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Investigations of the Stability of Vegetable Oils

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In Latvia rapeseed oil is the mostly investigated vegetable oil. There is a line of plants besides rape, from which seeds and fruits it is possible to obtain oils for various needs, for example, quince, oil radish, hemp, coriander, caraway, garlic, pumpkin, hazel nut, flax, poppy, plum kernel, red clover, grape kernel, hawthorn haw.

The aim of work – to determine oxidative stability of vegetable oils (obtained in Latvia) and to suggest effective antioxidant additives.

Investigation of oxidative stability of linseed, hempseed, colza and rapeseed oils has been carried out by using antioxidants BHT (2.6-ditertbutyl-4-methylphenol), ascorbylpalmitate and ascorbic acid as synergist. Antioxidants are added to oils 0.02 % m/m.

It has been discovered that 0.02 % BHT increases oxidative stability of linseed oil by 1.3 times, rapeseed oil – by 1.8 times; 0.02 % ascorbylpalmitate increases oxidative stability of colza oil by 1.8 times, linseed oil - by 2.3 times, hempseed oil - by 1.8 times; 0.02 % ascorbic acid increases oxidative stability of rapeseed oil by 4 times, colza oil – by 1.5 times, linseed oil – by 2.9 times, hempseed oil – by 1.5 times.

Vegetable oils, antioxidants, BHT, ascorbylpalmitate.

Introduction

Vegetable oils are source of valuable biological active compounds. Vegetable oils primarily contain tocopherol (*E* vitamin) as the natural antioxidant. Oil of wheat sprout is rich with tocopherols 520 mg/100 g, which also exists in other oils: sunflower 62.5, maize 33.7, grape 31.9, hazel nut 26.3, rapeseed 22.8, soybean 17.0, olive 12.8, peanut 10.3, lineseed 5.8 mg/100 g (Zariņš et al., 1999). Vegetable oils contain mono- and polyunsaturated fatty acids. The dietary requirement for lipids can be satisfied entirely by consumption of sufficient amounts of linoleic and linolenic acids. Both fatty acids are 18 carbon polyunsaturated fatty acids differing in the number and position of the double bonds. Linoleic acid has two double bounds with placement on the first double bond at the sixth carbon numbering from the omega carbon, which is the carbon on the opposite end of the carboxylic acid in the fatty acid chain. Linoleic acid has three double bonds with the first double bond on the third carbon from the omega carbon. The placement of the double bonds relative to the omega carbon is significant because it determines the capacity for endogenous synthesis of the fatty acid. Linoleic and linolenic fatty acids cannot be synthesized in human organism, while the oleic acid can be synthesized endogenously. Linoleic and linolenic acids are essential fatty acids in human nutrition.

Since vegetable oils contain many unsaturated acids, those oxidize during storage by the process of autooxidation. Oxidative degradation of vegetable oils may be initiated by various agents, such as light, temperature, ferments, traces of metals (Fe, CO, Cu, Mn), oxygen. As lipis oxidize, they form hydroperoxides, which are susceptible to further oxidation decomposition to secondary reaction products such as alcohols, aldehydes, ketones and acids. In many cases, these compounds adversely affect flavor, taste, nutritional value and averall quality (Angelo, 1992).

Autooxidation can not be checked easily since it is a chemical reaction with very low activation energy, estimated at 4-5 kcal/mole and 6-14 kcal/mole for the two reactions. The rates of autooxidation of methyl oleate, methyl linoleate, methyl linolenate are 1 : 12 : 25, so vegetable oils rich in polyunsaturated fatty acids are more exposed to autooxidation than the vegetable oils rich in saturated or unsaturated fatty acids. When the autooxidation of oil is followed experimentally by determining the peroxide value of the oil, it is found that the course of oxidation defines two distinct phases. During the initial phase, oxidation proceeds at a relatively slow and more or less uniform rate. Then, a certain critical amount of oxidation occures, the reaction enters the second phase, characterized by a rapidly accelerating rate of oxidation and by an eventual rate that is many times greater than that observed in the initial phase (Svobodnyje..., 1979).

