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Reduction of acrolein concentrations in palm cooking oil emissions through the addition of *Muntingia calabura* (aratiles) leaf extract to inhibit lipid peroxidation

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Abstract

Inhaling acrolein, an aldehyde present in cooking oil fumes, is detrimental to human health. Fatty acids in oils can undergo the degradation process of oxidation, which produces acrolein and other volatile aldehydes. To prevent such degradation, plant-based antioxidants could be added to the oil. This study aimed to measure and compare acrolein concentrations in emissions of palm oil with varying concentrations (w/w) of *Muntingia calabura* ethanol leaf extract. Four set-ups were subjected to volatilization and dissolution through Rancimat analysis. Volatile oxidation products from the Rancimat were analyzed for acrolein through high-performance liquid chromatography. Results showed that acrolein concentrations decreased as the concentration of the leaf extract in the oil treatment increased. Thus, the addition of *M. calabura* leaf extract to palm oil has the potential of controlling oxidation during cooking by reducing acrolein emissions; safer kinds of cooking oil could be formulated with the preference of incorporating natural antioxidants.

Introduction. - Inhaled fumes from degrading cooking oil is a major matter of concern worldwide [1,2] due to their detrimental effects on the health of people staying indoors [2,3,4], particularly in the Philippines [5]. Mitigating the effects of indoor air pollution from oil-based cooking is important as people spend more than 90% of their time indoors [6,7,8], especially during the COVID-19 pandemic [9]. Meanwhile, among the air pollutants, aldehydes and polycyclic aromatic hydrocarbons are found to be the major products of oil degradation [3] and the most harmful [10,11,12].

As cooking oil is heated beyond its threshold smoke point, fatty acids present in oil oxidize rapidly to form aldehydes and free radicals [12]. However, the smoke point of the oil is not a reliable basis of aldehyde formation, as the formation of volatile organic compounds occurs before the temperature of the oil reaches the smoke point [13]. Oxidative stability is the better indicator of oil performance [13] since it is measured in terms of the time it takes for oils to start generating volatile secondary reaction products (induction time) before the temperature reaches the smoke point [14]. In addition, the rate of production of aldehydes is inversely related to the oxidative stability [12,15] of the cooking oil and directly related to cooking temperatures [3], fatty acid content [16], and atmospheric oxygen [17]. Hence, in this study, the relationship between oxidative stability and the amount of aldehydes emitted by the treated palm cooking oil samples was investigated.

Most of the studies conducted on cooking oil have focused on the improvement of its oxidative stability [15,18], but there has been none in reducing the harmful constituents found in cooking oil fumes. In relation to aldehyde formation, studies have postulated a possible association between the addition of natural antioxidants to cooking oil and the amount of aldehydes released by cooking oil when heated [12,18]. This is because the production of aldehydes can be inhibited through the use of antioxidants by bonding towards the singlet oxygen in the reaction intermediate as fatty acids degrade, as established by the same studies. The cooking oil emissions will be analyzed for acrolein, the simplest unsaturated aldehyde and the most abundantly produced by palm oil and soybean oil [12]. With this, it is necessary to examine the correlation between the addition of natural antioxidants and acrolein emissions.

Among various methods to inhibit oxidation, the use of antioxidants is most effective, convenient, and economical [19]. Between the natural and the synthetic antioxidants, the former are safer for

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consumption due to toxicity problems associated with synthetic antioxidants [20]. Several studies investigated the effects of incorporating plant-based extracts, such as that from *Eucalyptus globulus* leaves, *Cinnamomum zeylanicum* bark, and *Sesamum indicum* coat, on the oxidative stability of cooking oil, resulting in longer induction times [15,18,20]. *Muntingia calabura* (aratiles) is one of the top five Philippine fruits to contain high antioxidant activity alongside *Diospyros blancoi* (velvet apple), *Tamarindus indica* (tamarind), *Sandoricum koetjape* (lolly fruit), and *Annona squamosa* (sugar apple) [22,23], making its extract a highly suitable additive, especially its leaves where the highest antioxidative activity can be found [24]. It is hypothesized that if *M. calabura* leaf extract is added to palm cooking oil (PCO), then its acrolein emissions will be reduced as an effect of the increase in its oxidative stability.

