Tomas Bata University in Zlín
Library

Oxidative stability of crude oils relative to tocol content from eight oat cultivars: Comparing the Schaal oven and Rancimat tests

Citation

POKHREL, Kshitiz, Lenka KOUŘIMSKÁ, Ondřej RUDOLF, and Servenaz Khalili TILAMI. Oxidative stability of crude oils relative to tocol content from eight oat cultivars: Comparing the Schaal oven and Rancimat tests. *Journal of Food Composition and Analysis* [online]. vol. 126, Academic Press, 2024, [cit. 2025-07-07]. ISSN 0889-1575. Available at https://www.sciencedirect.com/science/article/pii/S0889157523007925

DOI

https://doi.org/10.1016/j.jfca.2023.105918

Permanent link

https://publikace.k.utb.cz/handle/10563/1011837

This document is the Accepted Manuscipt version of the article that can be shared via institutional repository.

TBU Publications
Repository of TBU Publications
publikace.k.utb.cz

Oxidative stability of crude oils relative to tocol content from eight oat cultivars: Comparing the Schaal oven and Rancimat tests

Kshitiz Pokhrel[a,*], Lenka Kouřimská[a], Ondřej Rudolf[b], Sarvenaz Khalili Tilami[a]

[a]*Department of Microbiology, Nutrition and Dietetics, Czech University of Life Sciences Prague, Kamýcká 129, 165 00, Praha 6, Suchdol, Czech Republic*

[b]*Department of Fat, Surfactant and Cosmetics Technology, Tomas Bata University in Zlín, Vavrečkova, Czech Republic*

[*]*Corresponding author. E-mail address: Pokhrel@af.czu.cz (K. Pokhrel).*

ABSTRACT

Oats are a good source of oil containing polyunsaturated fatty acids and natural antioxidants such as tocols. This study investigated tocol composition relative to oxidative stability in crude oils from eight oat cultivars. Tocol content in the cultivars was as follows: Santini > Saul > Korok > Atego > Kamil > Patrik > Oliver > Marco Polo. Santini had the highest tocol content (155.3 mg/kg of oil), whereas Marco Polo contained only 62.0 mg/kg of oil. Extracted oat oils were subjected to the Schaal oven test at 60 $\pm$ 2 °C and the Rancimat test at 100 °C and 110 °C to determine oxidative stability. The induction period of oat samples ranged from 51 to 56 days in the Schaal test, with a protection factor of 7.8-8.6. The Rancimat test yielded induction periods ranging from 3.78 h to 9.31 h at 100 °C and 1.11 h to 4.25 h at 110 °C. Both tests revealed remarkable stability against oxidation. Therefore, oat oil, particularly from the Santini cultivar, could be a valuable and stable source of natural antioxidants for the food and pharmaceutical industries. However, a significant correlation between Schaal-derived induction period and tocopherols or tocotrienols was not identified. This outcome indicates that oil stability is due to more than tocol presence, with other antioxidants in the crude oil likely playing important roles. In contrast, the Rancimat test revealed that (3-tocotrienol content was correlated with induction period at both temperatures 100 ◦ C and 110 °C.

Keywords: Oxidative stability, rancimat test, schaal oven test, tocopherols, tocotrienols, avena sativa

1. Introduction

Oats (*Avena sativa L.*) are currently receiving global attention because of their unique composition and nutritional properties, with the food industry increasing the use of oats as a raw material (**Sang and Chu, 2017**). Oats contain more crude oil (**Chen et al., 2016**) and lipids (up to 18%) (**Banas et al., 2007**) than other cereal grains. While the fat content is still at lower levels than traditional oilseeds (e.g., sunflower or rape-seed), oats remain an appealing source of edible oils. However, a potential

disadvantage is that their high concentration of unsaturated fatty acids increases the risk of oxidation and the formation of free radicals, which are both harmful and generate unpleasant flavors.

Oat accounts for 2.9-9.51 g/100 g of lipid content (**Pokhrel et al., 2022**) and it contains 23% saturated fatty acids (mainly palmitic acid), 34% monounsaturated fatty acids (mainly oleic acid), and 43% polyunsaturated fatty acids (mainly linoleic acid) (**Kouřimská et al., 2021**). This composition has important implications for the flavor of oat products, as it also affects food oxidation, where unsaturated fatty acids content is particularly influential. Lipid reactions in oat products can result in bitter, astringent, or rancid flavors (**Molteberg et al., 1996; Peterson, 2001; Viscidi et al., 2004; Jaksics et al., 2023**). Food processing and storage can worsen this problem through lowering the effectiveness natural antioxidants in oats. For instance, peeling and grinding increases susceptibility to oxidation, while heat treatment destroys antioxidants. Unprocessed oats undergo slow hydrolysis and oxidation because of their low enzyme activity and strong antioxidants. Nevertheless, nonenzymatic oxidation can still occur. Storing oats stably requires balancing between enzyme inactivation and antioxidant preservation.

Oats are a rich source of antioxidants in free and bound forms. Natural antioxidants are primarily concentrated in the outer kernel layers and include tocols, phytic acid, phenolic compounds, and avenanthramides (**Peterson, 2001**). Also contributing to the crop's antioxidant properties which are flavonoids and sterols. Unlike other cereals, oat seeds contain up to 30 avenanthramides (Hernandez-Hernandez et al., 2021), secondary metabolites with antioxidant activity that contribute to fresh taste and protect against rancidity (**Molteberg et al., 1996**). Another key group is phenolic compounds, comprising one aromatic ring with an acidic group and one or more hydroxyl groups. Their antioxidant and anti-inflammatory effects give rise to numerous health benefits. Thus far, phenols identified and quantified in oat extracts include ferulic acid, p-coumaric acid, caffeic acid, vanillic acid, p-hydroxybenzoic acid, 4-hydroxyphenylacetic acid, vanillin, and catechol (**Xing and White, 1997; Banaś and Harasym, 2021**).

Total phenolic content in oats was significantly correlated with antioxidant activity (**Emmons et al., 1999**). Of particular note are to-copherols and tocotrienols, also known collectively as tocols or E-vitamers. Tocotrienols exhibit stronger antioxidant properties than tocopherols, and both have been detected in oat grains (**Gangopadhyay et al., 2015**). Together, a-tocopherol and a-tocotrienol account for 90% of all tocols present in oats (Bryngelsson et al., 2002). The Rancimat and Schaal oven tests are common methods used to determine oxidative stability, including anisidine, peroxide, and acid values (**Maszewska et al., 2018**). The Schaal oven test, also known as the accelerated oxidation method, is particularly convenient for evaluating oil stability and is frequently employed in the food industry for evaluating the lipid quality of cookies (Abasolo, 2021a). The Rancimat test is widely used to evaluate bakery and cosmetic products (**Abasolo, 2021b**).

Despite the availability of data on oat antioxidants and their properties, limited information is available regarding the stability of oils extracted from different oat varieties. Therefore, the principal objective of this study was to examine and compare the oxidative stability of crude oils obtained from eight different oat cultivars using two tests, namely the Rancimat and Schaal oven tests. Additionally, the study aimed to assess the tocopherol and tocotrienol contents within the same oats.

2. Materials and methods

2.1. Chemicals and reagents

Polyethylene glycol 3000 (PEG) was obtained from Sigma-Aldrich (Merck KGaA, Darmstadt, Germany). Petroleum ether 40-60 °C (PET), tert-butyl methyl ether ($tBME$), and n-heptane (HiPerSolv Chromanorm) of analytical purity and High-Performance Liquid Chromatography ($HPLC$) grade were obtained from VWR Chemicals (BDH Israel). Tocols (tocopherols and tocotrienols) were purchased from ChromaDex (Los Angeles, California, USA).

