

## The antioxidant effect of *Melissa officinalis* extract regarding the sunflower oil used in food thermal applications

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### Abstract

The aim of this paper was to study the effect of lemon balm (*Melissa officinalis*) compared with butylated hydroxytoluene regarding the retarding lipid oxidation of sunflower oil. Refined sunflower oil free of additives and supplemented by three concentration levels of lemon balm extract (200, 600, 1000 ppm) and one level of butylated hydroxytoluene (200 ppm) were subjected to convection heating for 1, 4, 8, 12, and 16 hours. To track the progress of lipid oxidation the following analyses were made: peroxid value (PV), p-anisidine value (p-AV) and TOTOX value. The measurements for the antioxidant characteristics of lemon balm are the FRAP value and Total phenolics value. According to the survey result that the lemon balm extract (LBE) shows a significantly inhibitory effect on lipid oxidation during heat treatment. Lemon balm extract in dose of 200 ppm inhibited the lipid oxidation in a similar manner to BHT and in dose of 600 ppm and 1000 ppm resulted an significant decrease of investigated indices. According to the results oxidative stability of sunflower oil can be improved using lemon balm natural extract.

**Keywords:** antioxidant; convective heating; extract; lemon balm; oxidative stability; sunflower oil.

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### 1. Introduction

Sunflower oil is one of the most common and widespread types of oils used in humans daily diet and also in food industry. The oil is high in vitamin E, vitamin K, essential linoleic acid and low in saturated fat. Sunflower oil is a vegetable triglyceride oil produced from sunflower seeds and contains 60-75% linoleic acid. Different types of sunflower oil were developed such as high oleic, mid oleic and high linoleic [1, 2].

The oxidative stability of sunflower oil it is affected by several factors such as storage conditions (temperature, light, time) but the main deterioration process occurs during thermal processing.

To protect against oxidation synthetic antioxidants are added for improving oxidative stability of vegetable oils.

This addition of synthetic antioxidants is not recommended because of their toxicity and carcinogenicity instead finding natural antioxidants sources is encouraged and the interest for these increased [3].

Lemon balm (*Melissa officinalis*) it is a herb, member of the Lamiaceae family and it is used as a culinary flavouring often in combination with other herbs such as spearmint. In medicinal field lemon balm has many uses such as mild sedative or calming agent, it can improve mental performance, it is claimed to have antiviral and antibacterial properties against herpes simplex [4] and that lemon balm extract has an exceptionally high antioxidant activity [5].

Lemon balm it is high in flavonoids and can provide antioxidant activity.

Other phytochemicals which can have an antioxidant effect from lemon balm are: rosmarinic acid, phenolic acids, caffeic acids and terpenes [6]. It has been reported that lemon balm has possesses strong antioxidant activity and lemon balm extract can improve the oxidative stability of sunflower oil [7, 8].

## 2. Material and method

**Materials.** The sun flower oil without addition of any antioxidants used was “Tip” oil.

Lemon balm extract (LBE) was obtained from dried leaves originated from Romania (Fares). All other chemicals and solvents used were purchased from Merck (Darmstadt, Germany) and were of analytical grade.

Rotary evaporator-Heidolph Laborata 4000  
Convection oven-Esmach, Italy, 1200W  
Spectrophotometer-Analytic Jena Specord 205

**Antioxidant Activity (FRAP Assay).** The antioxidant activity of lemon balm extract was measured using the ferric reducing antioxidant power (FRAP) assay [9]. In order to evaluate antioxidant activity, 0,1 g LBE, respectively BHT were mixed with 20 mL ethanol/water (70:30, v/v) for 10 min, then the solution was filtered and used for analysis. FRAP values were obtained reading the absorbance changes at 595 nm using a UV-VIS spectrophotometer. Results were expressed as mM Fe<sup>3+</sup> equivalents/g lemon balm extract, respectively BHT.

**Total Phenols Assay.** Total phenolic content of lemon balm was determined using the Folin-Ciocalteu colorimetric method [10]. A calibration curve using gallic acid was prepared and the absorbance of the standards and samples were measured at 750 nm. Results were expressed as mM gallic acid equivalents/g.

**Peroxid Value (PV).** Weigh 2.00 g oil sample into a 250 mL Erlenmeyer flask and then were added 10 mL of chloroform and 15 mL of glacial acetic acid. The flask was swirled until the sample was dissolved and then was added 1 mL of saturated potassium iodine (KI) solution. The flask was closed and let for 1 min in dark and after 5 more minutes to stand. 75 mL of distilled water was added and then in presence of starch was slowly titrate with 0.01 N sodium thiosulfate (Na<sub>2</sub>S<sub>2</sub>O<sub>3</sub>) [8].

