

HELIA, **30**, Nr. 47, p.p. 61-68, (2007)
DOI: 10.2298/HEL0747061B

UDC 633.854.78:631.53.011.026

CHANGES OF FATTY ACIDS CONTENT AND VIGOR OF SUNFLOWER SEED DURING NATURAL AGING

Balešević-Tubić, S.^{*}, Tatić, M., Miladinović, J. and Pucarević, M.

Institute of Field and Vegetable Crops, M. Gorkog 30, 21000 Novi Sad, Serbia

Received: November 10, 2006

Accepted: September 15, 2007

SUMMARY

Sunflower seed aging during storage affects seed vigor and content of fatty acids. In order to reveal severity of their influence, the following vigor tests were applied: standard laboratory germination test, cold test and Hiltner test. Five sunflower lines submitted to natural aging process for six and 12 months were tested under conventional storage and controlled conditions. The obtained results revealed that seed aging damaged the seed, which adversely affected seed vigor; most reliable results were obtained by the cold test and content of linoleic acids.

Key words: seed, sunflower, aging, vigor, fatty acids

INTRODUCTION

Viable seed is capable of producing new plant under both favorable and unfavorable climatic conditions. Emergence of the seed having high vigor is more uniform under field condition, and due to that it forms more vigorous plants which in turn provide higher yields (Milošević *et al.*, 1995). A large number of researchers use developmental stage of seedling as a tool for seed vigor estimation: length of the whole seedling, individual stem or root (McKersie and Tomes, 1982; Edwards and Sadler, 1992). Since individual tests do not satisfy all requirements for seed vigor estimation, several tests should be used (Milošević and Rajnpreht, 1993).

Vigor tests can be used as significant indicators of seed vigor during aging (Egli *et al.*, 1990; Miller and McDonald, 1994). Laboratory germination test is a good indicator of quality of seed stored under favorable conditions. However, it becomes less sensitive when storage conditions change to less favorable. In that case some of the tests for seed viability provide more reliable results (Elias and Copeland, 1994).

Since seed vigor reveals physiological changes the seed undergoes from swelling and beginning of germination to the full development of the seedling, and since it encompasses changes occurring in seed metabolism (Halmer and Bewley, 1984),

* Corresponding author: e-mail: ceca@ifvcns.ns.ac.yu

seed viability can be determined properly only by application of several vigor tests, and by combination and standardization of the results obtained by them.

Some reactions leading to lipid degradation in seed occur during seed storage (St. Angelo and Ory, 1983). It is understood that different composition of fatty acids means different susceptibility to peroxidation (Wilson and McDonald, 1986). In most of the plants species having seed rich in oil, lipids exposed to risk of autooxidation encompass oleic (18:1), linoleic (18:2) and linolenic (18:3) fatty acid chains. Degree of unsaturation has significant influence on degree of degradation (Priestley, 1986).

The aim of this investigation was to determine the degree and rate of seed deterioration during storage by submitting the seed to different storage conditions, which is significant from the aspect of maintaining the quality of seed.

MATERIALS AND METHODS

Experiment was performed with seed of five sunflower lines, ocms-74 (L1), cms-v-8931-3-4 (L2), ocms-22 (L3), ocms-98 (L4) and cms-ol-3 (L5), developed at Institute of Field and Vegetable Crops in Novi Sad. The seed was submitted to natural aging under conventional storage conditions (uncontrolled conditions) and under controlled conditions (at 4°C and relative humidity of 80 to 85%). Testing was done after six and 12 months of storage.

Standard laboratory test. Four replicates \times 100 seed of each line were tested. Wet blotter paper (roll type) was used as germination medium. Number of normal seedling was estimated after 10 days incubation period at 25°C and relative humidity of 95%.

Hiltner test. 4 \times 50 seeds were placed onto wet sand, and a 3 cm layer of cracked brick (previously sterilized and moistened with water) was placed upon them. Incubation period under optimal condition lasted for 10 days.

Cold test. 4 \times 50 seed were placed onto moistened soil (up to 40% of field capacity) at 5-8°C for seven days, and after that transferred to germination chamber at 25°C for four days.

Length of seedling stem was estimated in 10 normal seedlings.