The autooxidation may be prevented by keeping oils in the closed vessels or inert atmosphere, or by adding antioxidants. Antioxidants are effective only in case they are added before autooxidation has started (Romano, 1982; Privett et al., 1962; Emanuel et al., 1961).

Action of antioxidants is desribed in this manner:

$$RH \xrightarrow{\text{initiator}} R\cdot$$

radical reacted with oxygen:

$$R\cdot + O_2 \longrightarrow RO_2\cdot$$

competing reaction in chain process:

$$RO_2\cdot + RH \longrightarrow R\cdot + ROOH$$

This reaction is delayed by antioxidants (AOH):

$$RO_2\cdot + AOH \longrightarrow AO\cdot + ROOH$$

Antioxidant inhibits reaction while it is being completely consumed up (Romano, 1982; Privett, 1962).

Important and well-known antioxidants are BHA (2-or 3-tert-butyl-4-methoxyphenol) and butylhydroxytoluene, BHT (2,6-di-tert-butyl-4-methylphenol), vitamin C (ascorbic acid), 6-o-palmitoyl-L-ascorbic acid (ascorbylpalmitate) and vitamin E (tocopherol), which are legally per-

mitted food additives (Warner et al., 1989; Kolb et al., 2002; Sedlaček, 1975; Cort, 1974). Antioxidants found in nature such as tocopherols, L-ascorbic acid, citric acid, tartaric acid and its salts are acting also as synergists, preventing oxidation of oils (Emanuel et al., Warner et al., 1989; Kolb et al., 2002; Sedlaček, 1975; Cort, 1974; 1961; Melentjeva et al., 1995).

Ascorbic acid has synergistic effect on tocopherols. Synergists are aliphatic polyhydroxyacids, ascorbic acid, sodium ascorbinat, ascorbylpamitate. The mentioned compounds may be simultaneously synergists and antioxidants (Melentjeva et al., 1995).

It has been found out that exchange of H-atom between ascorbic acid and tocopherol radicals, which are arising in process of oxydation of fats with air oxygen, occurs. It is supposed, that oxydized fats at first are utilized ascorbic acid, then tocopherol. These results explain functions and action of synergism mechanism of ascorbic acid and tocopherols in the inhibition of fats oxydation (Melentjeva et al., 1995). Of various combination of antioxidants mixture of ascorbic acid, tocopherol and citric acid is the most effective ones. Ascorbic acid and high fatty acid 6-o-esters are frequently used as antioxidants for oils. Since mechanism of oxydation reduction of ascorbic acid and its 6-o-esters, probably, is identical, esters have synergists properties and they can be used together with tocopherols. The following ascorbic acid esters are used: stearat, palmitat, layrat, miristat, linoleat (Cort, 1974).

In recent time more attention has been paid to natural antioxidants. Various spices, aromatic and medicinal herbs as well as other plants can accumulate significant amounts of strong antioxidative compounds (Nguyen et al., 2000; Bandoniene et al., 2000; Schmidt et al., 2003; Niklova et al., 2001; Miskunova et al., 2001; Nguyen et al., 2001; Kozlova, 2003; Suja et al., 2004; Kumazawa et al., 2004; Zainol et al., 2003; Nuutila et al., 2003; Kim et al., 2003). So, evening primrose containes phenolic acids and their esters, gallic acid, methyl- and ethylgallates, protocatecheric acid and its methylesters. Evening primrose seed antioxidant activity has been tested for sunflower and rapeseed oils (Schmidt et al., 2003; Niklova et al., 2001).

Copper oleat exhibits good antioxidative properties as an additive in rapeseed oil and it has the advantage of lover volatility compared with synthetic phenolic antioxidants at higher temperatures (Huaiwen et al., 2003).