2.2. Plant material and fat extraction

Six naked (NO) (Kamil, Oliver, Patrik, Marco Polo, Santini, and Saul) and two hulled (HO) (Atego, Korok) oat varieties were obtained from the Selgen a.s. breeding station in Stupice, Czech Republic (GPS 50°313" N, 14°384" E, 287 m.a.s.l.). The information regarding the studied cultivars can be found in the Database of Plant Varieties/ Czech National List (accessible at **https://eagri.cz/public/app/sok/odrudyNouQF.do?la ng=en_US**). These oats are grown for food purposes and future use in breeding.

Oat samples were ground using a Scarlett Silver Line SL 1545 coffee grinder (Ariette-Scarlett, Firenze, Italy) for 3 min. Approximately 10 g of sample was placed into Cytiva Whatman grade high-performance cellulose extraction thimbles (Sweden) and covered with cotton and approximately 85 mL of petroleum ether, along with boiling chips, were added. Crude oat lipids were obtained using the Rendall hot extraction method (E4 Behrotest, Labor-Technik, Dusseldorf, Germany) at approximately 70 °C to avoid oxidation. Extraction lasted for 1 h, and for the next 40 min, most of the PET was evaporated. The remaining PET was removed using a rotary vacuum evaporator (Heidolph Hei-VAP Core HL G3, Heidolph Instruments GmbH). Approximately 100 g of raw oat oil was required to extract 5 g of purified oat oil.

2.3. Tocol analysis

The crude oat oil extracted via Rendall's hot extraction was used for stability testing (Rancimat and Schaal oven tests). To prevent potential oxidation during the analysis of tocol content, the extracted oat oil was obtained differently under room temperature (<27 °C) conditions. Ground oat samples were mixed with petroleum ether. Later, filtration was carried out to separate the oats, then petroleum ether was removed through vacuum evaporation at an operating temperature of 35 °C. Extracted oil (1 g) was weighed and placed into a 10 mL volumetric flask that was then filled with heptane. Tocopherols and tocotrienols (tocols) were analysed using a high-performance liquid chromatograph ($HPLC$) equipped with a fluorescence detector (FLD, G7121A). The detector was set at 298 nm for excitation and 330 nm for emission. Tocols were separated using a 4 × 250 mm id, 5 |rm particle size, Agilent LiChrospher DIOL column (Eschenstr. 582024; Taufkirchen, Germany), maintained at 35 °C. The mobile phase was a mixture of $tBME$ and n-heptane (5:95, v/v) with a flow rate of 1 mL/min. Analytical conditions followed previous reports (Aligent, 2010). Tocol standards (mixture of α, β, γ, and δ isomers) were dissolved in n-hexane for peak identification and quantification with six-point calibration curves per tocol. Respectively, the correlation coefficients of the calibration curves (≥ 0.998), Limit of detection (LOD) (μg/mL), and Limit of quantitation (LOQ) (μg/mL) of standard solutions were as follows: α-tocopherol (0.9982, 6.36, 19.29), β-tocopherol (0.9980, 2.20, 6.67), γ-tocopherol (0.9976, 3.46, 10.50), α-tocotrienol (0.9956, 2.35, 7.12), β-tocotrienol (0.9983, 1.47, 4.44),

and δ-tocotrienol (0.9947, 5.14, 15.58). Chromatogram of tocols of the analysed oat sample and the standards are in **Fig. 1**.

2.4. Schaal oven test

Extracted oat oil was subjected to the Schaal oven test along with refined rapeseed (K-classic, Czech Republic) and sunflower (GIANA, K-classic, Czech Republic) oils. Pork lard was used as the base for all samples because gravimetric detection of increased oxygen absorption (induction period) requires approximately 25 g of lipids per run, an unfeasible amount given the low oil yield from oats.

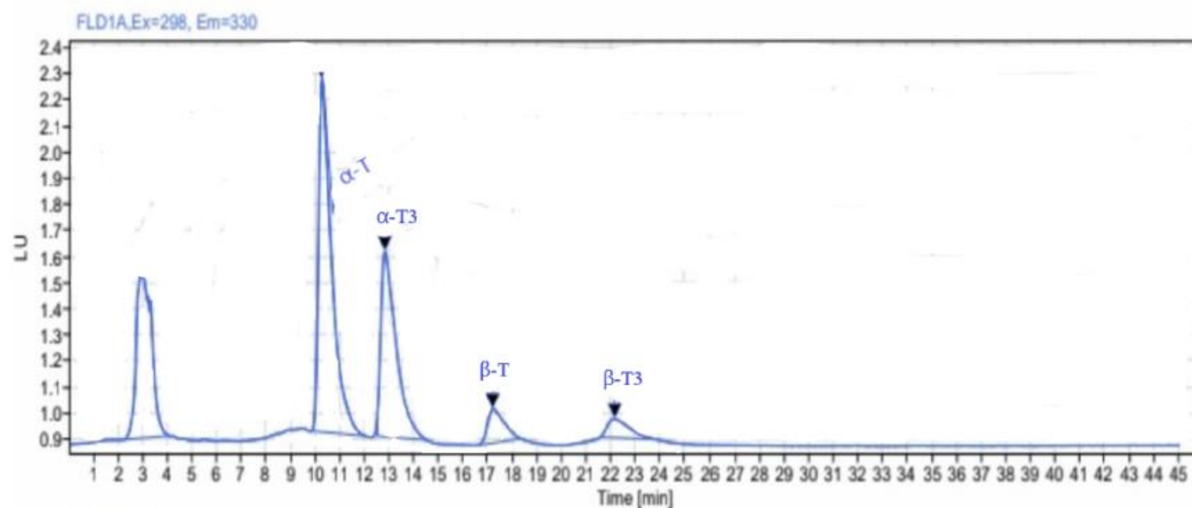
Crude oat oil, rapeseed oil, and sunflower oil (5 g each) plus 20 g of natural pork lard (K-classic, Czech Republic), and in addition 25 g of natural pork lard were added to 100 mL beakers of the same diameter (50 mm). These oils and fat were chosen as a control. The reason for choosing rapeseed and sunflower oils are the most common oils produced in the EU. Additionally, there were no added antioxidants in the oils as written in the label of oils. All samples were kept inside the oven (Binder GmbH, Tuttlingen, Germany) at 60 $\pm$ 2 °C. Sample weight was measured every 3-4 days on an analytical balance (Kern analytical balance ABS-N_ABJ-NM, Darmstadt, Germany) for 80 days until induction period (rapid increase in weight) was clearly detectable. The protection factor (PF) was the result of dividing the induction period (IP) per sample by the control IP (pork lard only). The extraction process was time-consuming extraction with low oil yield, due to this reason the Schaal oven test was conducted only once per oat variety. Relative weight change (RWC) was calculated using the following formula:

$$\mathrm{RWC[g/g]} = ((\mathrm{w2} - \mathrm{w}) - (\mathrm{w1} - \mathrm{w}))/(\mathrm{w1} - \mathrm{w}) \tag{1}$$

where w = weight of beaker without sample at day 0 [g], $w1$ = weight of beaker and sample with pork lard at day 0 [g], $w2$ = weight of beaker and sample with pork lard on any given day [g].

2.5. Rancimat test

The Rancimat test was applied on 5 g of crude oat oil, the samples mentioned in **Section 2.4**, as well as rice oil (GASTON, s.r.o., Zlín, Czech Republic) and butter (Madeta a. s., České Budějovice, Czech Republic). The analysis involved three replicates for oat oils and four replicates for the other oils. The characterization of various fats and oils (lard, butter, rapeseed, and sunflower oil) for comparison was accomplished by determining the fatty acid profile using GC-FID, following the method outlined in (**Lapčík et al., 2022**). The test was performed using a Metrohm Rancimat model 892 (Herisau, Switzerland), as recommended previously (**Metrohm, 2019**). The heat transfer medium was polyethylene glycol, suitable for non-traditional samples of oils, foodstuffs, and cosmetics (**Agilent, 2010 and Metrohm, 2019**). A stream of purified air 20 Lh— 1 was passed through 0.5 g of oil or fat and 3 g of PEG 3000 held at a constant temperature of 100 °C or 110 °C under conditions described in ISO method 6886:2016. Subsequently, effluent air from the oil sample was passed through a vessel filled with deionised water to create bubbles. Water conductivity was continuously monitored in StabNet 1.1 (Metrohm AG, Herisau, Switzerland), and the induction time was automatically calculated. The operating temperatures (100 °C and 110 °C), are recommended by Metrohm.