Total oil content was determined using the spectroscopic method on an NMR – analyzer. The content of oleic (18:1), and linoleic (18:2) fatty acids in seed was determined by esterification using tri-methyl sulfonium hydroxide (TMSH) according to Bute (1983).

RESULTS AND DISCUSSION

Autooxidation of lipids during sunflower seed storage led to changes in fatty acid content of seed of the tested sunflower lines. No significant changes of total oil

content were observed in these lines during natural aging process (Table 1). Ferguson *et al.* (1990) found no noteworthy changes in total oil content in naturally aging soybean seed. However, other authors (Gidrol *et al.*, 1989; Vrbaški *et al.*, 1996) mention a decreased total lipid content in aged sunflower seeds. Intensive enzyme activities participating in lipid metabolism, especially increased seed moisture content, and increased storage temperature lead to usage of lipids in the process of respiration causing significant decreases in sunflower seed oil content (Beratlife and Iliescu, 1997).

Natural sunflower seed aging caused no significant changes in oleic acid content. Linoleic acid content was reduced after 12 months of storage. The intensive degradation of linoleic acid occurred in sunflower line L5, in which the linoleic acid content dropped from 23%, measured in fresh seed, to 5.49% after 12 months of storage. As reported by other authors, that degree of degradation is significantly influenced by degree of fatty acid unsaturation, and that oleic acid is more resistant to peroxidative changes (Priestley, 1986). Many authors (Priestley and Leopold, 1983; Hailstones and Smith, 1988) determined slight declines in linoleic and linolenic acid contents after natural aging of soybean seed, while Ferguson *et al.* (1990) determined no changes in the content of oleic, linoleic, and linolenic acids after 12 months of soybean seed storage.

Table 1: Total oil content, oleic (18:1) and linoleic (18:2) contents in sunflower seed depending on duration and way of natural aging

| Lines | Constituent (%) | Fresh seed | Conventional conditions | | Controlled conditions | |
|-------|-----------------|------------|-------------------------|-----------|-----------------------|-----------|
| | | | 6 months | 12 months | 6 months | 12 months |
| L1 | Total oil | 37.04 | 35.94 | 35.43 | 36.56 | 35.57 |
| | Oleic | 27.64 | 27.05 | 27.39 | 28.51 | 26.81 |
| | Linoleic | 61.23 | 61.13 | 60.97 | 61.31 | 60.64 |
| L2 | Total oil | 52.90 | 51.11 | 51.02 | 52.24 | 51.06 |
| | Oleic | 31.02 | 30.08 | 30.65 | 33.49 | 31.97 |
| | Linoleic | 57.05 | 56.82 | 54.71 | 56.60 | 54.87 |
| L3 | Total oil | 47.58 | 47.09 | 46.67 | 47.24 | 46.90 |
| | Oleic | 33.19 | 32.04 | 32.21 | 33.23 | 33.01 |
| | Linoleic | 56.16 | 56.03 | 54.54 | 56.11 | 54.82 |
| L4 | Total oil | 43.49 | 42.86 | 41.72 | 43.52 | 42.51 |
| | Oleic | 22.62 | 22.49 | 22.08 | 22.64 | 22.25 |
| | Linoleic | 66.23 | 65.41 | 63.61 | 65.18 | 63.30 |
| L5 | Total oil | 48.75 | 48.11 | 45.86 | 48.73 | 47.75 |
| | Oleic | 65.75 | 82.91 | 82.12 | 83.35 | 83.34 |
| | Linoleic | 23.02 | 6.59 | 5.49 | 7.67 | 6.74 |

During natural seed aging, decline in seed vigor was observed in all tested sunflower lines. Significant differences were observed in seed stored for 12 months in relation to the vigor of fresh seed. Significant decrease in vigor of aged seed was observed in line L5, while the lowest rate of seed germination was observed in line

L4 (Table 2). The influence of aging and differences in reaction of the tested lines were noticeable when cold test was applied. Highest decreases in germination were observed in seed of lines L2 and L5 which had higher oil contents than the other tested lines. Damage of seed and loss of vigor could easily occur due to high oil content of stored sunflower seed (Christensen, 1971; Beratlić and Iliescu, 1997). Unfavorable storage conditions can cause large variations in seed vigor (Milošević and Čirović, 1994). According to Powell (1988), seed damage during storage leads to decreased seed vigor.