**The p-anisidine value (p-AV).** The p-anisidine value is a measurement of carbonyl content in the oils and was determined by the standard method according to AOCS [11].

It is based on the reactivity of the aldehyde carbonyl bond on the p-anisidine amine group, leading to the formation of a Schiff base that absorbs at 350 nm.

2 g of the sunflower oil samples were dissolved in 25 mL isooctane and absorbance (A<sub>1</sub>) of this fat solution was measured at 350 nm against a blank of isooctane. An aliquot (5 mL) of this solution, respectively 5 mL of isooctane was transferred to each of two test tubes of 10 mL and 1 mL of anisidine solution (0.25% g/v glacial acetic acid) was added to each. After 10 min the absorbance (A<sub>2</sub>) was measured at 350 nm against isooctane containing p-anisidine. p-AV was calculated according to the formula:

$$p-AV = 25 \times 1.2 \times A_2 - A_1 / w$$

**TOTOX value:** The total oxidation value (TOTOX) was used to estimate the oxidative deterioration of lipids. Totox value is defined as the sum of both values (PV and p-AV) to total oxidation and was calculated according to the formula:

$$TOTOX \text{ value} = 2 \times PV + p-AV$$

## 3. Results and discussion

Antioxidant characteristics of lemon balm extract (LBE) and butylated hydroxytoluene (BHT) are presented in table 1.

**Table 1.** Antioxidant characteristics of LBE and BHT

Sample	FRAP value (mM Fe <sup>3+</sup> /g)	Total phenolics (mM GAE/g)
Lemon balm extract (LBE)	45.06	743.6
BHT	1.36	-

Peroxid value was used as indicator for the primary oxidation of sunflower oil. The primary products of lipid oxidation are hydroperoxides and determination of peroxides can be used as oxidation index for the early stages of lipid oxidation. The results of PV are presented in table 2.

According to the results we can state that LBE in a dose of 200 ppm behave similarly to BHT. In a dose of 600 ppm and 1000 ppm lemon balm extract shows better results regarding the peroxid value of sunflower oil than the synthetic antioxidant.

p-AV value is a measurement of the secondary oxidation products (alcohols, ketones, aliphatic aldehydes and acids). Results regarding p-AV value are presented in table 3.

The results obtained following the p-AV determination confirm that lemon balm extract in doses of 200 behave similiary to BHT and in a dose of 600 and 1000 ppm reduced the lipid oxidation better than BHT.

**Table 2.** Effect of LBE and BHT on peroxid value during sunflower heating in convection oven

Time (hours)	Control	BHT 200 ppm	LBE 200 ppm	LBE 600 ppm	LBE 1000 ppm
1	4.64	4.06	3.53	3.28	3.05
4	7.98	7.88	7.01	6.63	5.98
8	12.13	11.92	11.13	11.00	10.55
12	18.65	18.12	17.59	17.15	16.31
16	19.79	19.43	18.70	18.04	17.56

**Table 3.** Effect of LBE and BHT on p-AV during sunflower oil heating in convection oven

Time (hours)	Control	BHT 200 ppm	LBE 200 ppm	LBE 600 ppm	LBE 1000 ppm
1	17.25	15.32	14.53	13.84	13.22
4	28.14	25.98	24.43	23.57	22.86
8	43.44	42.64	40.61	39.23	38.04
12	56.12	53.76	51.42	50.10	49.31
16	68.16	66.36	65.38	64.11	63.55

#### 4. Conclusion

The present research was carried out in refined sunflower oil free of additives, supplemented by three concentration levels of lemon balm extract (200, 600, 1000 ppm) and one level of BHT (200 ppm). The samples thermal treated in convective oven for 1, 4, 8, 12 and 16 hours.

According to the results it can be observed that because of thermal treatments oxidation of sunflower oil increased. Supplementation with lemon balm extract and BHT reduced the lipid oxidation resulting significant differences. The inhibitory effect of lemon balm against primary oxidation of lipids was concentration-dependent in chase of all three levels concentration (200, 600 and 1000 ppm).

Significant diferences were also observed at p-AV results and it can be seen that phenolic compounds from lemon balm had a strong inhibitory effect on the secondary lipid oxidation. The efficiency of lemon balm extract regarding oxidative stability of sunflower oil during thermal applications increased with increasing concentration of the natural extract and prove that lemon balm extract is a very efficient inhibitor of lipid oxidation.

#### Compliance with Ethics Requirements

Authors declare that they respect the journal's ethics requirements. Authors declare that they have no conflict of interest and all procedures involving human and/or animal subjects (if exists) respect the specific regulations and standards.

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