Recently papers about investigation of antioxidants in various vegetable oils have appeared. A chemiluminescence method has been used to study the effectiveness of the antioxidants of linseed oil in quenching peroxy radicals. Antioxidant efficacy decreases with the duration of the oil storage time. Components with potential antioxidant activity (tocopherols, carotenoids, ubiquinone and phosphatidylcholine) have been quantified in the fresh oil (Prozorovskaja et al., 2003). The antioxidative properties of sunflower, rapeseed and grapeseed oils have been evaluated. Rapeseed oil has the highest content of antioxidants, stability and total radical-trapping antioxidative potential (Ciz et al., 2002). The lipid soluble antiradical material contains 4-vinyl-2,6-dimetoxyphenol as antioxidant is extracted from the rapeseed oil (Shigeru et al., 2003).

Methods of reseach

Seed oils of rape (Brassica napus), colza (Brassica rapa), line (Linum usitatissimum) and hemp (Cannabis sativa), grown in Latvia, were used in experients. Oils were obtained by cold pressing, at the temperature of 40 °C and filtrated. The role of antioxidants BHT, ascorbylpalmitate and ascorbic acid, that preserve oxidative stability of these vegetable oils, was investigated.

Oxidative deterioration was realized in a thermostat at 60 °C. Antioxidants BHT and ascorbylpalmitate were dissolved in 100 ml of rapeseed (lineseed, colza, hempseed) oil at the concentration of 0.02 % (m/m), antioxidants were added directly to the oil and mixed for 10 min at 80 °C. Ascorbic acid was dissolved in distilled water (0.02 g ascorbic acid in 0.08 ml water) and was added directly to the oil and mixed for 10 min at 60 °C.

The samples were placed in open 50 ml volume 4 beakers (25 ml). A control sample was prepared under the same conditions, without addition of any antioxidant. All samples were placed in thermostat. These samples were used for determining the peroxide value and acid value.

For the determination of peroxide value (Matiseks et al., 1998) approximately 1 g of the sample of oil was weighed to within 0.1 mg, into a 250 ml flask, 10 ml chloroform and 10 ml acetic acid were added, immediately followed by 0,5 ml of saturated potassium iodide solution. The sample was shaken and put in dark for 3 minutes and then 100 ml of distilled water were added. The liberated iodine was titrated with 0,01 N sodium thiosulfate solution by shaking, and using starch as indicator.

For the determination of acid value approximately 2 g of the sample of oil was weighed, to within 0.1 mg, into a 250 ml flask. Sample was dissolved in 100 ml of previously neutralized solvent diethyl ether and 95 % ethanol (1 : 1). The mixture was titrated by swirling with the potassium hydroxide 0,1 M solution to the endpoint when the addition of a single drop generates a slight, but definite colour change persisting for at least 15 s.

Fatty acid content in oils was determined as follows: oil glycerides were transesterificated to fatty acid methylesters, solved in heptan and analyzed by method of gas-liquid chromatography. Chromatograph CHROM-5 with flame ionization detector and column 0.3 mm x 1.5, filled with 10 % DEGA/chrom WAW 60/80 was used (carrier gas – nitrogen, pressure 0.85 atm, temperature in thermostat 210 °C (Niewiadomski, 1990)).

Results and discussion

Oxidative stability of rapeseed, colza, linseed and hempseed oils with has been investigated by means of accelerate method (60 °C temperature). BHT, ascorbylpalmitate, ascorbic acid (0,02 % mass) have been used for stabilization of vegetable oils.

Process of oxidation has been controlled according to peroxide value. Peroxide value depends on chemical composition of oils.

Rapeseed and colza oils are similar by content of high fatty acids. They contain ~ 30 % polyunsaturated (linoleic- and linolenic-) acids, while lineseed oil - ~ 70 %, and hempseed oil - 78,2 % this acids (Table).

