Development of Hybrids With Various Oil Quality

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Abstract

The objective behind this study was the introduction of genes for various oil quality into the parental lines of the hybrids NS-H-45, NS-H-626 and NS-H-680, which are resistance to *Phomopsis*.

Given the existence of isogenic lines possessing genes for various oil quality, three various oil qualities have been introduced into the parental lines of the hybrids NS-H-45, NS-H-626 and NS-H-680. The first version has Ol + tph_1, the second Ol + tph_2 and the third Ol + tph_1 tph_2 genes.

Due to the newly created isogenic lines, it is now possible to produce the three hybrids in at least three different versions with regard to oil quality. The new types of oil boast a significantly greater oxidative stability when compared to the common type. They also enable us to significantly widen the range of oil use in the food processing and other industries.

Key words: Sunflower oil, alfa, beta, gamma and delta tocopherols, oleic acid, oil oxidative stability

Introduction

The cultivated sunflower (*Helianthus annuus L.*) is one of the most important oil crops of the world. Although sunflower is primarily grown for the extraction of seed oil, there is nevertheless a limited production of non-oilseed types used for human confectionary or as a bird seed. (FRIEDT ET AL., 1994). The sunflower oil is generally considered a premium edible oil, because of its high linoleic and oleic acid content (C 18:2 and C 18:1, respectively). These two acids comprise over 90% of the fatty acid content of the sunflower oil, which, in addition, contains the following acids: palmitic (C 16:0), stearic (C 18:0), minor amounts of myristic (C 14:0), myristoleic (C 14:1), palmitoleic (C 16:1), arachidic (C 20:0), behenic (C 22:0), as well as some others in traces.

Great many authors have established the existence of variability in the composition and amount of fatty acids that is dependant on the genotype and environmental factors.

SOLDATOV (1976) made a significant contribution to a change of the C 18:1 composition and developed a high oleic sunflower genotype by means of induced mutation. Treatments of the variety VNIIMK8931 with dimethylsulfate (DMS) produced plants with more than 50% of oleic acid and further selections of 80-90% C 18:1 were used to derive the high oleic variety Pervenets.

The genetic basis and modes of inheritance of high oleic acid content in F_1 and F_2 generations were subjects of a number of studies (MILLER AND ZIMMERMANN, 1983; URIE, 1985; FERNANDEZ-MARTINEZ ET AL. 1989, etc).

The standard sunflower oil contains predominantly α tocopherol. Significant contribution to a change in sunflower oil tocopherol was made by DEMURIN(1993), who discovered the genes tph_1 (line L6-15), tph_2 (line L6-17) and tph_1 tph_2 (line L6-24). Lines containing these genes enable us to develop hybrids with various content of α, β, γ and δ tocopherols in oil.

The objective of this study was the introduction of the genes: a. Ol-genes + tph_1; b. Ol-genes + tph_2; and Ol-genes + tph_1 tph_2 into the parental lines of the hybrids resistant to *Phomopsis*: NS-H-45, NS-H-680 and NS-H-626 and subsequent development of hybrids with three new oil types based on the newly developed lines.

Materials and Methods

The research made use of donor lines for desirable genes created by VNIIMK, Krasnodar (Russia). In the B-lines, the carriers of donor genes were:

1. LG-21 (Ol + tph_1)
2. LG-25 (Ol + tph_2)
3. LG-24 (tph_1 tph_2)
4. VK-OL-373 (Ol)

In the restorer lines, the carriers of donor genes were the folowing lines:

1. VK-66-1 (tph_1 tph_2)
2. VK-66-2 (Ol + tph_1)
3. VK-66-3 (Ol + tph_2)

Parental lines from the hybrids resistant to *Phomopsis*: NS-H-45, NS-H-626 (Amelia) and NS-H-680 (Asturia) served as recepients of desirable genes.

In the summer of 1993, female lines from the hybrids: NS-H-45, NS-H-626 and NS-H-680, namely Ha-74, Ha-981 and CMS-3-8, were crossed in the field conditions with LG-21, LG-25, LG-24 and VK-OL-373. In the same year, furthermore, restorer lines from the hybrids NS-H-45 (RHA-SNRF-b), NS-H-626 and NS-H-680 (RHA-583) were crossed with VK-66-1, VK-66-2 and VK-66-3.

