morphological and physiological characters of 136 camelina DH lines. The different colours (Green = negative correlations, Red = positive correlations).

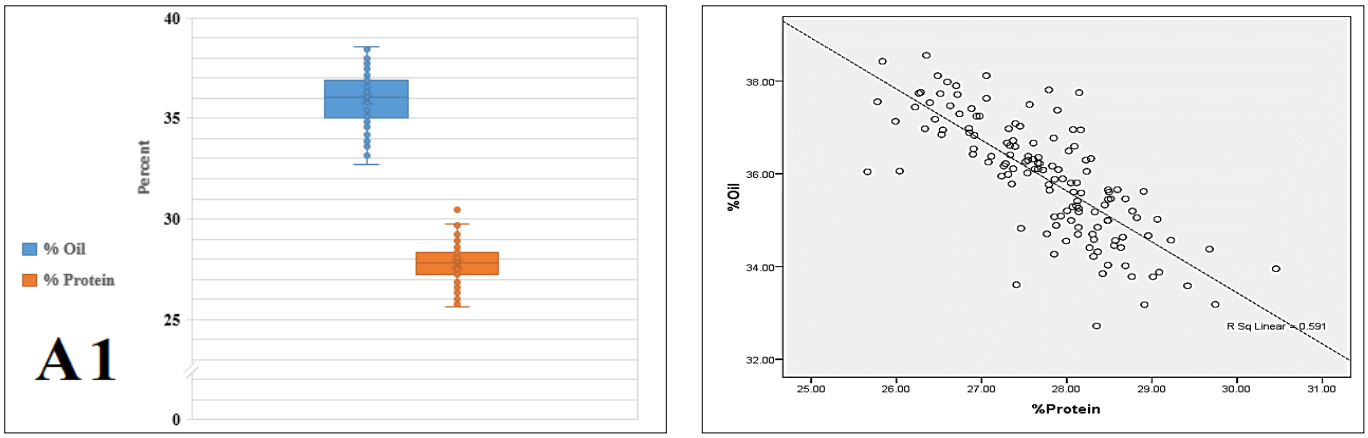

Fig. 5. A .The percentage of oil and protein in the seed oil of 136 camelina DHs. B. Pearson's linear correlation between oil and protein in the seed oil 136 camelina DHs.

ALA (12). LA and ALA are important because they are precursors of linoleic and linolenic acids, respectively. The analysis of linolenic acid content in the lines examined in this research revealed that the minimum and maximum values were 28.95 % and 34.90 %, corresponding to lines 83 and 65, respectively. The minimum and maximum levels of linoleic acid were 16.07 and 22.24, respectively, with the lowest and highest values recorded in lines 128 and 96. The quantity of linolenic acid can be considered a stability parameter for Camelina oil. Various researchers have conducted studies that reported similar percentages of linolenic and linoleic acids (Table 6).

Table 6. The content of linolenic and linoleic acids in Camelina oil based on previous reports (4)

Fatty acids	Content
Linolenic acid (C18:3)	30.5-50.3 %
	32.6 %
	35.6 %
	28.0-33.4 %
	32.8-33.0 %
	38.1 %
	35.2 %
Linoleic acid (C18:2)	16.6-19.3 %
	19.6 %
	20.9 %
	18.5-22.4 %
	18.3-18.5 %
	16.0 %
	16.9 %

The correlation between the fatty acid composition

It is crucial to inquire if there is a biochemical connection between the levels of fatty acids in Camelina oils. To answer this, one must determine if there is a correlation between the fatty acid levels (Table 7). Table 7 presents the correlation results of fatty acids in the oil of 136 doubled haploid Camelina lines. The linkage between the fatty acid composition contents of the Camelina oils studied can be discussed using this plot as a basis. The correlation between citric acid (C18:0) and other fatty acids was minimal except for fatty acid (C20:0-C22:0). The strongest correlation between fatty acids C18:0-C20:0 ($r = 0.833$, $P < 0.01$) and fatty acids C20:0-C22:0 ($r = 0.959$, $P < 0.01$) was observed. (Fig. 6).

Principal component analysis (PCA) - based biplot

All plant cells need lipids as an essential component. Fatty acids are mandatory structural components of almost all lipids, including triacylglycerols (storage and non-storage). There has been a description of more than 1000 different fatty acids (21). Plant seed oils contain a vast array of fatty acids. The grouping of a fatty acid into major, minor, or unusual fatty acids is arbitrary and should only be used for comparative studies (21). The simultaneous visualisation of