Table. Fatty acid content in vegetable oils
Lentelė. *Riebalų rūgščių kiekis*

N° *Nr.*	Vegetable oil *Augalinis aliejus*	Fatty acid content in oil, % *Riebalų rūgščių kiekis %*				
		Palmitic acid *Palmitino rūgštis* $C_{16:0}$	Stearic acid *Stearino rūgštis* $C_{18:0}$	Oleic acid *Oleino rūgštis* $C_{18:1}$	Linoleic acid *Linolo rūgštis* $C_{18:2}$	Linolenic acid *Linoleno rūgštis* $C_{18:3}$
1.	Hempseed *Kanapių sėklų*	6,1	2,0	12,1	57,7	20,5
2.	Pumpkin seed *Moliūgų sėklų*	9,5	0,1	7,8	82,6	-
3.	Oil pumpkin seed *Aliejingųjų moliūgų sėklų*	10,2	6,0	49,3	34,5	-
4.	Hazel nut *Lazdynų riešutų*	4,0	0,9	87,6	7,5	-
5.	Lineseed: *Linų sėmenų:*					
	stock *pluoštinių*	4,4	0,5	24,9	15,5	54,7
	oil *aliejinių*	5,6	0,5	26,3	12,5	55,1
6.	Poppy seed *Aguonų sėklų*	11,1	1,2	20,0	65,7	2,0
7.	Rapeseed *Rapsų sėklų*	3,5	2,8	66,6	20,2	6,9
8.	Rye sprout *Rugių daigelių*	17,0	1,4	21,2	33,4	27,0
9.	Red clover *Raudonųjų dobilų*	11,1	0,5	27,6	57,7	3,2
10.	Grapeseed *Vynuogių sėklų*	5,9	0,3	16,0	76,9	0,5
11.	Colza *Kolza (rapsų)*	4,5	2,3	62,6	22,0	8,7

Lineseed oil contains mainly linolenic acid (~ 55 %), and it must be more unstable. However, oxydation process depends not only on content of unsaturated fatty acids of oil, but also on presence of natural antioxidants and other compounds of oils. According to the results of the experiments, tested oils by their oxidative stability can be arranged as follows: colza oil > lineseed oil > rapeseed oil > hempseed oil.

Addition of antioxidants BHT and ascorbic acid to rapeseed oil increases oxidative stability 1,7 and 4,0 times, accordingly (Fig.1).

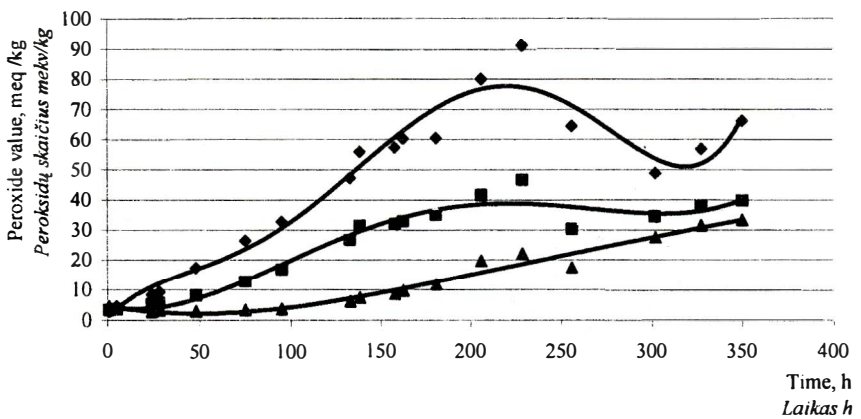

Fig. 1. Oxidative stabilization of rapeseed oil at 60 °C: ♦ - control, ■ – 0.02 % BHT, ▲- 0.02 % ascorbic acid
1 pav. *Rapsų aliejaus stabilizavimas 60 °C: ♦ - kontrolė, ■ –0.02 % BHT, ▲- 0.02 % askorbo rūgštis*

Addition of antioxidants BHT, ascorbylpalmitate and ascorbic acid to colza oil increases oxidative stability 1.2; 1.8 and 1.5 times, accordingly (Fig. 2). As colza oil is more stable, its oxidative stability is insingnificantly affected by antioxidants.

Addition of antioxidants BHT, ascorbylpalmitate and ascorbic acid to lineseed oil increases oxidative stability 1.3, 2.3 and 2.9 times, accordingly (Fig. 3).