The plants used as females were on a daily basis subjected to a manual removal of stamens in the early morning hours. Throughout the vegetation period, pollen from the donor lines was applied with the brush and on a daily basis as well. During the autumn and winter of 1993 and 1994, F_1BC_{p1}, F_2 and F_3 combinations were produced in the greenhouse. In the course of 1994, new generations of breeding material were produced in the field. After the harvest, seeds of individual plants were analysed for tocopherol and fatty acid content.

Tocopherol composition was determined by means of thin-layer chromatography (TLC) followed by the Emmerie-Engel reaction and densitometer quantitation (POPOV AND ASPIOTIS, 1991) and fatty acid composition by gas chromatography (GC) of methyl esters.

In the autumn of 1994, a selection of plants with desirable traits was sown in the greenhouse in order to create a new generation. After the harvest, a new selection was made and the plants were sown again in the greenhouse in January, 1995. After a harvest at the end of April, selected plants were sown in the field, where there was yet another selection of plants. Subsequent to harvesting, these were analysed in the laboratory for tocopherols and fatty acid composition.

With regard to the restorer lines, the introduction of desirable genes can be considered over, whereas with the B-lines, it is almost complete - all that needs to be done is a translation into the CMS-form.

Results

Isogenic lines with different tocopherol and fatty acid composition of oil served as donors of certain traits introduced into the parental lines of the *Phomopsis*-resistant hybrids NS-H-45, NS-H-626 and NS-H-680. The objective was a simultaneous introduction of Ol-genes for high oleic acid oil content and those for various tocopherol types (tph_1; tph_2; tph_1 tph_2) into the parental lines of the above mentioned hybrids. Using certain breeding methods and laboratory analysis in the process of the introduction of genes, three isogenic lines with different quality of oil were created for each of the parental lines (Tables 1 and 2). Significantly, the lines were made to retain all of the other good traits as well, especially resistance to *Phomopsis*.

On the basis of the achieved results, it can be said that each parental line of the hybrids NS-H-45, NS-H-626 and NS-H-680 now exists in three new forms, namely with: a. Ol-genes + tph_1; b. Ol-genes + tph_2; c. Ol-genes + tph_1 tph_2 (Tables 1 and 2). The process of introducing various oil qualities into the hybrids' restorer lines has now been completeted. With regard to the female lines, nevertheless, the process is yet to be concluded, since the new isogenic lines with various oil quality still need to be translated into the CMS-form.

The newly created lines will enable us to produce the hybrids NS-H-45, NS-H-626 and NS-H-680 in at least three new types with regard to oil quality, thereby also enabling us to widen the range of sunflower oil use in the food processing, pharmaceutic, cosmetic and other industries.

It should be stressed that, in addition to the isogenic parental lines of the three hybrids, new lines with various oil quality which differ from the source lines with regard to agronomic and other traits have also been created. They could be used for the development of new hybrids.

Discussion

The results of DEMURIN ET AL. (1996) show that oil oxidative stability can be increased by increasing the content of β, γ and δ tocopherols simultaneously with oleic acid. The authors illustrate this by an example in which the presence of γ-tocopherols in oil and its high oleic acid content increased oxidative stability 16,4 times in comparison to the common sunflower oil.

Synergism in the effect of high oleic acid content and high β and γ tocopherols brings about a strong antioxidant activity, offering new possibilities in sunflower breeding for various oil quality. The realization of this fact was what prompted us to conduct these researches in the first place.

What needs to be addressed is the question of future users of the sunflower oil of various quality. Great possibilities undoubtedly exist, regardless of the fact that its application will certainly not be all that rapid.

One of the things that might present a problem is the possibility that not all of the Ol-genes for high oleic acid oil content are in the homozygous state. Futhermore, the exact number of the Ol-genes is not yet clear, although the literature offers various figures.

Conclusion

By means of appropriate breeding procedure and isogenic lines containing the genes Ol + tph_1, Ol + tph_2 and Ol + tph_1 and tph_2, parental lines of the hybrids NS-H-45, NS-H-626 and NS-H-680 were translated into isogenic lines with three different oli qualities. The first version of each of the parental lines contains the Ol + tph_1, the second one Ol + tph_2, and the third one Ol + tph_1 tph_2 genes. The newly created isogenic lines retained all the positive characters from before, especially resistance to *Phomopsis*. These lines enable us to produce the hybrids NS-H-45, NS-H-626 and NS-H-680 with at least three new types of oil.