Table 2: Vigor of sunflower seed (1-standard laboratory test, 2-cold test, 3-Hiltner test) depending on duration and method of seed storage (S-conventional conditions, C-controlled conditions)

| Test | Duration of storage | Germination (%) | | | | | Length of seedling stem (mm) | | | | |
|---------------------|---------------------|-----------------|-----|-----|-----|-----|------------------------------|-----|-----|-----|-----|
| | | L1 | L2 | L3 | L4 | L5 | L1 | L2 | L3 | L4 | L5 |
| 1 | Fresh seed | 97 | 85 | 87 | 92 | 89 | 141 | 138 | 122 | 148 | 125 |
| | 6 month S | 94 | 80 | 85 | 90 | 80 | 136 | 130 | 112 | 140 | 109 |
| | C | 94 | 83 | 86 | 91 | 81 | 140 | 131 | 113 | 138 | 110 |
| | 12 month S | 91 | 72 | 79 | 89 | 60 | 116 | 95 | 90 | 119 | 84 |
| | C | 91 | 75 | 83 | 90 | 69 | 129 | 103 | 93 | 106 | 84 |
| LSD _{0,05} | | 4,0 | 4,3 | 4,4 | 4,2 | 3,7 | 4,4 | 6,9 | 3,0 | 4,2 | 4,4 |
| 2 | Fresh seed | 94 | 82 | 85 | 90 | 79 | 84 | 62 | 51 | 71 | 80 |
| | 6 month S | 90 | 77 | 80 | 86 | 71 | 85 | 61 | 32 | 57 | 68 |
| | C | 91 | 79 | 82 | 86 | 70 | 83 | 63 | 39 | 62 | 58 |
| | 12 month S | 86 | 68 | 72 | 83 | 58 | 63 | 47 | 26 | 54 | 50 |
| | C | 89 | 70 | 78 | 85 | 63 | 65 | 50 | 29 | 56 | 53 |
| LSD _{0,05} | | 3,5 | 2,8 | 4,5 | 4,0 | 4,2 | 3,0 | 3,4 | 3,8 | 4,8 | 4,8 |
| 3 | Fresh seed | 95 | 79 | 78 | 91 | 79 | 141 | 131 | 100 | 141 | 123 |
| | 6 month S | 94 | 78 | 76 | 90 | 77 | 132 | 119 | 94 | 125 | 116 |
| | C | 94 | 79 | 79 | 91 | 76 | 133 | 125 | 101 | 127 | 118 |
| | 12 month S | 90 | 73 | 74 | 85 | 57 | 130 | 108 | 46 | 114 | 98 |
| | C | 90 | 76 | 76 | 87 | 69 | 136 | 115 | 77 | 115 | 105 |
| LSD _{0,05} | | 2,3 | 3,6 | 2,8 | 2,8 | 3,7 | 4,6 | 3,8 | 4,5 | 3,3 | 3,8 |

If results of individual vigor tests are compared, it can be seen that aged sunflower seed had the lowest germination value in the cold test. In the standard laboratory germination test, the seeds formed normal seedlings due to optimal humidity and temperature conditions during testing. However, because of a stress caused by the application of the cold test, the seed damaged during aging had lower vigor than fresh seed. The cold test was more reliable for estimation of aged seed vigor and reaction of seed under field conditions. This is in accordance with the results obtained by Trawatha *et al.* (1995), who noted that the cold test registered a 10% reduction of vigor in old seed compared with fresh seed.

Hiltner test exerts a physical stress on seed, predicting the capability of seed to germinate in crusted soil. Germination rate of aged sunflower seed was higher after

application of Hiltner test in relation to cold test, which was an indication that Hiltner test was not sufficiently reliable to reveal the decreased aged sunflower seed vigor as it was the case with cold test.