Sanitini

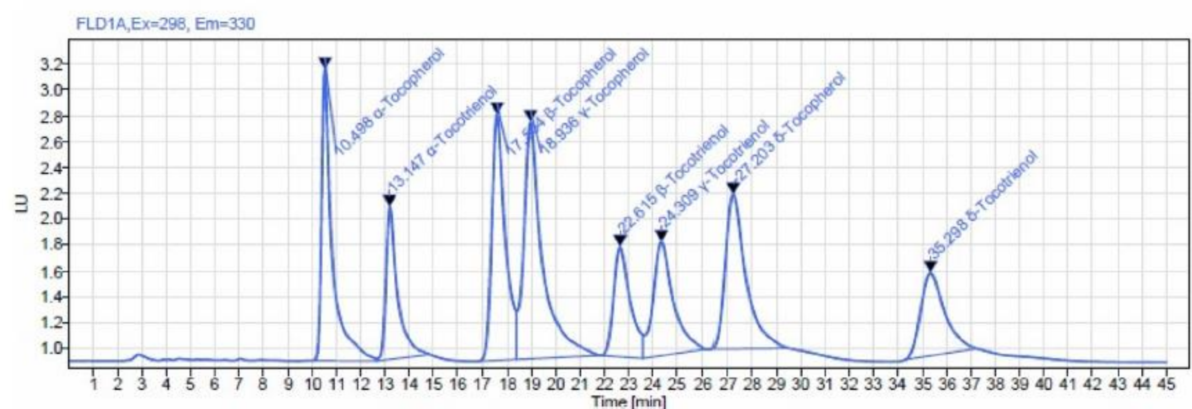


Fig. 1. Chromatogram of tocols of oat sample (Santini) and standard ($\alpha - T$: α-tocopherols, α-$T3$: α-tocotrienol, $\beta - T$: $\beta -$ tocopherol, $\beta - T3$: $\beta -$ tocotrienol).

2.6. Statistical analysis

Data were analysed in IBM SPSS version 29 (Armonk, New York, USA) and expressed as mean ± standard deviation (SD). One-way analysis of variance (ANOVA) and Tukey's post-hoc tests were used to determine between-group differences. Significance was set at $p \leq 0.05$. Pearson's Correlation coefficients were calculated to determine the relationship between the Schaal oven and Rancimat tests data.

3. Results

3.1. Tocopherols and tocotrienols in oats

The contents and compositions of tocols were determined in oat cultivars (**Table 1**). Significantly, variations were identified in the contents of individual tocols, ranging from 28.31 to 89.81 α-tocopherol mg/ kg of oil, 21.76 to 65.65 α-tocotrienol mg/kg of oil, 1.84 to 7.50 β-tocopherol mg/kg of oil, 4.14 to 8.76 β-tocotrienol mg/kg of oil, 0.73 to 2.05 γ-tocopherol mg/kg of oil and 0.27 to 1.80 γ-tocotrienol of oil (**Table 1**). The total tocols content ranged from 62.05 to 155.29 mg/kg. Among the oat cultivars, total tocol content was highest in Santini (155.3 mg/kg of oil), followed by Saul (144.5 mg/kg of oil) and Korok (105.9 mg/kg of oil). Tocol content was lowest in Marco Polo (62.1 mg/ kg of

oil). Tocopherol was the most abundant form of tocol in all culti-vars except Patrik and Saul, where α-tocotrienol was the predominant form. Interestingly, Saul had the highest α-tocotrienol content (8.8 mg/ kg of oil), a potentially valuable trait for oat breeding programs. Notably, β-tocopherol was detected at low concentrations in all cultivars, with the highest content reaching 7.5 mg/kg of oil in Santini. Sunflower oil was chosen as a representative, highly unsaturated plant oil with reasonable tocopherol content. In contrast, pork lard is high in saturated fats with low to no tocol content. Sunflower oil had comparable tocopherol content (78.1 mg/kg of oil) as most oat samples (**Table 1**). As expected, pork lard was low in tocol content (approximately 2 mg/kg). Hulled oats, Atego, and Korok contained higher tocol content than most naked oats, except for Santini and Saul. The variations in tocol content among different cultivars were notably significant. The of total tocols, tocopherols, and tocotrienols content in naked and hulled oats, along with control samples, is depicted in the **Fig. S1**.

Table 1 Concentration of tocopherols and tocotrienols in oats, sunflower oil and pork lard (mg/kg).

	α-tocopherol	α-tocotrienol	β-tocopherol	β-tocotrienol	γ-tocopherol	γ-tocotrienol	Total tocopherols	Total tocotrienols	Total tocols
Saul	63.44 ± 1.52^{c}	65.65 ± 4.98^{a}	3.61 ± 0.09^{bcd}	8.76 ± 1.06^{a}	1.20 ± 0.21^{b}	1.80 ± 0.31^{a}	68.25 ± 1.90^{b}	76.21 ± 7.95^{a}	144.46 ± 6.18^{a}
Marco Polo	28.31 ± 3.77^{f}	23.53 ± 1.59^{d}	4.69 ± 0.37^{bc}	4.51 ± 0.96^{b}	0.73 ± 0.65^{c}	0.27 ± 0.06^{c}	33.73 ± 2.81^{d}	28.32 ± 2.98^{e}	62.05 ± 3.79^{d}
Patrik	32.35 ± 3.33^{f}	35.55 ± 4.48^{c}	2.42 ± 0.74^{cd}	6.01 ± 2.38^{ab}	ND	ND	34.76 ± 3.04^{d}	41.57 ± 2.34^{cd}	76.33 ± 4.07^{cd}
Santini	89.81 ± 7.47^{a}	48.54 ± 2.52^{b}	7.50 ± 1.37^{a}	6.91 ± 0.83^{ab}	2.05 ± 0.35^{a}	0.47 ± 0.18^{b}	99.36 ± 6.75^{a}	55.93 ± 1.40^{b}	155.29 ± 6.31^{a}
Oliver	44.07 ± 4.34^{e}	21.76 ± 2.84^{d}	1.84 ± 0.76^{d}	7.21 ± 0.40^{ab}	ND	ND	45.92 ± 4.71^{cd}	28.97 ± 3.02^{e}	74.89 ± 7.68^{cd}
Atego	56.67 ± 3.33^{cd}	35.16 ± 3.01^{c}	5.81 ± 0.72^{ab}	4.14 ± 1.63^{b}	ND	ND	62.47 ± 3.73^{b}	39.3 ± 4.63^{de}	101.78 ± 8.20^{b}
Korok	48.72 ± 4.34^{de}	46.48 ± 2.51^{b}	5.45 ± 2.0^{ab}	5.32 ± 0.84^{ab}	ND	ND	54.17 ± 6.02^{bc}	51.80 ± 1.81^{bc}	105.97 ± 7.56^{b}
Kamil	44.23 ± 3.47^{e}	40.48 ± 1.45^{bc}	2.26 ± 0.74^{cd}	6.42 ± 2.31^{ab}	ND	0.51 ± 0.08^{b}	46.49 ± 3.38^{cd}	47.41 ± 3.38^{bcd}	93.90 ± 5.77^{bc}
Sunflower oil	78.16 ± 5.09^{b}	ND	1.48 ± 0.13^{d}	ND	0.16 ± 0.03^{d}	ND	79.77^{a}	ND	79.77^{cd}
Pork lard	2.09 ± 0.36^{g}	ND	ND	ND	ND	ND	2.09^{f}	ND	2.09^{e}

ND = not detected. Different lowercase letters in a column indicate significant differences ($p \leq 0.05$). Values are expressed as means $\pm$ SD of independent analyses in triplicate ($n = 3$).

