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Assessment of Spineless Safflower (*CarthamusTinctorius,* L.) Mutant Lines for Seed Oil Content and Fatty Acid Profiles.

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ABSTRACT

This study was conducted to assess the new spineless mutants that previously induced through gamma radiation and hybridization techniques in the advanced generation for seed oil content and fatty acid profiles

The obtained results cleared that oil percentages of all seven safflower mutants were increased than local variety Giza (1) and the new mutant hybrid 2 line (white petals) had the highest increase value of oil percentage (10%) but the mutant line M14 (dark red petals) had the lowest increase value of oil percentage (3.1 %)

The mutant line M7 (yellow petals) had the highest value of total saturated fatty acid (40.38%), because it had the highest value of palmitic fatty acid (25.16%), comparing to 10.01% value for local variety Giza (1), followed by mutant line hybrid 2 (white petals) which had (39.88%) because it had the highest value of caprylic, capric, lauric, myristic and stearic fatty acids.

All safflower mutant lines had higher value of oleic fatty acid than that of the local variety Giza (1) the two new safflower mutant lines M7 (yellow petals) and hybrid 2 (white petal) had the highest value of oleic fatty acid 41.22% and 39.88% respectively in comparison with 13.5% for local variety Giza (1), the obtained results are indicating to seed oil content negative correlation between oelic and linoleic acids.

Key words: safflower/ seed oil content/fatty acid profiles.

INTRODUCTION

Safflower (*Carthamus tinctorius* L.) is an ancient oil crop planted in Egypt. It was used as a row material for many industries in pharaonic era. Its oil was used in nutrition as well as in painting. The petals were used as a source of natural dye.

Safflower is a member of the compositae family and it was first cultivated in the Near East thousands of years ago as a source of dye and other products that can be derived from the plant [(1)].
In Egypt, safflower well adapted to Egypt conditions and it has a specific importance as winter oil crops, since oil crops were sown in summer and could not competed with the major crops like cotton, rise and maize. Moreover, the local safflower variety Giza (1) and land races were characterized with spineless which have been a major deterrent to he acceptance of the safflower crop. In a previous program for induced mutations by gamma radiation at Atomic Energy Authority some induced spineless mutants on safflower through irradiation and hybridization were selected and characterized with improved oil content and fatty acid profiles [(2)].

The original cultivars of safflower contained high levels of linoleic acid. These varieties also have the highest ratio of unsaturated to saturated fats compared to other oilseed crops. [1] In the 1950's, an oleic variant of safflower was first described by [(3)]. This variant was genetically described in 1963 by [(4)]. Subsequent breeding resulted in the release of UC1, an oleic variety of safflower [(5)]. Oleic safflower varieties were subsequently released commercially, and in recent years have come to dominate the safflower market. The release of oleic safflower types market the first time that one species was represented by varieties differing substantially in fatty acid type. Later discoveries accented the variability present in the safflower gene pool.

Safflower oil primarily comprises the fatty acids palmitic (C16:0), stearic (C18:0) as saturated fatty acids, oleic (C18:1), and linoleic acids (C18:2) as unsaturated fatty acids.

Numerous health studies have been conducted in recent years linking the types of fats consumed to health issues, especially cholesterol levels. It has been recognized that unsaturated fatty acids have superior health benefits compared to saturated fatty acids, especially those containing fewer than eighteen carbon atoms. Saturated fats with eighteen or more carbon atoms seem to have little or no effect on cholesterol levels [(6)].

Recent studies regarding fatty acids and health issues, especially heart disease, indicate an advantage of oleic acid over other vegetable oil fatty acids, or at least fewer disadvantages than such fatty acids. There have been conflicting results among studies, but two recent findings are noteworthy. It has been found that oleic acid is superior to linoleic acid regarding cholesterol level and levels of HDL [(7)]. Trans fatty acids, created through hydrogenization of unsaturated fatty acids, have the effect of elevating cholesterol levels.

From a health standpoint, a vegetable oil with increased oleic acid as well as decreased palmitic acid would be highly desirable. Since palmitic acid is the only saturated fat with fewer than eighteen carbon atoms found in safflower in significant amounts, a safflower variety with decreased palmitic acid and increased oleic acid would be highly desirable.

Higher oleic acid levels also have potential for industrial uses. Increases in oleic acid are highly correlated with lower levels of linoleic acid [(8)]. Low levels of linoleic acid correlate positively with oil stability [(9)]. Highly stable oils have uses in a number of markets. Manufacturers use oleic acid for infant formula and food supplements due to the greater shelf life of the oil. Oils with greater stability are being sought as an agent in spray-on flavor additives. If the oleic acid level could reach or exceed 90% additional markets would be available.

The objectives of this study are to assess the new seven spineless safflower mutant lines for seed oil content and fatty acid profiles in advanced generation.