To address the issue of aldehyde emissions, this study aimed to measure and compare the acrolein concentrations in cooking oil fumes from palm oil with varying percent mass of *M. calabura* leaf extract. This study specifically aimed to:

(i) Determine the radical scavenging activity (RSA) and total phenolic content (TPC) of the *M. calabura* leaf extract through 2,2-diphenyl-1-picrylhydrazyl (DPPH) UV-visible radical scavenging assay;

(ii) Measure the induction time and antioxidative activity index (AAI) of the set-ups through Rancimat analysis;

(iii) Quantify the aldehyde concentrations derived from the fumes of the set-ups through high-performance liquid chromatography (HPLC);

(iv) Determine if there are significant differences among the induction times of the set-ups; and

(v) Determine if there are significant differences among the aldehyde concentrations from the fumes of the set-ups.

Methods. - The methods of this experimental study are composed of: (1) leaf harvesting and extraction, (2) DPPH assay, (3) treatment of oil samples, (4) Rancimat analysis, and (5) HPLC analysis. Four set-ups were prepared, with one set-up as the negative control and the other three as treatments.

Leaf Harvesting and Extraction. *M. calabura* leaves were washed and air-dried for one day, processed into smaller chunks, then transferred into a reagent bottle with 99% ethanol, following a 1:20 leaf to solvent ratio (w/v). The bottles were stored in a cool area for five days. Subsequently, the solution underwent vacuum filtration and rotary evaporation (60 °C heating bath temperature, 90 rpm, 5 hours). Lastly, the solution was reconstituted and air-dried in an exhaust for 3 days.

DPPH Assay. The RSA and TPC of the *M. calabura* leaf extracts were calculated through DPPH radical scavenging assay. Gallic acid was used as a standard to calculate the TPC. An extract-ethanol solution series (40, 80, 120, 160, 200 ppm) and a gallic acid solution (40 ppm) were prepared. Four milliliters (4 mL) of 0.1 mM DPPH-ethanol solution was added to 1 mL of each solution. All solutions were wrapped with aluminum foil and stored for 30 minutes in a cool dark area before the analysis. Absorbance values of the extracts and gallic acid were measured at a wavelength of 508.40 nm using a UV-vis spectrophotometer (Shimadzu UV-1800).

Treatment of Oil Samples. Four set-ups consisting of PCO samples with 0, 0.1%, 0.3%, and 0.5% mass of *M. calabura* leaf extract were prepared to achieve a total of 25 g. The maximum treatment concentration of 0.5% was based on the study of Mohamed et al. [25], which considered the limitations in the amount of additives safe for consumption. Oil samples were agitated for 5 minutes in an up-and-down motion before the analysis.

Rancimat Analysis. The Rancimat (Metrohm 892 Professional Rancimat) was set to a temperature of 150 °C and heated until it was stable to prevent temperature fluctuation. The gas flow of oxygen was set at the default setting of 20.0 L/hr while 60 mL of pure water (Merck, Elix Type II) was poured into each measuring vessel. The reaction vessels containing 3 g of the samples were heated in their respective heating blocks until the last sample had reached the stop criteria of 400 µS/cm conductivity. After 5 hours of heating, the resulting water samples were collected for further analysis of aldehydes. A total of 4 replicates were performed.