3.2. Schaal oven test

The oxidative kinetics of oats and control are shown in **Figs. 2-4**. **Fig. 2** displays the oxidation kinetics of Oliver, Kamil, Patrik, and Atego, while **Fig. 3** illustrates the oxidation kinetics of Korok, Marco Polo, Santini, and Saul oats. **Fig. 4** portrays the oxidation kinetics of pork lard, rapeseed oil, and sunflower oil. Adding pork lard to rapeseed and sunflower oils increased stability through elevating natural antioxidants levels compared with lard-only samples.

The induction period (Ips) for all oat samples in lard exceeded 50 days (**Table 2**). The protection factor (PFs) of all hulled and naked oat samples ranged from 7.8 to 8.6 days. Atego, Patrik, and Santini exhibited the highest PFs. Oats with higher tocol levels (Santini) had longer IP while Patrik, which had the second lowest tocol content, exhibited a similar IP.

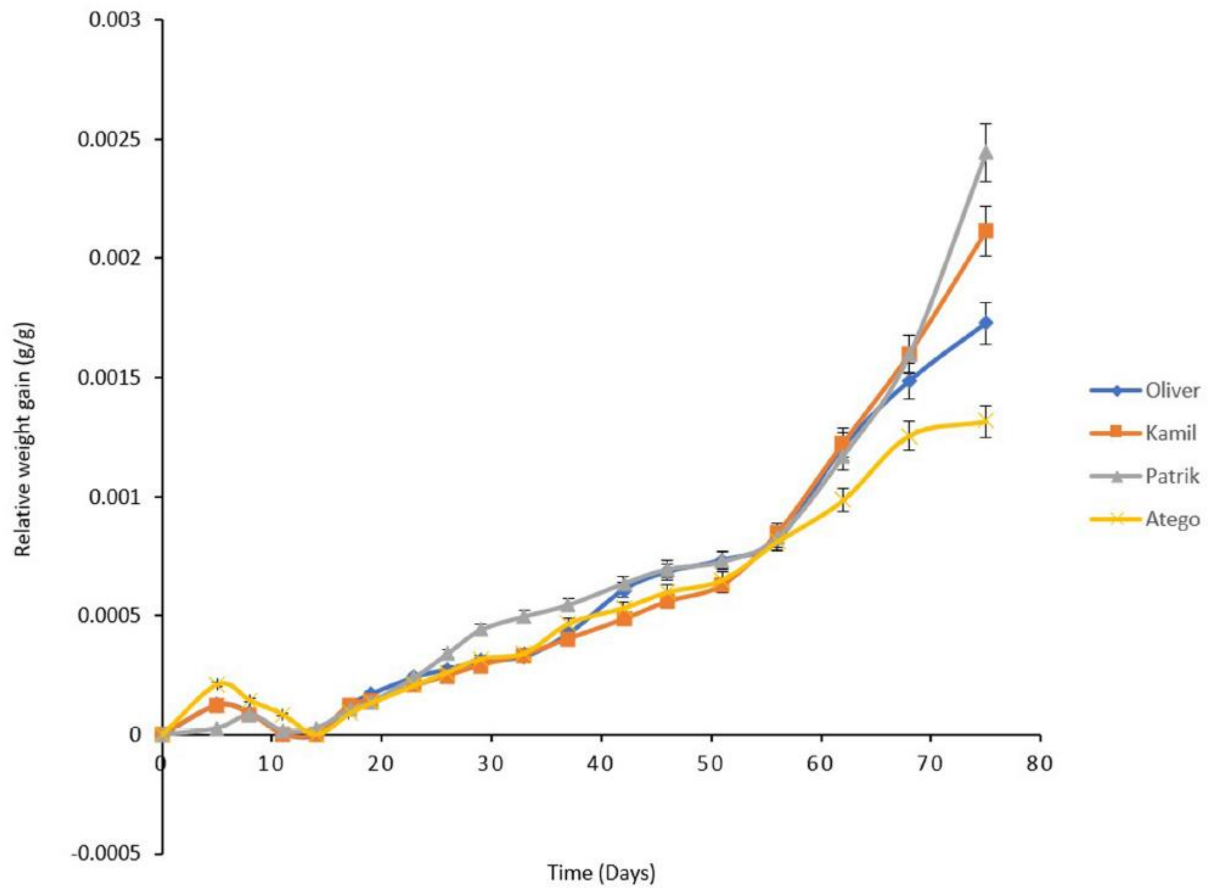


Fig. 2. Oxidation time-course of yellow oat oil mixed with pork lard in the Schaal oven test.

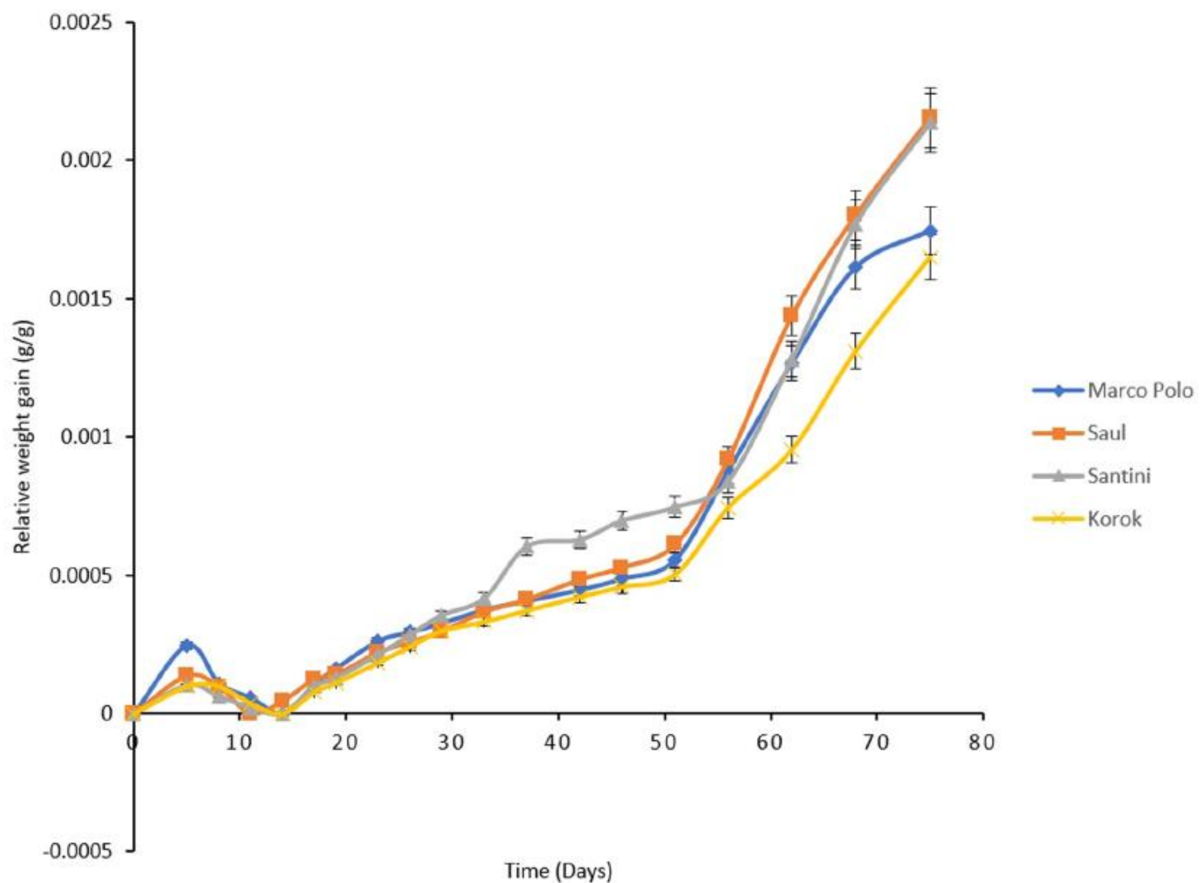


Fig. 3. Oxidation time-course of hulled and naked oat oils mixed with pork lard in the Schaal oven test.