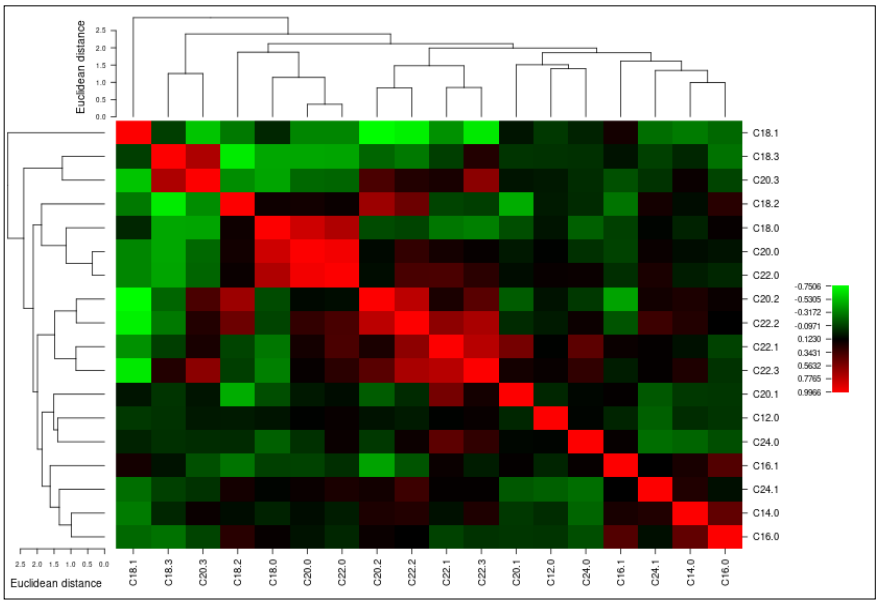

Fig. 6. Cluster analysis and heat map correlations were performed on the fatty acids of Camelina oil extracted from 136 Camelina DH lines. The different colors (Green = negative correlations, Red = positive correlations).

genotypes and variables can be achieved using a Principal Component Analysis (PCA) - based biplot. Fatty acids were divided into five groups. These groups had common fatty acids as well. This can be attributed to the common production pathways of fatty acids. The first category includes fatty acids (C22:2, C22:3, C18:1, C20:2, C22:1 C18:2, C20:0 C24:1 and C22:0), the second category (C22:3, C22:1, C22:0, C18:3, C18:0 C20:3, C18:2, C20:0 and C20:1), the third category (C20:2, C22:1, C22:0, C18:0, C18:2, C20:0, C20:1, C16:1 and C24:0), the fourth category (C14:0, C16:0, C16:1, C24:1 and C12:0) and the fifth category (C18:3, C18:0, C20:3, C18:2, C20:0 and C24:0) (Fig. 7).

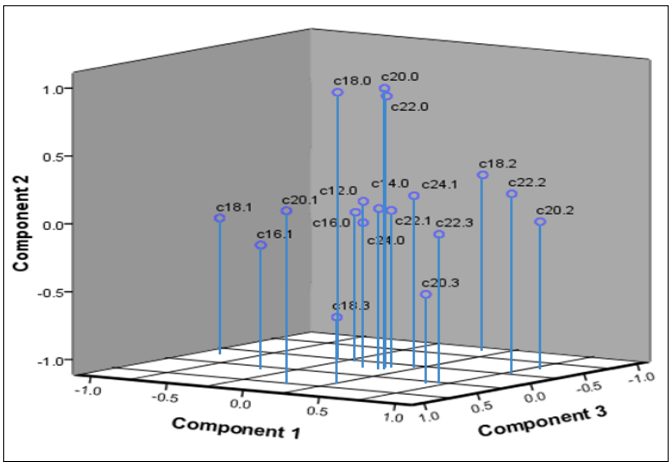

Fig. 7 .Three-dimensional scatter plots of the correlation between 18 fatty acids in 136 Camelina DHs.

Discussion

This study on the fatty acid composition of Camelina oil has identified eighteen distinct types of fatty acids. The reported values of fatty acids in Camelina oil vary across different studies, primarily due to factors such as growth environment, extraction methods and genetic variations. In the present research, cold press extraction was utilised to obtain the oil, a technique widely adopted in various industries for its efficiency and practicality. This method was chosen explicitly for oil extraction in this study. Additionally, due to their genetic purity, doubled haploid lines are an excellent tool for elucidating the potential of oil-related traits and other characteristics.

A breeding program can be designed more effectively by improving our understanding of the morphological traits associated with grain yield formation. This study's evaluation of doubled haploid lines in camelina reveals significant diversity, underscoring their breeding potential. This diversity enables the selection of desirable traits among the doubled haploid lines. Furthermore, doubled haploids offer distinct advantages in breeding and genetic programs, as their genotypes and genetic variance ratios are equivalent to those of gametes, making them a powerful tool for genetic improvement. According to the correlation matrix of morphological and physiological traits, the most significant correlations with grain yield (GY) were observed for NPP, BY, SHW and PSW traits. The number of pods per plant (NPP) showed a high correlation with the number of pods per main branch plant (NPMB) ($r=0.959$, $P<0.01$) and the number of pods per lateral branch plant (NPLB) ($r=0.896$, $P<0.01$). Notably, both NPMB and NPLB exhibited a strong positive correlation with the length of the main branch and the length of the lateral branch. These findings suggest that increasing the shoot length of camelina plants may lead to a higher number of pods and seeds per plant, thereby enhancing yield. Selecting a few key traits in plant breeding often leads to more successful outcomes (22). Therefore, targeting the length of both the primary and lateral branches could be an effective strategy to improve yield in camelina. This approach aligns with broader principles of crop improvement of 14 vegetable oils, including rapeseed oil (23). Their findings indicated limited correlations between specific fatty acids (C16:0-C18:0) and other fatty acids, underscoring the complexity of trait interactions in plant breeding. In the present study, Palmitic acid (C16:0) correlated poorly with different fatty acids. This finding aligns with the complex nature of Palmitic acid, which serves as a precursor for most fatty acids through modifications such as desaturation, elongation, hydroxylation and oxidation. These extensive