HPLC Analysis. Acrolein concentrations in the samples obtained from Rancimat analysis were determined through HPLC analysis (Shimadzu Prominence LC-20A UFLC Stack HPLC System), equipped with a UV-vis detector and a C8 150 x 4.6 mm 5µm column kept at 25 °C in the column oven. The analysis was carried out using an isocratic mode of elution using vacuum filtered distilled water as a mobile phase at a 2.0 mL/min flow rate. HPLC-grade acrolein (CPI International) standard solution (3, 6, 9, 12, and 15 ppm) was used to establish the calibration curve for quantification. The filtered (0.45 µm) samples were individually kept in a glass vial filled up to 2 mL volume, while 20 µL aliquots per sample were collected by the autosampler for analysis. A total of 4 replicates for each treatment were performed.

Computation of Parameters. The absorbance values obtained from the UV-vis spectrophotometer were used to calculate the RSA of the extract and gallic acid using Equation 1.

$$(1) \quad \%\ Inhibition = \frac{\lambda_{blank} - \lambda_{sample}}{\lambda_{blank}} \times 100\%$$

where λ_{blank} is the wavelength of the DPPH solution at maximum absorbance, and λ_{sample} is the wavelength of the DPPH-extract solution.

The TPC of the extract was calculated from the RSA of both gallic acid and the extract as shown in Equation 2.

$$(2) \quad TPC\ (\text{mg GAE/g}) = \frac{R_E \times 100}{R_G \times M_E}$$

where R_E is the RSA of the extract, R_G is the RSA of gallic acid, and M_E is the mass of extract used to calculate R_E.

Lastly, the AAI of the extract was calculated using Equation 3 [14].

$$(3)\quad AAI = \frac{I_{treatment}}{I_{control}}$$

where $I_{treatment}$ is the induction time of the treatment group and $I_{control}$ is the induction time of the control group.

Data Analysis. One-way analysis of variance (ANOVA) at α=0.05 was performed to determine if a significant difference exists between the percent mass extract in oil and the induction time among the four set-ups, as well as between the percent mass extract in oil and the acrolein concentration in its emissions. Pairwise comparisons using t-tests were then conducted to calculate the *p*-values between set-ups. All statistical analyses were performed using R software (RStudio®, v1.2.5033).

Safety Procedure. The samples and solutions were prepared under a fume hood. Personal protective equipment was worn at all times whenever handling reagents, samples, glassware, and equipment. Adherence to the respective safety data sheet of the chemicals was also observed. The use of equipment was done with proper protocol and training. Chemical and biological waste were placed in their respective waste containers to prevent further chemical reactions.

Results and Discussion. - After the DPPH assay was performed, the RSA was calculated, followed by the TPC. Through Rancimat analysis, the induction times of the samples were measured, and the AAI was determined. The water samples used from the Rancimat analysis were then analyzed for acrolein using the HPLC.

RSA and TPC. The RSA and TPC of the extracts were calculated based on the absorbance values obtained through UV-vis spectrophotometry. The RSA of extracts from *M. calabura* at different concentrations is shown in Table 1.

Table 1. Summary of radical scavenging activity (RSA), and total phenolic content (TPC) of increasing concentrations of leaf extract to DPPH solution. Data used to calculate for RSA and TPC are expressed in average by the UV-vis spectrophotometer.

Concentration (ppm)	RSA (%)	TPC (mg GAE/g)
40-Gallic Acid	94.24	N/A
40	54.59	57.92
80	56.63	60.09
120	60.47	64.16
160	63.47	67.35
200	66.59	69.38

RSA determines how likely the antioxidants present in *M. calabura* prevent the release of free radicals. DPPH is a purple-colored solution that gradually changes to a yellowish solution depending on the presence of antioxidants or antioxidative activity [25]. The absorbance of the gallic acid-DPPH aliquot at 40 ppm was measured to obtain the TPC in terms of mg GAE/g. In DPPH assays with gallic acid as the standard, a TPC value greater than 10 mg GAE/g is considered to have a high antioxidative capability [27]. *M. calabura* leaves are abundant in flavones, isoflavones, and polyphenols, which explain the high values for the TPC [28].