3.3. Rancimat test

The results of the Rancimat test showed that Kamil oxidized faster than other oat cultivars, with an average IP of 4.12 h at 100 °C and 1.49 h at 110 °C (**Table 3**). In contrast, Saul oxidized the slowest, with an average IP of 9.02 h at 100 °C and 4 h at 110 °C. The IPs of other oat samples ranged between 5 to 7 h at 100 °C and between 2 to 3 h at 110 °C. When comparing the IPs of other oils and fats with oats, rice oil had lower oxidation stability; with its IP measuring 2.14 and 1.26 h at 100 °C and 110 °C, respectively. The proportion of the main fatty acid groups (SFA: MUFA: PUFA 1:2:2) is similar between rice oil and oat oil.

Crude oat oils showed longer stability with IPs ranging from 4 to 9 h, while refined edible oils/fats had shorter IPs, averaging around 1 h (**Fig. 5**). Crude oat oils were extremely stable at both test temperatures. Oat oils and other oils differed in IPs at 110 °C (**Fig. 6**). However, oat oil was more stable (IP range of 2.8 to 5.9 h) than other edible oils and fats (IP range of 0.66 to 1.26 h). These results suggest that refining processes decrease natural antioxidant content of edible plant oils and lower oxidation stability. Notably, butter and lard, having naturally low antioxidant levels, are inherently less stable.

4. Discussion

This study is one of the few to determine the oxidative stability of oat oil using the Schaal oven and Rancimat methods and to investigate tocol concentrations in oats. The tocol levels analysed in the studied oat oil samples in average are α-tocopherol at 59.06 mg/kg of oil, α-tocotrienol at 43.705 mg/kg of oil, β-tocopherol at 4.67 mg/kg of oil, β-tocotrienol at 6.45 mg/kg of oil, γ-tocopherol at 1.39 mg/kg of oil, and γ-tocotrienol at 1.035 mg/kg of oil. Total tocol concentration in studied oat oil samples aligned with previous research that reported 90 ppm a-tocopherol, 13 ppm γ-tocopherol, and 6.5 ppm δ-tocopherol (**Saga et al., 2013**).

The α-form of tocopherol and tocotrienol is most prominent, followed by the β-form, which is consistent with the previous findings of **Panfili et al. (2003); Gutierrez-Gonzalez et al. (2013); Shammugasamy et al. (2013); Redaelli et al. (2016)**. Beyond the a and p forms of to-copherols and tocotrienols, **Gutierrez-Gonzalez et al. (2013); Shammugasamy et al., (2013)** have reported the presence of the γ-form in trace amounts, which aligns with the observations in the current study. The δ-form was found in some oat cultivars, as reported by **Shammugasamy et al.** (**2013**), whereas the current study did not find the δ-form of to-copherols and tocotrienols. Unlike other studies, the current study has found higher content of α-tocopherol in six out of eight oat varieties.¨

Study focusing on hulled and naked oat genotypes found significantly lower a-tocopherol concentrations (4.5-12.3 mg/kg) than current study (**Sterna et al., 2016**). Overall, the data suggest that a variety of factors influence tocol concentration in oat oil, including genotype, environmental conditions, growing condition, and the extraction method.

All the cited authors have used similar types of methods, which require hot saponification and subsequent solvent extraction of tocols from oat grain matrix. The variation in tocols content in the current and previous study is due to differences in the methods used to identify and quantify tocols, different cultivars and the growing conditions of oats. Direct methods have certain benefits compared to hot saponification. It can prevent potential harm to unsaturated tocotrienols occurring due to severe alkaline conditions. It can also prevent questionable interference with tocol detection. Lastly, it helps keep sensitivity and selectivity high, notably when using fluorescence detection (**Peterson et al., 2007**).

Oat oils possess remarkable oxidative stability properties. The PF for oats was 1.8 in a previous study (**Holasova et al., 2002**), considerably lower than our results in general. This disparity can be attributed to differences in the experimental design. Our study extracted and measured crude oat oil, whereas the other study measured dried ground oat samples mixed with 10 g lard at 70 °C (**Holasova et al., 2002**). Similarly, **Saga et al. (2013)** indicated that adding 5% and 10% crude oat oil to fish oil increased stability by approximately two-fold, based on Schaal oven test results at 70 °C. Moreover, pure oat oil did not increase in weight after 50 days at 70 °C, indicating that oat oil is highly resistant to oxidation, corroborating our findings of high stability.

Furthermore, a study examining the rancidity resistance of diacylglycerol-enriched soybean oil and palm olein when exposed to heat (**Wang et al., 2010**) found that soybean oil had an IP of 9.46 h at 110 °C. In contrast, diacylglycerol-enriched soybean oil had an IP of only 4.21 h; its higher unsaturated fatty acid concentrations and lower tocopherol levels decreased its oxidative stability. At the same temperature, palm olein had an IP of 21.53 h, whereas diacylglycerol-enriched palm olein only had an IP of 5.40 h. Both soybean and palm olein oils had longer IPs than the oat samples analysed in our study.

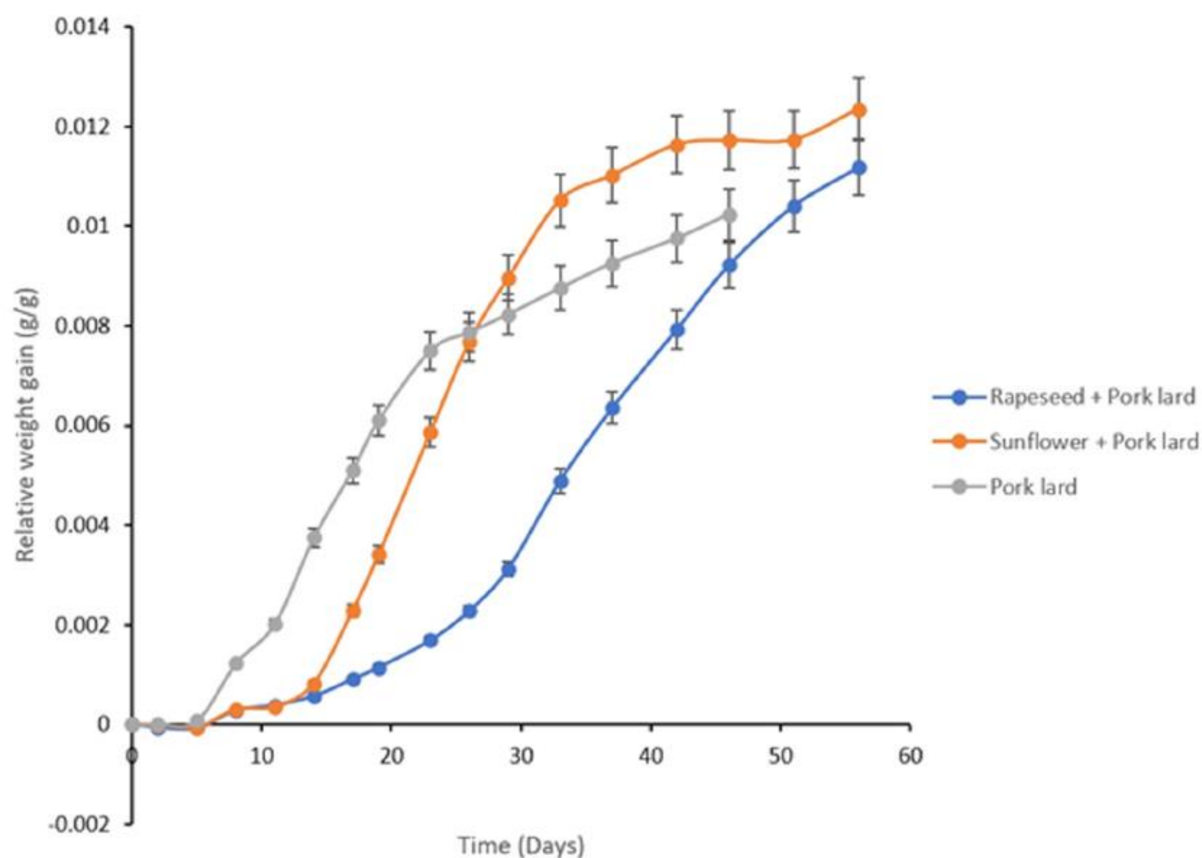


Fig. 4. Oxidation time-course of pork lard alone (control), as well as rapeseed and sunflower oils mixed with pork lard in the Schaal oven test