MATERIALS AND METHODS

In a program designed for improving safflower through the use of gamma irradiation induced mutation, eleven mutants were selected from M_2 generation after seed irradiation of local variety Giza(1), with 10, 20, 30, 40, 50, and 60 Krad [(10)]. From this program some mutants with improved oil content and quality were selected [2].

To improve the spineless mutants, two induced mutant (Mut. 1 and Mut. 2) were crossed together and with the local variety Giza 1 as well as the exotic variety $A_{25}SK_1$ (introduced from Germany). Some mutant lines with improved oil content and fatty acid profiles were selected, seven of these mutants were characterized by stability in their character over 10 years, therefore these mutant lines were assessed in advanced generation for oil seed content and fatty acid composition compared to local variety Giza 1 in Principal Central Laboratory – faculty of Agriculture – Cairo University.

Complete block design with three replications was used. Five seed samples from each plot for each line were used for determination of seed oil content and for acid profiles the lipid extraction for each replication of each entry according to the method of [11], and separation of fatty acids according to [12].

Identification and determination of fatty acids were done by gas liquid chromatograph (GLC) with a method described by [13].
The data was statistically analyzed according to [14] .

RESULTS AND DISCUSSIONS

(1) Total seed oil content.

Total oil seed percentage (%) of new safflower mutant lines and local variety Giza (1) were showed in table (1). All seven mutant lines were increased than local variety Giza 1 (orange petals) by 5.8%, 4.7%, 7.9%, 5.8%, 10.0%, 6.3% and 3.1% for mutant lines M7 (orange petals), M7 (yellow petals), selected 96 (yellow petals), M14 (yellow petals), Hybrid 2 (white petals), Hybrid 9 (white petals) and M14 (Dark red petals), respectively.
The mutant lines Hybrid 2 (white petals) had the highest increase value of oil percentage (10%), but the mutant line M14 (dark red) had the lowest increase value of oil percentage (3.1 %). Similar results were obtained by [15, 16 and 17], they found an increase in seed oil for new lines of safflower after treating seeds with gamma rays and chemical mutagenesis, but [18] found that the differences between the oil content and number of capitulum.

Table (1): Total seed oil percentage (%) for new safflower mutant lines comparing with local variety Giza (1).

Trait / Mutant Lines	Oil %	% Increase over Giza (1)
Giza 1 (Orange)	**19.1**	**__**
M7 (Orange)	**20.2**	**5.8%**
M7 (Yellow)	**20.2**	**4.7%**
Selected 96 (Yellow)	**20.6**	**7.9%**
M14 (Yellow)	**20.2**	**5.8%**
Hybrid 2 (White)	**21.0**	**10.0%**
Hybrid 9 (White)	**20.3**	**6.3%**
M14 (Dark Red)	**19.7**	**3.1%**

L.S.D. 0.05 **0.7263 %**

(2) Fatty acid profiles.

2-1 saturated fatty acids.

Saturated fatty acids profiles of seed oil for safflower mutant lines comparing to local variety Giza (1) were shown in Table (2).

- Caprlyic fatty acid (C:8.0): the mutant line hybrid 2 (white petals) had the highest value of caprylic fatty acid 2.27% but the M7 (orange petals) had the lowest value of it (0.04%) compared to 0.21 for Giza (1).
- Capric fatty acid (C:10.0): the hybrid 2 (white petals) mutant line had the highest level of capric fatty acid 1.24% and the M14 (dark red petals) mutant line had the lowest level of it 0.20% compared to 0.05% for Giza (1).
- Lauric fatty acid (C:12.0): hybrid 2 (white petals) mutant line was given the biggest value of lauric fatty acid 1.29% but Lauric fatty acid was disappeared in both Giza (1) and M7 (orange petals).
- Myristic fatty acid (C:14.0): the highest value of myristic fatty acid was detected in hybrid 2 (white petals) mutant line 9.62%, and lowest value was founded in M14 (yellow petals) mutant line (0.10%) compared to 0.0% level in mutant line M7 (orange petals) indicating that M7 (orange petals) mutant line suggesting that the orange petals could be used as morphological for free lauric and myristic fatty acid.
- Palmitic fatty acid (C:16.0): it is important saturated fatty acid, it was increased to 18.42% and 25.16% in the hybrid 2 (white petals) and M7 (yellow petals) mutant line, respectively. But the selected 96 (yellow petals) had the lowest value of palmitic fatty acid (7.17%) compared to 7.67% for Giza (1).
- Stearic fatty acid (C:18.0): hybrid 2 (white petals) had the highest level of stearic fatty acid (6.67%) and hybrid 9 (white petals) had the lowest level of it comapred to 2.03% for Giza (1).