Induction time and AAI. The induction times of the samples were obtained through Rancimat analysis, while the AAI values of the samples were calculated based on the induction times. The induction time of cooking oil is one way to determine oxidative stability. The longer the induction time, the more stable the cooking oil is. The time indicates when the oil starts to break down at a certain temperature, in this case at 150 °C. The AAI explains the relative antioxidative activity between the control and the treated samples; the higher the index, the higher the antioxidant activity of the oil compared to the other [14].

Among the four set-ups, the set-up with 0.5% mass of extract shows the highest induction time and antioxidative index. Both induction time and AAI increased as the percent mass of extract added to PCO increased (0<0.1%<0.3%<0.5%). Meanwhile, the acrolein concentration decreased (0.5%<0.3%<0.1%<0) as shown in Table 2.

Table 2. Summary of mean values for induction time (IT), antioxidative activity index (AAI), and acrolein concentration (AC) of palm cooking oil with increasing percent mass of leaf extract. Data are expressed in terms of mean ± standard deviation (n = 4).

Extract (% Mass)	IT (h)	AAI	AC (ppm)
0	2.30 ± 0.20	1.00 ± 0.00	9.747 ± 0.67
0.1	2.51 ± 0.05	1.10 ± 0.09	8.120 ± 0.22
0.3	2.66 ± 0.06	1.16 ± 0.10	6.914 ± 0.48
0.5	2.78 ± 0.08	1.22 ± 0.08	5.262 ± 0.99

Each treatment group is significantly different (p<0.05) from the control group, which indicates that there is an increase in the oxidative stability of PCO as the mass percent composition of *M. calabura* leaf extract is increased. However, no significant difference (p>0.05) can be found from the pairwise comparisons of the 0.1% and 0.3% set-ups, and the 0.3% and 0.5% set-ups, as reflected in Figure 3.

Acrolein concentrations. To quantify the acrolein concentrations, HPLC analysis was performed on the water samples used for the Rancimat analysis. The acrolein concentrations from cooking oil fumes were observed to decrease as the concentration of extract is increased, as indicated in Table 2. Each pair of set-ups was also significantly different (p<0.05) from one another, as shown by Figure 3. Furthermore, an increase in the percent mass of extract by increments of 0.2% significantly decreased the acrolein

concentrations in the fumes of the oil, unlike the results obtained for induction time.

The approach of analyzing acrolein in PCO fumes through the use of the Rancimat is a novel method that utilizes the concept of polarity and solubility. The volatile oxidation products generated due to the lipid oxidation are directed by the air stream to the Rancimat vessels containing pure water [29]. Since acrolein and water are both polar, acrolein dissolves in water.

Fullana et al. [30] have investigated the level of aldehydes emitted at different times at 180 °C and 240 °C but were unable to analyze acrolein. Aside from their study, no studies have analyzed the aldehyde concentrations in fumes in cooking oil with antioxidant additives but such analysis was recommended [12,18].

The results of Endo et al. [31] show that acrolein in cooking oil is not formed from glycerol backbones in triacylglycerol but from methyl linolenate and methyl linoleate derived from polyunsaturated fatty acids, such as α-linolenic and linoleic acids, produced at high levels during heating. Thus, cooking with vegetable oils at high temperatures is not advisable as it induces the formation of acrolein. Furthermore, Da Silva and Pereira [12] formulated a reaction mechanism on how unsaturated fatty acids degrade to acrolein which provides several unstable singlet oxygens in intermediate reactions accompanied by the production of free radicals along the way. In the nucleophilic attack, antioxidants bind to singlet oxygen molecules to prevent the reduction of carboxylic acids to aldehydes. These antioxidants also bind to the free radicals produced due to oxidation, as shown in Figure 2 [12].

Comparing the values, it can be inferred that the induction time of oil is directly proportional to the antioxidant activity of the oil and inversely proportional to its acrolein emissions. This confirms the proposed association between the amount of natural antioxidants and the aldehyde emissions of the oil [12,18].