modifications make it challenging to accurately calculate the correlation of Palmitic acid (C16:0) with other fatty acids. Structural membrane glycerolipids in plant cells are predominantly composed of C-16 and C-18 fatty acids, further highlighting the significance of Palmitic acid in cellular processes. The concentration of Palmitic acid (C16:0) consistently decreases during the growth period of Camelina seeds, suggesting that this fatty acid undergoes multiple transformations (24). These transformations likely contribute to the study's low correlation between Palmitic acid and other fatty acids. ALA (C18:3) and LA (C18:2) are essential fatty acids within the PUFA category. This study measured the linolenic acid (C18:3) content across 137 lines, with minimum and maximum values of 28.95 and 34.90, respectively. Similarly, the linoleic acid (C18:2) content ranged from 16.07 to 22.24. While the stability of an oil depends on the overall composition of fatty acids rather than a single type, linolenic acid has been identified as a key factor in the stability of rapeseed oil. Consequently, the quantity of linolenic acid can serve as a reliable stability parameter for Camelina oil.

One of the key outcomes of this research is the concurrent analysis of physiological-morphological traits and fatty acids (as breeding targets) in camelina. This approach facilitates the selection of correlated characteristics and significantly improves the efficiency of breeding programs for this plant. Additionally, the composition and concentration of fatty acids in camelina seed oil were precisely identified and characterised. Paying special attention to fatty acids' amount, type and correlation is essential to Camelina production. Selecting lines for specific purposes can be made easier by investigating the correlation between fatty acids. The investigation and reporting of the relationship between 18 types of fatty acids in Camelina lines was carried out in this study. The minimum and maximum amounts of Camelina oil were calculated to be (32.72 - 38.55 %). Camelina oil had an estimated protein content of 25.66 % to 30.45 %. This research estimated the amount of oil and protein to have a negative correlation. The present study's conclusions on the correlation of oil and protein content (9, 25). The research observed a negative correlation between oil and protein content in Camelina seeds by examining six distinct genotypes. However, in the present study, this inverse relationship was confirmed with greater statistical confidence by analysing a significantly larger population of lines (136 lines) (26, 27).

Research indicates that only seven fatty acids of camelina's spring and autumn cultivars were identified and analysed (27). However, identifying a more significant number of fatty acids would have allowed for a more accurate evaluation of the acceptance or rejection of the research hypotheses. In the research that focused on the analysis of Camelina fatty acids and their potential applications in food products, the quantified levels of most fatty acids aligned closely with the values reported in the current study.

The results of reported Camelina genetic diversity will significantly impact future breeding programs. The different accessions were analysed and several fatty acids were found. This could potentially be useful for industrial applications in the future. *C. sativa* line with low levels of very long chains and polyunsaturated fatty acids were identified during the screening of the fatty acid composition of 136 *C. sativa* accession samples, Suggesting that their oil would have a greater ability to withstand thermo-oxidation and a lower solidification point (28). The observed diversity among the studied lines of our Camelina plants can influence classical breeding and the discovery of functional molecular markers for genotype selection. Based on this, it is recommended that the variety of traits and their correlation with the type and amount of fatty acid in camelina be investigated.

Conclusion

This study investigated the fatty acid composition of Camelina oil and identified eighteen distinct types of fatty acids. This research demonstrates significant diversity in the traits examined among doubled haploid lines of camelina, indicating that this broad range of variation can facilitate the design of targeted and multipurpose breeding programs for this plant. In addition to conventional uses, these findings can contribute to the improvement and recommendations for industrial applications of camelina. Significant correlations between morphological and physiological traits with grain yield and fatty acid composition provide opportunities for selecting desirable traits to enhance both yield and oil quality. The analysis of genetic diversity and trait correlations will pave the way for future research to improve yield alongside oil quality in this plant.

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Authors' contributions

FF, as the principal investigator, derived this article from their PhD thesis and was responsible for writing plant cultivation and data collection. DK served as the supervisor for this research, assisting and overseeing data analysis and the development of double haploid lines. AR collaborated on cultivation and selecting the type of statistical design. AZ was involved in the validation of double haploid lines. LZ assisted in writing and field data collection. HD supervised the extraction of fatty acids and selecting appropriate methods."

Compliance with ethical standards

Conflict of interest: Authors do not have any conflict of interest to declare.

Ethical issues: None

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