The viability values of aged seed of the tested sunflower lines obtained by the cold and Hiltner tests were in positive correlation with laboratory germination, with significant correlation coefficient obtained for lines L1 ($r=0.78$ cold test, $r=0.74$ Hiltner test), L2 ($r=0.90$ and $r=0.72$, respectively) and L5 ($r=0.98$ and $r=0.94$, respectively), confirming that vigor tests are reliable tools for predicting degree of seed damage during storage. Studying the reliability of vigor tests, Rajnpreht (1993) concluded that there was a high agreement between test results relative the possibility of prediction of seed behavior under field conditions, as indicated by correlation coefficient reaching even 0.98.

The capacity for seedling growth declined with sunflower seed storage duration. Differences in reaction of sunflower lines to seed aging were more pronounced in seedling growth than in seed germination. This was in agreement with conclusions of Anderson and Baker (1983) that, with prolonged storage, seed gradually loses germination capacity while seedling growth slows down. In this study, the lowest rate of seedling growth from old seed was obtained in the cold test. Largest reductions in seedling stem growth were registered in lines L3 and L5. Reduced seedling growth and increased number of abnormal seedlings developing from aged seeds were also observed by Roberts (1981) and other authors.

Initial plant growth is crucial for further plant development and survival under unfavorable environmental conditions. Quick formation of assimilation area and root system in an early stage of plant life gives these plants certain advantages during later stages of growth and development. The applied vigor tests showed that the stored sunflower seed suffered changes that adversely affected the growth of seedlings, especially under unfavorable conditions.

CONCLUSIONS

It was found that individual genotypes differed in their tolerance to changes occurring in seed during aging, revealing that degree of seed damage and capability of seed to resist negative aging consequences are influenced by duration and method of seed storage as well as other characteristics of sunflower seed.

A decline in seed vigor of naturally aged seed compared to the vigour of fresh seed was observed when vigor tests were applied. Cold test was the most reliable indicator of changes occurring in aged sunflower seed. The positive correlations obtained between the vigor tests indicated that all of them can be used for estimation of seed vigor in the tested sunflower lines.

REFERENCES

- Anderson, J.D. and Baker, J.E., 1983. Deterioration of seed during aging. *Phytopathology* 53: 321-325.
- Beratlić, C. and Iliescu, H., 1997. Highlights of proper sunflower seed storage. *Helia* 20(26): 121-137.
- Bute, J., 1983. Rapid derivatization of acids for GC by pre-column transesterification of glycerides. *J. Chromatography*.
- Christensen, C.M., 1971. Evaluating conditions and storability of sunflower seeds. *J. Stored Prod. Res.* 7(3): 163-169.
- Edwards, L.M. and Sadler, J.M., 1992. Growth vigor at some crop species and cultivars when fallseeded as winter cover in the Atlantic region of Canada. *Can. J. Plant Sci.* 72: 421-429.
- Egli, D.B., TeKrony, D.M. and Wiralage, R.A., 1990. Effect of soybean seed vigor and size on seedling growth. *J. of Seed Technol.* 14(1): 1-12.
- Elias, S.G. and Copeland, L.O., 1994. The effect of storage conditions on canola (*Brassica napus* L.) seed quality. *J. of Seed Technol.* 18: 21-22.
- Ferguson, J.M., TeKrony, D.M. and Egli, D.B., 1990. Changes during early soybean seed and axes deterioration: II. Lipids. *Crop Sci.* 30: 179-182.
- Gidrol, X., Serghini, H., Noubhani, A., Mocoquot, B. and Mazliak, P., 1989. Biochemical changes induced by accelerated aging in sunflower seeds. I. Lipid peroxidation and membrane damage. *Physiol. Plant.* 76: 591-597.
- Hailstones, M.D. and Smith, M.T., 1988. Lipid peroxidation in relation to declining vigor in seeds of soya (*Glycine max* L.) and cabbage (*Brassica oleracea* L.). *J. Plant Physiol.* 133: 452-456.
- Halmer, P. and Bewley, J.D., 1984. A physiological perspective on seed vigor testing. *Seed Sci. and Technol.* 12: 561-575.
- McKersie, B.D. and Tomes, D.T., 1982. A comparison of seed quality and seedling vigor in birdsfoot trefoil. *Crop. Sci.* 22: 1239-1241.
- Miller, B. and McDonald, J., 1994. Seed lot potential: viability, vigor and field performance. *Seed Sci. and Technol.* 22: 421-425.
- Milošević Mirjana i Rajnpreht Jarmila, 1993. Značaj setve deklarisanog semena pšenice za sortu i prinosa. *Zbornik radova Instituta za ratarstvo i povrtarstvo, Novi Sad*, 21: 343-350.
- Milošević Mirjana i Ćirović, M., 1994. Seme. *Institut za ratarstvo i povrtarstvo, Novi Sad*.
- Milošević Mirjana, Rajnpreht Jarmila, Graovac Milka i Zlokolica Marija, 1995. Uticaj starenja na osnovu pokazatelja životne sposobnosti semena važnijih ratarskih biljnih vrsta. *Agronomska saznanja br. 3*: 2-5.
- Priestley, D.A. and Leopold, A.C., 1983. Lipid changes during natural aging of soybean seeds. *Physiol. Plant.* 59: 467-470.
- Priestley, D.A., 1986. Seed aging implications for seed storage and persistence in the soil. *Comstock Publishing Associates, A division of Cornell University Press Ithaca and London*: 137-165.
- Powell, A.A., 1988. Seed vigor and field establishment. *Advances in Research and Technology of Seeds* 11: 29-61.
- Rajnpreht Jarmila, 1993. Pouzdanost metoda vigor testa na semenu pšenice proizvedenom u različitim ekološkim i agrotehničkim uslovima. *Doktorska disertacija, Univerzitet u Novom Sadu, Poljoprivredni fakultet, Novi Sad*.
- Roberts, E.H., 1981. Physiology of ageing and its application to drying and storage. *Seed Sci. and Technol.* 9: 359-372.
- St. Angelo, A.J. and Ory, R.L., 1983. Lipid degradation during seed deterioration. *Phytopathology* 53: 315-317.
- Trawatha, S.E., TeKrony, D.M. and Hildebrand, D.F., 1995. Relationship on soybean quality to fatty acid and C₆-aldehyde levels during storage. *Crop Sci.* 35: 1415-1422.
- Vrbaški, Ž., Budinčević, M., Turkulov, J., Škorić, D. and Vranac, K., 1996. Oxidation stability of sunflower oil of altered sunflower after seed storage. *Helia* 19(24): 73-78.
- Wilson, D.O. and McDonald, M.B., 1986. The lipid peroxidation model of seed ageing. *Seed Sci. and Technol.* 10: 269-300.