The basis for the oxidative stability of cooking oil majorly depends on its polyunsaturated fatty acid composition [32]. Hence, aside from the addition of antioxidants in cooking oil, the mixing of polyunsaturated fatty acid-rich oil with monounsaturated fatty acid-rich oil can be done to increase its oxidative stability [33]. The fatty acid composition of the oil is also the basis in determining the nature of volatile aldehyde formation [30,33]. The four parameters, namely fatty acid composition, antioxidant concentration, induction time, and volatile aldehyde concentration, are likely interrelated and can be the subject of further studies.

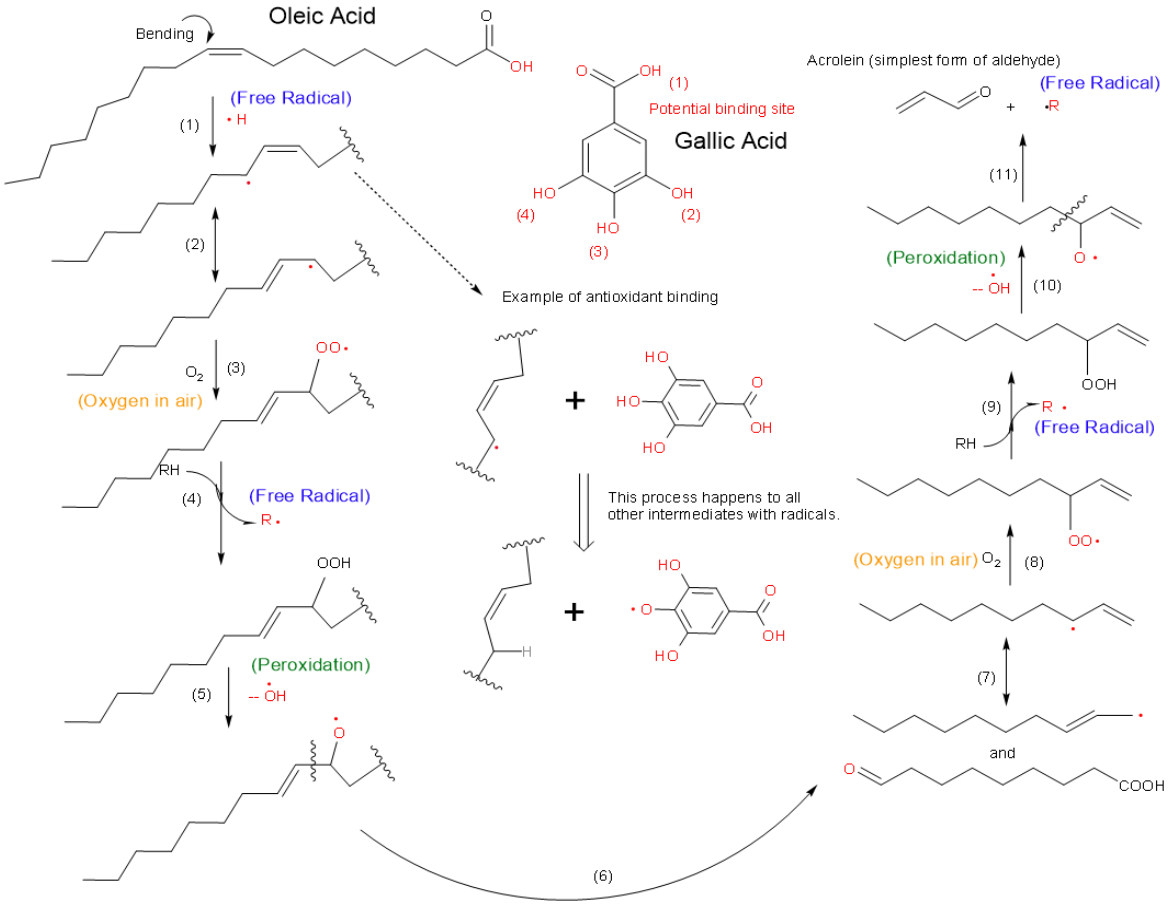

Figure 2. Reaction mechanism of the degradation of oleic acid—a monounsaturated fatty acid in palm cooking oil—into acrolein and the interaction of an antioxidant, gallic acid, in the prevention of such reaction [12].