Table 2 Induction period and protection factor of oats, rapeseed, and sunflower oil with pork lard.

Oil sample (5 g) + pork lard (20 g)	Induction period (days)	Protection factor
Atego (hulled oat)	56.0	8.6
Kamil (naked oat)	54.0	8.3
Korok (hulled oat)	55.5	8.5
Marco Polo (naked oat)	51.0	7.8
Oliver (naked oat)	55.0	8.5
Patrik (naked oat)	56.0	8.6
Santini (naked oat)	56.0	8.6
Saul (naked oat)	52.0	8.0
Rapeseed oil	23.0	3.5
Sunflower oil	14.0	2.2
Control - pork lard (25 g)	6.5	1.0

Table 3 Induction periods of samples and controls (0.5 g samples + 3 g polyethylene glycol-PEG 3000) at 100 °C and 110 °C in the Rancimat test.

Samples	100 °C Induction period (h)	110 °C Induction period (h)
Kamil	4.12 ± 0.30^{b}	1.49 ± 0.47^{b}
Saul	9.02 ± 0.27^{a}	4.00 ± 0.30^{a}
Marco Polo	6.80 ± 1.06^{ab}	2.90 ± 0.15^{b}
Oliver	7.29 ± 0.5^{ab}	3.33 ± 0.32^{ab}
Atego	5.06 ± 0.27^{b}	2.22 ± 0.09^{b}
Patrik	5.45 ± 0.47^{b}	3.50 ± 0.33^{ab}
Santini	5.96 ± 0.59^{b}	2.92 ± 0.23^{b}
Korok	6.86 ± 1.15^{ab}	2.83 ± 0.11^{b}
Pork lard	1.18 ± 0.04	0.77 ± 0.05
Butter	1.07 ± 0.06	0.66 ± 0.08
Rapeseed oil	1.34 ± 0.07	0.75 ± 0.02
Sunflower oil	1.32 ± 0.08	0.81 ± 0.06
Rice oil	2.14 ± 0.05	1.26 ± 0.19

Values are expressed as means ± standard deviation (SD) of independent analyses run in triplicate. Different lowercase letters in a column indicate significant differences ($p \leq 0.05$). Only oat samples were included in the ANOVA.

Based on our analysis of fatty acid profile, oat oil (24% saturated fatty acid [SFA], 39% monounsaturated fatty acid [MUFA], 36% polyunsaturated fatty acid [PUFA]) was expected to have a shorter IP than what we obtained from the Rancimat test.

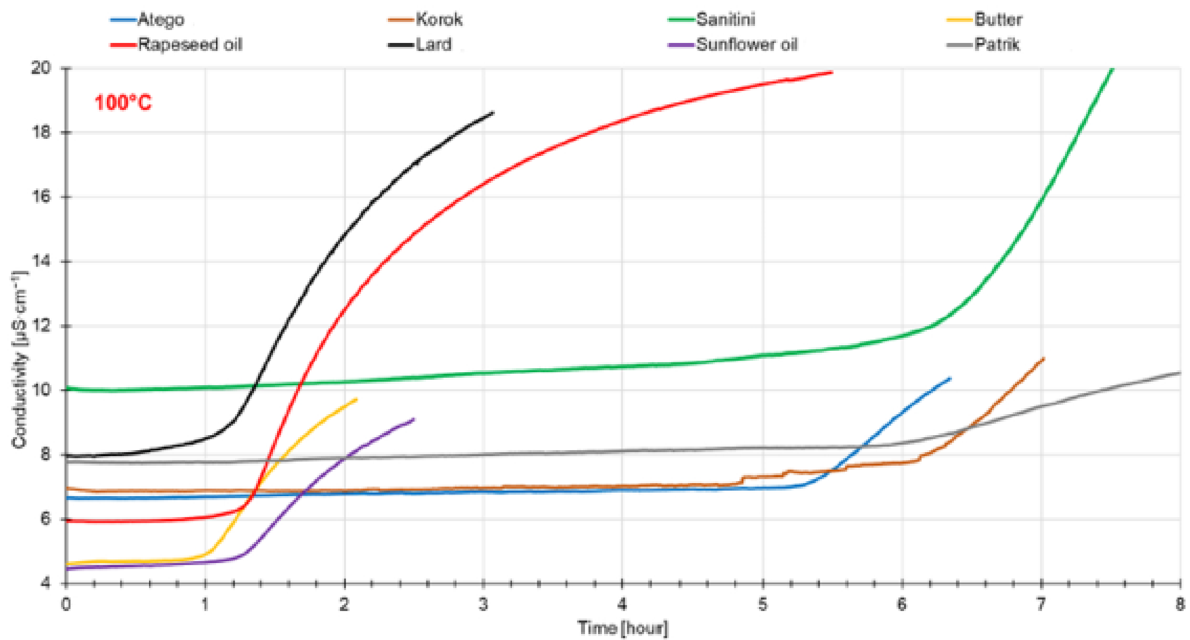


Fig. 5. Stability curves for selected varieties of oat oils, edible oils, and fats at 100 °C in the Rancimat test.

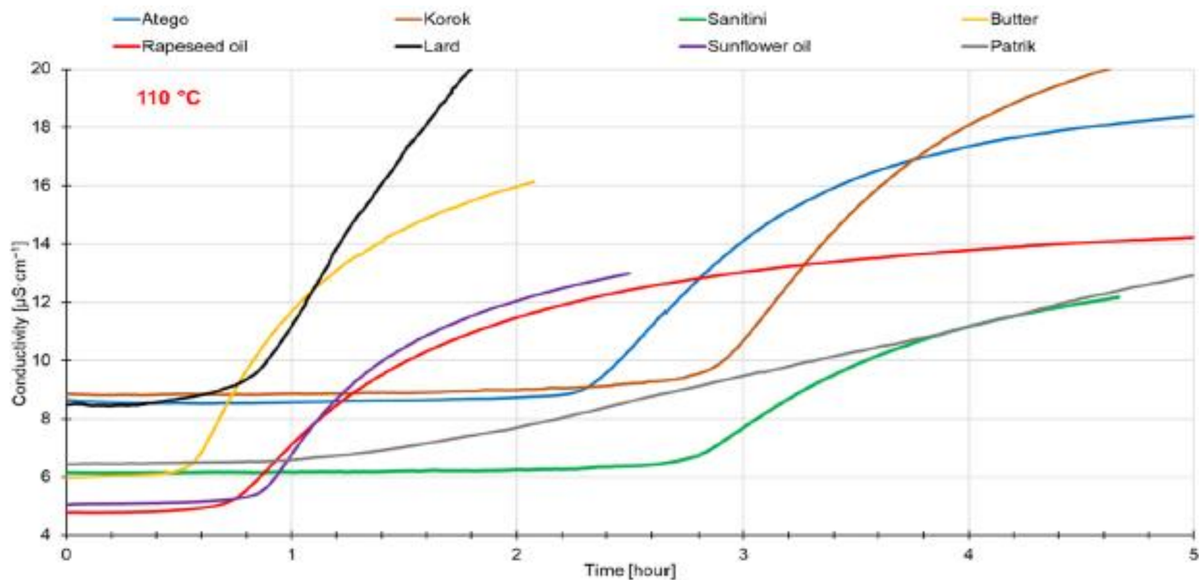


Fig. 6. Stability curves for selected varieties of oat oils, edible oils, and fats at 110 °C in the Rancimat test.