Table 1: Composition of tocopherol and fatty acids content in isogenic lines

<table>
<tr><th rowspan="2">Genotype with genes</th><th colspan="4">Tocopherol composition (TLC)-%</th><th colspan="4">Fatty acid composition - %</th></tr>
<tr><th>α</th><th>β</th><th>γ</th><th>δ</th><th>16:0</th><th>18:0</th><th>18:1</th><th>18:2</th></tr>
<tr><td>Common sunflower seeds</td><td>95-100</td><td>0-3</td><td>0-2</td><td>-</td><td>6</td><td>4</td><td>30</td><td>60</td></tr>
<tr><td>Ol+tph1</td><td>50</td><td>50</td><td>-</td><td>-</td><td rowspan="3">4-6</td><td rowspan="3">3-4</td><td rowspan="3">85-87</td><td rowspan="3">6</td></tr>
<tr><td>Ol+tph2</td><td>0-5</td><td>-</td><td>95-100</td><td>-</td></tr>
<tr><td>Ol+tph1tph2</td><td>8-40</td><td>0-25</td><td>25-84</td><td>8-10</td></tr>
</table>

Table 2: Isogenic parental lines of hybrids NS-H-45, NS-H-626 and NS-H-680

<table>
<tr><th>Hybrid</th><th>Line</th><th>Genes</th></tr>
<tr><td rowspan="2">NS-H-45</td><td>1. Female line Ha - 74</td><td>Ol+tph1
Ol+tph2
Ol+tph1 tph2</td></tr>
<tr><td>2. Male line RHA-SNRF-b</td><td>Ol+tph1
Ol+tph2
Ol+tph1 tph2</td></tr>
<tr><td rowspan="2">NS-H-626</td><td>1. Female line Ha - 981</td><td>Ol+tph1
Ol+tph2
Ol+tph1 tph2</td></tr>
<tr><td>2. Male line RHA-583</td><td>Ol+tph1
Ol+tph2
Ol+tph1 tph2</td></tr>
<tr><td rowspan="2">NS-H-680</td><td>1. Female line CMS-3-8</td><td>Ol+tph1
Ol+tph2
Ol+tph1 tph2</td></tr>
<tr><td>2. Male line RHA-583</td><td>Ol+tph1
Ol+tph2
Ol+tph1 tph2</td></tr>
</table>

References

Demurin Ya, 1993 - Genetic variability of tocopherol Composition in sunflower seeds. HELIA, Vol.16. No. 18: 59-62

Demurin Ya., Škorić, D., Popov P., Efimenko S., Bochkovoy A.,1994 - Tocopherol genetics in sunflower breeding for oil quality. Proc. of EUCARPIA - Symposium on Breeding of Oil and Protein Crops. pp.193-197. Albena, Bulgaria. 22-24.09.1994.

Demurin Ya., Škorić D., Karlović Đ., 1996.- Genetic variability of tocopherol composition in sunflower seeds as a basis of breeding for improved oil quality (in Press)

Fernandez-Martinez J., Jimenez A., Dominguez J., Garcia J.M., Garces R. and Mancha M., 1989.- Genetic analysis of the high oleie acid content in cultivated sunflower-Euphytica 41: 39-51

Friedt W., Ganssmann M., Korell M., 1994 - Improvement of sunflower oil quality. Proc. of EUCARPIA - Symposium on Breeding of Oil and Protein Crops pp. 1-30. Albena, Bulgaria, 22-24. 09.1994.

Miller J.F. and Zimmermann D.C., 1983 - Inheritance of high oleie fatty acid content in sunflower. Proc. Sunflower Research Workshop, Fargo N.D. 26. January, National Sunflower Association, Bismark, N.D.

Popov P.S., Aspiotis E.H., 1991 - Biochemical methods for breeding material estimation (In Russien). Biology, breeding and growing of sunflower: 77-80. Agropromizdat, Moscow.

Urie A.L., 1985 - Inheritance of high oleic acid in sunflower. Crop. Sci. 24:1113-1115.