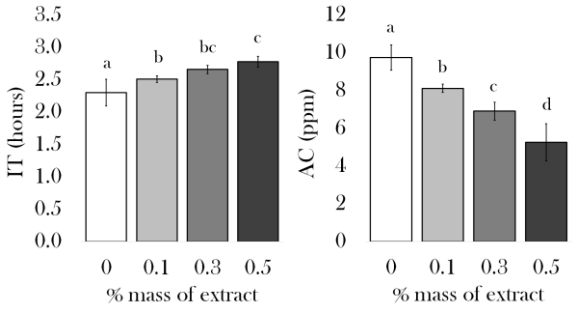

Figure 3. Induction time (IT) and acrolein concentrations (AC) in the fumes of palm cooking oil with varying % mass of *M. calabura* ethanol leaf extract. Data are presented as mean ± standard deviation (n=4). Values with different superscript letters represent significant differences among set-ups (p<0.05).

Limitations. The results of this study partially support the claims in the literature that the addition of antioxidants contributes to the reduction of aldehydes present in cooking oil fumes. However, the researchers were not able to identify specific antioxidants present in the extract. Second, the extract was not freeze-dried to maximize solubility in PCO. Third, the cooking oil used was already incorporated with butylated hydroxytoluene, a synthetic antioxidant. Fourth, other aldehydes aside from acrolein were not analyzed.

Conclusion. - Antioxidant capacity to reduce aldehyde emissions of palm cooking oil was investigated. Before treatment of the oil, it has been verified that *M. calabura* leaves contain high antioxidative activity, total phenolic content, and antioxidative activity index. Induction times of the treated oil samples increased, implying that oxidative stability increased. Hence, the addition of natural antioxidants, specifically *M. calabura* leaf extract, to palm cooking oil increases oxidative stability of the oil and reduces the aldehyde concentrations in palm cooking oil fumes. Significant differences were observed between the induction times of all pairs of set-ups except for the 0.1% and 0.3% pair and the 0.3% and 0.5% pair. Meanwhile, significant differences were observed between the acrolein concentrations of all pairs of set-ups (0 & 0.1%, 0 & 0.3%, 0 & 0.5%, 0.1% & 0.3%, 0.1% & 0.5%, and 0.3% & 0.5%). Lastly, this study can serve as a basis for the establishment of the correlation between the addition of antioxidants in cooking oil and its emission of aldehydes as fumes.

Recommendations. - It is recommended that similar studies are done with the use of gas chromatography-mass spectrometry which is more suited for volatile analytes. Other cooking oils, aldehydes, and sources of natural antioxidants should also be analyzed to compare the power and versatility of the natural antioxidants. Another recommendation is to test the treatments at varying higher temperatures and heating duration. Sensory analysis of the treated oil is also encouraged. Moreover, it is recommended to analyze the fatty acid composition and aldehyde concentrations of oil subjected to high heating temperatures to determine if the results are consistent with the aldehyde emissions. Lastly, antioxidant profiling and the determination of antioxidant binding ability with acrolein could be performed.

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References

[1] Harrison MR, Hester RE, editors. 2019. Indoor air pollution. [Internet]. The Royal Society of Chemistry; [cited 2020 Jan 27]. Available from: https://rsc.li/2lYHDjH.

[2] Chen HC, Wu CF, Chong IW, Wu MT. 2018. Exposure to cooking oil fumes and chronic bronchitis in nonsmoking women aged 40 years and over: a health-care based study. BMC Public Health. 18(246): 1–11. doi: 10.1186/s12889-018-5146-x.