The fatty acid profile is in contrast to lard (35% SFA, 47% MUFA, 18% PUFA) and butter (66% SFA, 30% MUFA, 4% PUFA); the lower antioxidant levels of saturated oils resulted in shorter IPs than the unsaturated oat oils. Next, in our study unsaturated rapeseed (8% SFA, 66% MUFA, 26% PUFA) and sunflower (11% SFA, 35% MUFA, 54% PUFA) oils had slightly longer IPs than saturated fats, but noticeably shorter IPs than oat oils. Thus, fatty acid composition appears to exert a smaller effect on oxidation than the presence of antioxidants in oat oils. Differences in the results obtained between the Schaal oven and Rancimat tests have been observed, and these disparities could be attributed to several factors rather than a specific one. Firstly, the Rancimat analysis mechanism is designed to measure the IP by detecting the formation of volatile acids during oil oxidation (**Aktar and Adal, 2019**). This method is highly dynamic because a significant volume of air, which has been filtered and purified, is bubbled through the heated sample at a rate of 20 L/h. Moreover, reactions take place throughout the entire sample volume. On the other hand, the Schaal test is much more static, typically without the mixing of samples and without air or oxygen bubbling. This method focuses more on storage conditions than the Rancimat test. Secondly, the Schaal oven test involves the measurement of sample weight every 3-4 days using an analytical balance, while the Rancimat test is automated. Additionally, factors such as free fatty acids, metals, and their ions, and chlorophylls can also influence the oxidation and induction period of oat oils.

We did not identify a correlation between tocol content and the IP of oat oils when running the Schaal oven tests. While tocopherols and tocotrienols are important in enhancing oxidative stability of oat oils, other components in oats may also have protective effects. Components with antioxidant potential include avenanthramides, phenolic compounds, phytic acids, flavonoids, and sterols. In Rancimat test, β-toco-trienol content correlated modorately with IP at both temperatures (correlation coefficient $(r) = 0.4392$ at 100 °C and at 110 °C $(r) = 0.4389$). Hence, other compounds with antioxidant activity did not appear to be effective under the Rancimat test conditions. Correlation between tocols and oxidative stability observed in this study provides insights for the selection and optimization of processing conditions to preserve the quality and stability of oat oil during heat treatment. This knowledge can be utilized to develop healthier and more stable oat-based products.

In the present scenario, oat oil is primarily acquired in larger quantities as a byproduct during the bio-refinement of oat grains to isolate functional components like β-glucans. Currently, the number of

operational biorefineries is limited. There is a lack of sustainable biomass utilization, both in food and non-food sectors. Furthermore, recent advancements in the fractionation of oat grains through novel methods and techniques unlock the potential for oat to serve as a versatile raw material across numerous industrial domains. Oat grains and their derivatives have the capacity to contribute to the creation of innovative products in various sectors, including pharmaceuticals, chemicals, cosmetics, and food, catering to both medicinal and dietary needs (Liu, 2014; **Sang and Chu, 2017**; Banas and Harasym, 2021).

Previous research using the Rancimat test demonstrated that the IP of sunflower oil was 226.2 min at 110 °C and 127.2 min at 120 °C (**Almoselhy, 2021**). The IP period lasted for 5 h during the first month of storage. However, in the twelfth month of storage, when subjected to a Rancimat test at 120 °C, the IP period reduced to 3.2 h (**Maszewska et al., 2018**). These values are higher than the IPs observed for rapeseed and sunflower oil at 100 °C in current study (1.34 h and 1.32 h, respectively).

Interestingly, Saul, contained reasonable amount of total tocol content, exhibited the longest IP in the Rancimat test. Our findings are consistent with reports that the Rancimat test yielded an IP of 6.9 h for oat oil and its fractions at 110 °C (**AG, 2019**). At concentrations of 1-5%, oat oil increased the IP of lard and tallow two- to eight-fold from non-antioxidant levels. When the Rancimat method (100-130 °C) was used to assess the stability of olive oil (semi-fine, fine, refine, and extra virgin) (**Farhoosh and Hoseini-Yazdi, 2014**), the findings revealed that extra virgin olive oil was the most stable, lasting for 55.1 h at 100 °C and 5 h at 130 °C, while semi-fine oil only lasted 15.6 h at 100 °C and 1.6 h at 130 °C. Notably, however, olive oil was more stable than oat oil at both temperatures.

In our study, we demonstrated that oat oil had slow oxidation during heating and cooking than rapeseed and sunflower oils (**Figs. 4 and 5**). Introducing new oat cultivars with increased yield and higher antioxidant content widens their applicability in various food and industrial sectors. Furthermore, the stability of these cultivars will improve processing to obtain refined oat oil.

In summary, the result of current study indicates that oats have longer stability than common edible vegetable oils (rapeseed and sunflower) rice oils and saturated fats (butter and lard), contributing to their superior oxidative stability. Other bioactive components such as ave-nanthramides also play a role in the stability of oat oil against oxidation.

5. Conclusions

This study provided valuable comparative data on the oxidative stability of crude oat oil using Schaal oven and Rancimat tests. Schaal oven test results were more similar across cultivars, whereas between-cultivar differences emerged in the Rancimat test. Interestingly, oat oil was more stable than commercial oils and fats. The Saul variety had the highest IP and a-tocotrienol content. There were differences in the concentration of tocopherols and tocotrienols among the oat varieties. Additionally, hulled oats contained higher tocol content than most of the naked oats (except Santini and Saul). Tocopherol and tocotrienol concentrations in oat oils were not significantly correlated with the Schaal-derived IP. Nevertheless, β-tocotrienol was correlated with Rancimat-derived IP at both temperatures. The study concluded that in addition to the tocopherols and tocotrienols, other components with antioxidant potential, such as avenanthramides, phenolic compounds, and phytic acid, could contribute to the stability of oat against oxidation. Future research should focus on investigating the specific antioxidant mechanisms of oat components like avenanthramides, phenolic compounds, and phytic acid, and their contributions to oat oil stability.

References

Abasolo, C., 2021a. Accelatated Oxidation Test: The Schaal Mehods [WWW Document]. BTSA. URL (**www.btsa.com/en/accelerated-oxidation-tests-the-schaal-method**).

Abasolo, C., 2021b Accelatated Oxidation Test: The RANCIMATE Methods [WWW Document]. BTSA. URL (**www.btsa.com/en/accelerated-oxidation-tests-rancimat-method/**) Accessed 23 May 2023.

AG, M., 2019. Oxidation stability of coffee.

Agilent, 2010. Quality Analysis of Virgin Olive Oils - Part 4 Nutritive benefi ts - Quantifi cation of tocopherols and tocotrienols in virgin olive oil using the Agilent 1260. Appl. Note.

Aktar, T., Adal, E., 2019. Determining the Arrhenius kinetics of avocado oil: oxidative stability under rancimat test conditions. Foods 8, 1-13. **https://doi.org/10.3390/ foods8070236**.

Almoselhy, R.I.M., 2021. Comparative study of vegetable oils oxidative stability using DSC and Rancimat Methods. Egypt. J. Chem. 64, 299-312. **https://doi.org/ 10.21608/EJCHEM.2021.51238.3051**.