In general, M7 (yellow petals) had the highest value of total saturated fatty acid (40.38%), because it had the highest value of palmitic fatty acid (25.16%) comparing to value (10.01%) for local variety Giza (1) and the mutant line hybrid 2 (white petals) had the second highest value of total saturated fatty acid (39.51&), because it had the highest value of caprylic, capric, lauric, myristic and stearic fatty acids.

- Similar findings were obtained by [19] where several mutants with increased contents of stearic and [16] found significant positive change of composition of saturated fatty acid (palmitic and stiaric).

Table (2): Saturated fatty acid profiles as percentage of oil content for safflower mutant lines comparing to local variety Giza(1).

Lines / Fatty Acid	Giza 1 (Orange)	M7 (Orange)	M7 (Yellow)	Selected 96 (Yellow)	M14 (Yellow)	Hybrid 2 (White)	Hybrid 9 (White)	M14 (Dark Red)	L.S.D
Caprylic C:8.0	0.21	0.04	1.22	0.11	0.64	2.27	0.10	1.45	0.897
Capric C:10.0	0.05	0.97	0.98	0.41	0.43	1.24	0.89	0.20	0.199
Lauric C:12.0	---	----	0.67	0.19	0.60	1.29	0.09	0.16	0.139
Myristic C:14.0	0.05	----	4.60	0.12	0.10	9.62	0.11	0.48	0.742
Palmitic C:16.0	7.67	7.49	25.16	7.17	7.22	18.42	8.23	8.3	0.955
Stearic C:18.0	2.03	1.86	7.75	2.27	2.63	6.67	1.41	2.13	0.172
Total	10.1	10.83	40.38	10.27	11.62	39.51	10.83	12.72	----

2-2 Unsaturated fatty acids.

Unsaturated fatty acid profiles as percentage of oil seed for safflower mutant lines comparing to local variety Giza (1) were cleared in table (3).

2-2-1 Oleic fatty acid

The oil which contained high percentage of oleic fatty acid was considered useful for man's health and the best oil for industrial products.

All new safflower mutant lines except for M7 (orange) had high values of oleic fatty acid, these were increased to 41.22%, 39.88%, 15.17%, 15.60%, 14.09 and 14.06 for mutant lines M7 (yellow petals), hybrid 2 (white petals), selected 96 (yellow petals), M14 (yellow petals), hybrid 9 (white petals) and M14 (dark red petals) respectively, comparing to 13.50% for local variety Giza (1), and the increase percentages were 205.3%, 195.4%, 12.4%, 15.6%, 4.4% and 4.2% respectively.

The safflower mutant lines M7 (yellow petals) and hybrid 2 (white petals) had the highest value of oleic fatty acid, 41.22% and 39.88% respectively, but the new safflower mutant line M7 (orange petals) had the lowest value of oleic fatty acid, where the percentage of oleic fatty acid was decreased to 13.21% comparing to 13.50% for local variety, the percentage of decrease was 2.2%.

The results were agreed with the results which obtained by [19] and [17].

Table (3): Unsaturated fatty acid profiles as percentage of oil seeds for safflower mutant lines comparing to local variety Giza (1).

Fatty Acid / Lines	Oleic C:18.1	Linoleic C:18.2	Total
Giza 1(Orange)	**13.5**	**74.53**	**88.01**
M7 (Orange)	**13.21**	**75.57**	**88.87**
M7 (Yellow)	**41.22**	**17.17**	**58.39**
Selected(96Yellow)	**15.17**	**73.76**	**88.93**
M14(Yellow)	**15.60**	**72.12**	**87.72**
Hybrid 2(White)	**39.88**	**18.65**	**58.53**
Hybrid 9(White)	**14.09**	**74.2**	**88.29**
M14(Dark Red)	**14.06**	**72.42**	**86.48**
L.S.D	**0.602**	**0.613**	**----**

2-2-2 Linolenic fatty acid

The values of linoleic fatty acid were decreased by increasing oleic fatty acid for safflower mutant lines, the values of linoleic fatty acid were 17.17%, 73.76%, 72.12%, 18.65%, 74.2% and 72.42% for M7 (yellow petals), selected 96 (yellow petals), M14 (yellow petals), hybrid 2 (white petals), hybrid 9 (white petals) and M14 (dark red petals), respectively, comparing to value 74.53% for local variety Giza (1), but the value of linoleic of safflower new mutant line M7 (orange petals) was increased to 75.57% comparing to 74.53% for local variety Giza (1).

The safflower mutant line selected 96 (yellow petals) had the highest value of total unsaturated fatty acids (88.93%), but the new safflower mutant line M7 (yellow petals) had the lowest value (58.39%) of total unsaturated fatty acids, and the safflower mutant line hybrid 2 (white petals) had the second lowest value (58.53%) of total unsaturated fatty acids.

(17) found the same trend where the linoleic fatty acid was significantly increased after treating safflower seeds with gamma rays , [Ems], sodium acid and N-methyl-N-nitrosourea.

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