Addition of antioxidants BHT, ascorbylpalmitate and ascorbic acid to hempseed oil increases oxidative stability 1.2, 1.8 and 1.5 times, accordingly (Fig. 4.). It has been found out, that the antioxidants, used for preserving oxidative stability of oils, may be arranged in the following line: ascorbylpalmitate ≥ ascorbic acid > BHT.

Determination of acid value can also give interpretation of the mechanism of oil oxidation. At this moment definite conclusions about acid value changes can not be made (Fig. 5, 6).

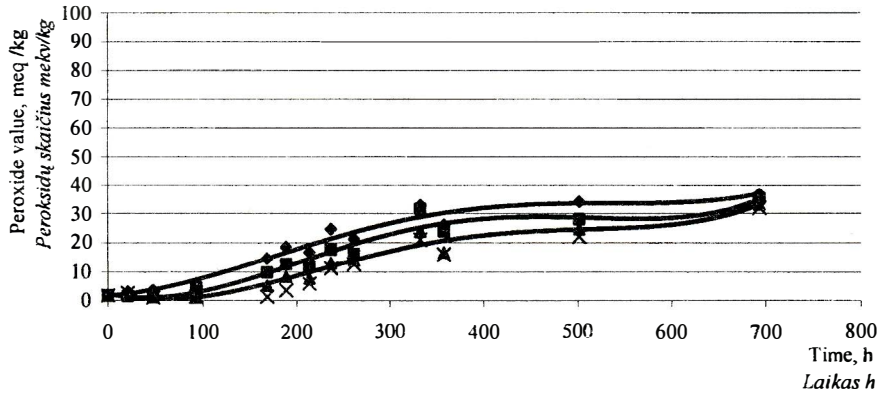

Fig. 2. Oxidative stabilization of colza oil at 60 °C: ♦ - control, ■ – 0.02 % BHT, ▲- 0.02 % ascorbic acid, × - 0.02 % ascorbylpalmitate

***2 pav.** Rapsiukų aliejaus stabilizavimas 60 °C: ♦ - kontrolė, ■ – 0.02 % BHT, ▲- 0.02 % askorbo rūgštis, × - 0.02 % askorbilo palmitatas*

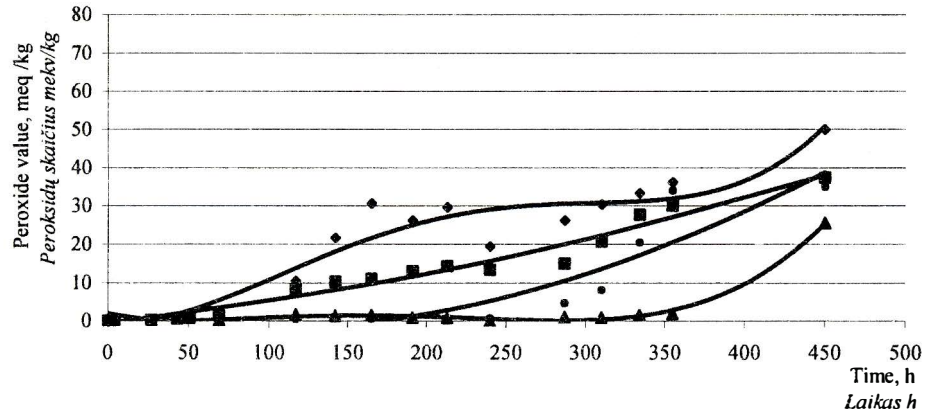

Fig. 3. Oxidative stabilization of linseed oil at 60 °C: ♦ - control, ■ – 0.02 % BHT, ▲- 0.02 % ascorbic acid, • - 0.02 % ascorbylpalmitate

***3 pav.** Linų sėmenų aliejaus stabilizavimas 60 °C: ♦ - kontrolė, ■ – 0.02 % BHT, ▲- 0.02 % askorbo rūgštis, • - 0.02 % askorbilo palmitatas*