Banaś, A., Debski, H., Banas, W., Heneen, W.K., Dahlqvist, A., Bafor, M., Gummeson, P. O., Marttila, S., Ekman, Å., Carlsson, A.S., Stymne, S., 2007. Lipids in grain tissues of oat (Avena sativa): differences in content, time of deposition, and fatty acid composition. J. Exp. Bot. 58, 2463-2470. **https://doi.org/10.1093/jxb/erm125**.

Banaś, K., Harasym, J., 2021. Current knowledge of content and composition of oat oil—future perspectives of oat as oil source. Food Bioprocess Technol. 14, 232-247. **https://doi.org/10.1007/s11947-020-02535-5**.

Bryngelsson, S., Mannerstedt-Fogelfors, B., Kamal-Eldin, A., Andersson, R., Dimberg, L. H., 2002. Lipids and antioxidants in groats and hulls of Swedish oats (Avena sativa L). J. Sci. Food Agric. 82, 606-614. **https://doi.org/10.1002/jsfa.1084**.

Chen, H., Qiu, S., Gan, J., Li, Z., Nirasawa, S., Yin, L., 2016. New insights into the antioxidant activity and components in crude oat oil and soybean oil. J. Food Sci. Technol. 53, 808-815. **https://doi.org/10.1007/s13197-015-1991-0**.

Emmons, L.C.M., Peterson, D.L., Paul, G., 1999. Antioxidant capacity of Oat (Avena sativa L.) extracts. 2. in vitro antioxidant activity and contents of phenolic and tocol antioxidants. J. Agric. Food Chem. 47, 4894-4898. **https://doi.org/10.1021/ jf990530i**.

Farhoosh, R., Hoseini-Yazdi, S.Z., 2014. Evolution of oxidative values during kinetic studies on olive oil oxidation in the Rancimat test. JAOCS, J. Am. Oil Chem. Soc. 91, 281-293. **https://doi.org/10.1007/s11746-013-2368-z**.

Gangopadhyay, N., Hossain, M.B., Rai, D.K., Brunton, N.P., 2015. A review of extraction and analysis of bioactives in oat and barley and scope for use of novel food processing technologies. Molecules 20, 10884-10909. **https://doi.org/10.3390/ MOLECULES200610884**.

Gutierrez-Gonzalez, J.J., Wise, M.L., Garvin, D.F., 2013. A developmental profile of tocol accumulation in oat seeds. J. Cereal Sci. 57, 79-83. **https://doi.org/10.1016/j. jcs.2012.10.001**.

Hernandez-Hernandez, O., Pereira-Caro, G., Borges, G., Crozier, A., Olsson, O., 2021. Characterization and antioxidant activity of avenanthramides from selected oat lines developed by mutagenesis technique. Food Chem. 343, 128408 **https://doi.org/ 10.1016/J.FOODCHEM.2020.128408**.

Holasova, M., Fiedlerova, V., Smrcinova, H., Orsak, M., Lachman, J., Vavreinova, S., 2002. Buckwheat - the source of antioxidant activity in functional foods. Food Res. Int. 35, 207-211. **https://doi.org/10.1016/S0963-9969(01)00185-5**.

Jaksics, E., Nemeth, R., Schall, E., Szentmiklóssy, M.K.J., Bidlo, G., Simon, K., Tomoskozi, S., 2023. Study of the effects of heat treatment on the composition, functionality, and oxidative and hydrolytic stability of oat. Cereal Chem. 708-720. **https://doi.org/10.1002/cche.10646**.

Kouřimská, L., Pokhrel, K., Božík, M., Tilami, S.K., Horčička, P., 2021. Fat content and fatty acid profiles of recently registered varieties of naked and hulled oats with and without husks. J. Cereal Sci. 99 **https://doi.org/10.1016/j.jcs.2021.103216**.

Lapčík, L., Lapčíkova, B., Gautam, S., Vasina, M., Valenta, T., Repka, D., Cepe, K., Rudolf, O., 2022. Acoustic and mechanical testing of commercial cocoa powders. Int. J. Food Prop. 25, 2184-2197. **https://doi.org/10.1080/10942912.2022.2127760**.

Maszewska, M., Florowska, A., Dluzewska, E., Wroniak, M., Marciniak-Lukasiak, K., Zbikowska, A., 2018. Oxidative stability of selected edible oils. Molecules 23, 15-17. **https://doi.org/10.3390/molecules23071746**.

Metrohm A.G., 2019. Oxidation stability of chocolate chocolate without sample preparation.

Molteberg, E.L., Magnus, E.M., Bj0rge, J.M., Nilsson, A., 1996. Sensory and chemical studies of lipid oxidation in raw and heat-treated oat flours. Cereal Chem. 73, 579-587.

Panfili, G., Fratianni, A., Irano, M., 2003. Normal phase high-performance liquid chromatography method for the determination of tocopherols and tocotrienols in cereals. J. Agric. Food Chem. 51, 3940-3944. **https://doi.org/10.1021/jf030009v**.

Peterson, D.M., 2001. Oat antioxidants. J. Cereal Sci. 33, 115-129. **https://doi.org/ 10.1006/JCRS.2000.0349**.

Peterson, D.M., Jensen, C.M., Hoffman, D.L., Mannerstedt-Fogelfors, B., 2007. Oat tocols: saponification vs. direct extraction and analysis in high-oil genotypes. Cereal Chem. 84, 56-60. **https://doi.org/10.1094/CCHEM-84-1-0056**.

Pokhrel, K., Kouřimská, L., Pazderů, K., Capouchová, I., BoZik, M., 2022. Lipid content and fatty acid profile of various European and Canadian hulled and naked oat genotypes. J. Cereal Sci. 108 **https://doi.org/10.1016/jjcs.2022.103580**.

Redaelli, R., Dimberg, L., Germeier, C.U., Berardo, N., Locatelli, S., Guerrini, L., 2016. Variability of tocopherols, tocotrienols and avenanthramides contents in European oat germplasm. Euphytica 207, 273-292. **https://doi.org/10.1007/s10681-015-1535-8**.

Saga, L.C., Kristinova, V., Kirkhus, B., Jacobsen, C., Skaret, J., Liland, K.H., Rukke, E.O., 2013. Characterization of oxidative stability of fish oil- and plant oil-enriched skimmed milk. JAOCS J. Am. Oil Chem. Soc. 90, 113-122. **https://doi.org/10.1007/ s11746-012-2148-1**.

Sang, S., Chu, Y., 2017. Whole grain oats, more than just a fiber: role of unique phytochemicals. Mol. Nutr. Food Res. 61, 1600715. **https://doi.org/10.1002/ mnfr.201600715**.

Shammugasamy, B., Ramakrishnan, Y., Ghazali, H.M., Muhammad, K., 2013. Combination of saponification and dispersive liquid-liquid microextraction for the determination of tocopherols and tocotrienols in cereals by reversed-phase high-performance liquid chromatography. J. Chromatogr. A 1300, 31-37. **https://doi. org/10.1016/j.chroma.2013.03.036**.

Sterna, V., Zute, S., Brunava, L., 2016. Oat grain composition and its nutrition benefice. Agric. Agric. Sci. Procedia. **https://doi.org/10.1016Zj.aaspro.2016.02.100**.

Viscidi, K.A., Dougherty, M.P., Briggs, J., Camire, M.E., 2004. Complex phenolic compounds reduce lipid oxidation in extruded oat cereals. Lwt 37, 789-796. **https:// doi.org/10.1016/j.lwt.2004.03.005**.

Wang, Y., Zhao, M., Tang, S., Song, K., Han, X., Ou, S., 2010. Evaluation of the oxidative stability of diacylglycerol-enriched soybean oil and palm olein under rancimat-accelerated oxidation conditions. JAOCS J. Am. Oil Chem. Soc. 87, 483-491. **https://doi.org/10.1007/s11746-009-1521-1**.