[3] Katragadda HR, Fullana A, Sidhu S, Carbonell-Barrachina AA. 2010. Emissions of volatile aldehydes from heated cooking oils. Food Chem. 120: 59–65. doi: 10.1016/j.foodchem.2009.09.070.

[4] Neghab M, Delikhoon M, Baghani AN, Hassanzadeh J. 2017. Exposure to cooking fumes and acute reversible decrement in lung capacity. Int J Occup Env Med. 8(4):207–216. doi: 10.15171/ijoem.2017.1100.

[5] WHO (World Health Organization). 2018. [Cited 9 September 2019]. Available from https://bit.ly/3ekZASi.

[6] EPA (Environmental Protection Agency). 2019. [Cited 27 January 2020]. Available from https://bit.ly/2SrEuZT.

[7] Rivas I, Fussell J, Kelly F, Querol X. 2019. Indoor Sources of Air Pollutants. Issues Environ Sci Technol. 48: 1–15. doi: 10.1039/9781788016179-00001.

[8] Tham KW. 2016. Indoor air quality and its effects on humans — A review of challenges and developments in the last 30 years. Energy Buildings. 130: 637–650. doi: 10.1016/j.enbuild.2016.08.071.

[9] Haddad M. c2020. Coronavirus: How much more time are people spending at home? [Internet]. Aljazeera Media Network. [cited 2020 December 9]. Available from: https://bit.ly/3h6LHcb.

[10] Ghobadi S, Akhlaghi M, Shams S, Mazloomi SM. 2018. Acid and peroxide values and total polar compounds of frying oils in fast food restaurants of Shiraz, Southern Iran. Int J Nutr Sci. 3(1): 25–30.

[11] Lu F, Shen B, Yuan P, Li S, Sun Y, Mei X. 2019. The emission of PM2.5 in the respiratory zone from Chinese family cooking and its health effects.

Sci Total Env. 654: 671–677. doi: 10.1016/j.scitotenv.2018.10.397.

[12] Da Silva TO, Pereira PA. 2008. Influence of time, surface-to-volume ratio, and heating process (continuous or intermittent) on the emission rates of selected carbonyl compounds during thermal oxidation of palm and soybean oils. J Agric Food Chem. 56(9): 3129–3135. Doi: 10.1021/jf0734525.

[13] Guillaume C, De Alzaa F, Ravetti L. 2018. Evaluation of chemical and physical changes in different commercial oils during heating. Acta Scientific Nutritional Health. 2: 2–11.

[14] Metrohm. 2012. Oxidation stability of oils and fats – Rancimat method. Available from: https://bit.ly/3eYtMBU.

[15] Ali S, Chatha SAS, Ali Q, Hussain AI, Hussain SM, Perveen R. 2016. Oxidative stability of cooking oil blend stabilized with leaf extract of *Eucalyptus citriodora*. Int J Food Prop. 19(7): 1556–1565. doi: 10.1080/10942912.2015.1047514.

[16] Chowdhury K, Banu L, Khan S, Latif A. 2008. Studies on the fatty acid composition of edible oil. Bangladesh J Sci Ind Res. 42(3): 311–316. doi: 10.3329/bjsir.v42i3.669.

[17] Fujisaki M, Endo Y, Fujimoto K. 2002. Retardation of volatile aldehyde formation in the exhaust of frying oil by heating under low oxygen atmospheres. J Amer Oil Chem Soc. 79(9): 909–914. doi: 10.1007/s11746-002-0578-3.

[18] Shahid MZ, Saima H, Yasmin A, Nadeem MT, Imran M, Afzaal M. 2018. Antioxidant capacity of cinnamon extract for palm oil stability. Lipids Health Dis. 17(1): 1–8. doi: 10.1186/s12944-018-0756-y.