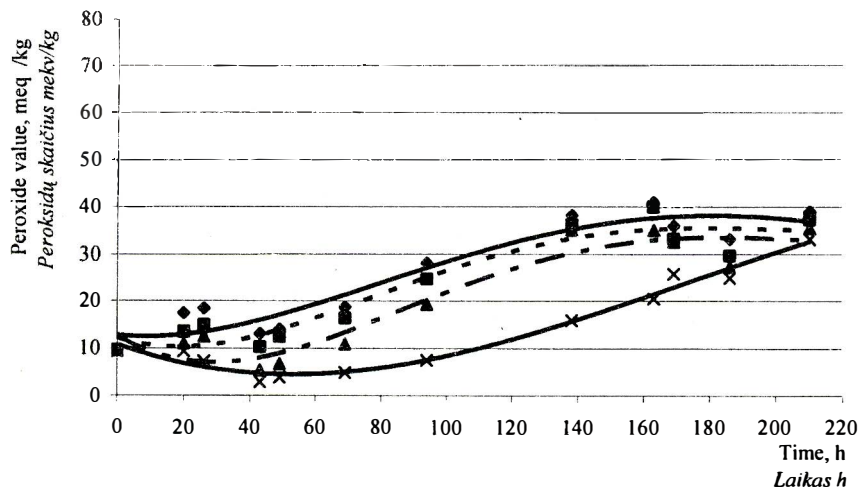

Fig. 4. Oxidative stabilization of hempseed oil at 60 °C: ♦ - control, ■ – 0.02 % BHT, ▲- 0.02 % ascorbic acid, × - 0.02 % ascorbylpalmitate

***4 pav.** Kanapių sėklų aliejaus stabilizavimas 60 °C: ♦ - kontrolė, ■ – 0.02 % BHT, ▲- 0.02 % askorbo rūgštis, × - 0.02 % askorbilo palmitatas*

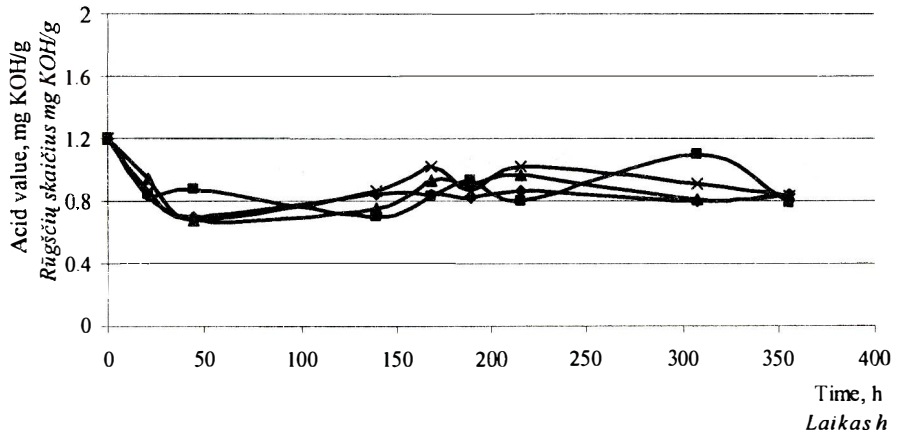

Fig. 5. Changes of acid value at oxidative stabilization of rapeseed oil at 60 °C: ♦ - control, ■ – 0.02 % BHT, ▲- 0.02 % ascorbic acid, × - 0.01 % ascorbylpalmitate

***5 pav.** Rūgščių skaičiaus pokyčiai stabilizuojant rapsų aliejų 60 °C: ♦ - kontrolė, ■ – 0.02 % BHT, ▲- 0.02 % askorbo rūgštis, × - 0.01 % askorbilo palmitatas*