CAMBIOS DE CONTENIDO DE ÁCIDOS GRASOS Y GERMINACIÓN DURANTE EL ENVEJECIMIENTO NATURAL DE LA SEMILLA DE GIRASOL

RESUMEN

El envejecimiento de la semilla de girasol durante el almacenamiento, influye en la energía de germinación y el contenido de ácidos grasos en la semilla. Para determinar la intensidad de su influencia, fueron investigadas las siguientes pruebas de la energía de germinación: la prueba de germinación de laboratorio, estándar, prueba fría y la prueba de Hiltner. Cinco líneas de girasol, expuestas al envejecimiento natural durante seis y 12 meses, fueron investigadas en las condiciones de almacenamiento convencional y almacenamiento en las condiciones controladas. Los datos obtenidos demuestran que el envejecimiento de la semilla causó deterioros que tuvieron influencia negativa en la energía de germinación. Los resultados más ciertos fueron obtenidos sobre la base de la prueba fría y el contenido del ácido linólico.

CHANGEMENTS DU CONTENU D'ACIDES GRAS ET DE LA VIGUEUR DE LA GRAINE DE TOURNESOL AU COURS DU VIEILLISSEMENT NATUREL

RÉSUMÉ

Le vieillissement de la graine de tournesol durant l'entreposage affecte la vigueur de la graine et le contenu d'acides gras. Les tests suivants ont été appliqués dans le but d'établir l'importance de leur influence: test de faculté germinative standard en laboratoire, test au froid et test Hiltner. Cinq lignées de tournesol soumises au vieillissement naturel au cours de six et douze mois ont été étudiées dans des conditions d'entreposage conventionnel et d'entreposage contrôlé. Les données obtenues montrent que le vieillissement de la graine avait provoqué des dommages qui affectaient négativement la vigueur de la germination. Les résultats les plus fiables ont été obtenus par le test au froid et celui du contenu d'acides linoléiques.