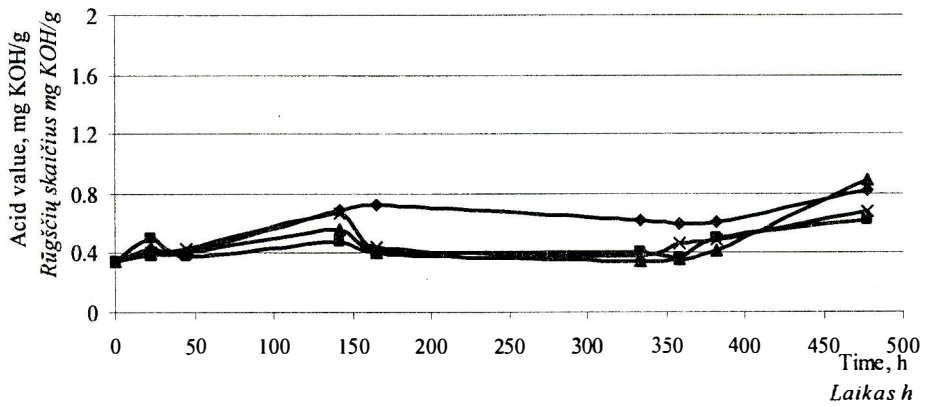

Fig. 6. Changes of acid value at oxidative stabilization of linseed oil at 60 °C: ♦ - control, ■ – 0.02 % BHT, ▲- 0.02 % ascorbic acid, × - 0.01 % ascorbylpalmitate

***5 pav.** Rūgščių skaičiaus pokyčiai stabilizuojant linų sėmenų aliejų 60 °C: ♦ - kontrolė, ■ – 0.02 % BHT, ▲- 0.02 % askorbo rūgštis, × - 0.01 % askorbilo palmitatas*

Conclusions

1. By their oxidative stability vegetable oils may be arranged in the following line: colza > lineseed oil > rapeseed oil hempseed oil.

2. The best antioxidants for the investigated oils are ascorbylpalmitate and ascorbic acid.

3. 0.02 % (mass %) ascorbylpalmitate increases oxidative stability for colza oil 1.8 times, for linseed oil – 2.3 times, for hempseed oil – 1.8 times.

4. 0.02 % (mass %) ascorbic acid increases oxidative stability for rapeseed oil 4 times, for colza oil – 1.5 times, for linseed oil – 2.9 times, for hempseed oil – 1.5 times.

5. Acid value of oils changes insignificantly in the conditions of the experiment.

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Augalinio aliejaus stabilumo tyrimai

Santrauka

Rapsų aliejus yra labiausiai ištirtas augalinis aliejus Latvijoje. Be rapsų, yra daug augalų (svarainiai, aliejinis ridikas, kanapės, kalendra, kmynai, česnakai, moliūgai, lazdynai, linai, aguonos, slyvų sėklos, raudonieji dobilai, vynuogių sėklos, gudobelės vaisiai), iš kurių sėklų ar vaisių galima išgauti aliejų, skirtą įvairiausioms reikmėms.

Darbo tikslas – įvertinti Latvijoje pagaminto augalinio aliejaus atsparumą oksidacijai ir siūlyti efektyvius antioksidantus jai didinti.

Linų sėmenų, kanapių, rapsų ir rapsiukų aliejų atsparumas oksidacijai buvo tirtas naudojant antioksidantus: BHT (2,6-ditertbutilo 1-4 metilfenolį), askorbilo palmitatą ir askorbo rūgštį kaip sinergentą. Antioksidantų buvo dedama 0,02 % aliejaus masės.

Nustatyta, kad naudojant 0,02 % BHT priedą, sėmenų aliejaus atsparumas oksidacijai padidėja 1,3 karto, rapsų aliejaus – 1,8 karto. 0,02 % askorbilo palmitato priedas padidina rapsų aliejaus atsparumą oksidacijai 1,8 karto, sėmenų aliejaus 2,3 karto, kanapių aliejaus 1,8 karto. Naudojant 0,02 % askorbo rūgšties priedą, rapsų aliejaus atsparumas oksidacijai padidėja 4 kartus, rapsiukų aliejaus – 1,5 karto, sėmenų aliejaus- 2,9 karto, o kanapių aliejaus 1,5 karto.

Augalinis aliejus, antioksidantai, BHT, askorbilo palmitatas.

Gauta 2004 m. birželio mėn., atiduota spaudai 2005 m. kovo mėn.

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