[19] Shahidi F, Zhong Y. 2010. Lipid oxidation and improving the oxidative stability. Chem Soc Rev. 39(11): 4067–4079. doi: 10.1039/B922183M.

[20] Taghvaei M, Jafari SM. 2015. Application and stability of natural antioxidants in edible oils in order to substitute synthetic additives. J Food Sci Technol. 52(3): 1272-1282. doi: 10.1007/s13197-013-1080-1.

[21] El-Roby AM, Hammad KSM, Galal SM. 2020. Enhancing oxidative stability of sunflower oil with sesame (*Sesamum indicum*) coat ultrasonic extract rich in polyphenols. J Food Process Preserv. e14564. doi: 10.1111/jfpp.14564.

[22] Recuenco MC, Lacsamana MS, Hurtada WA, Sabularse VC. 2016. Total phenolic and total flavonoid contents of selected fruits in the Philippines. Phil Journal Sci. 145(3): 275–281. doi: 10.1100/2012/728613.

[23] Singh A, Gupta R, Pandey R. 2017. Exogenous application of rutin and gallic acid regulate antioxidants and alleviate reactive oxygen generation in *Oryza sativa L*. Physiol Mol Biol Plants. 23(2): 301–309. doi: 10.1007/s12298-017-0430-2.

[24] Y MAK, Mundasada SC, Ramadas D. 2015. Antioxidative activity: root, leaves and fruits aqueous extracts of *Muntingia calabura*. JIPBS. 2(4): 363–368.

[25] Mohamed S, Mohd NF, inventors; University Putra Malaysia, Selangor, assignee. 2013. Cooking oil composition with additives to reduce oil adsorption. United States Patent Application No. 8.425,969 B2.

[26] Kedare SB, Singh RP. 2011. Genesis and development of DPPH method of antioxidant assay. J Food Sci Technol. 48(4): 412–422. doi: 10.1007/s13197-011-0251-1.

[27] Mahmood ND, Nasir NLM, Rofiee MS, Tohid SFM, Ching SM, Teh LK, Salleh MZ, Zakaria ZA. 2014. *Muntingia calabura*: a review of its traditional uses, chemical properties, and pharmacological observations. Pharm Biol. 52(12): 1598–1623. doi: 10.3109/13880209.2014.908397.

[28] Haerani A, Chaerunisa AY, Subarnas A. 2019. Antioxidant Activities of *Muntingia calabura*, *Syzygium cumini*, *Ocimum basilicum*, and *Eleutherine bulbosa* using DPPH Method. Indonesian Journal of Pharmaceutics. 1(2): 50–54. doi: 10.24198/idjp.v1i2.21531.

[29] Tinello F. Lante A, Bernardi M, Cappiello F, Galgano F, Caruso MC, Favati F. 2018. Comparison of OXITEST and RANCIMAT methods to evaluate the oxidative stability in frying oils. Eur Food Res Technol. 244(4): 747–755. doi: 10.1007/s00217-017-2995-y.

[30] Fullana A, Carbonell-Barrachina AA, Sidhu S. 2004. Comparison of volatile aldehydes present in the cooking fumes of extra virgin olive, olive, and canola oils. J Agric Food Chem. 52(16): 5207–5214. doi: 10.1021/jf035241f.

[31] Endo Y, Hayashi C, Yamanaka, T, Takayose K, Yamaoka M, Tsuno T, Nakajima S. 2013. Linolenic acid as the main source of acrolein formed during heating of vegetable oils. Journal of the American Oil Chemists' Society. 90(7): 959–964. doi: 10.1007/s11746-013-2242-z.

[32] Bhatnagar AS, Prasanth Kumar PK, Hemavathy J, Gopala Krishna AG. 2009. Fatty acid composition, oxidative stability, and radical scavenging activity of vegetable oil blends with coconut oil. J Am Oil Chem Soc. 86(10): 991–999. doi: 10.1007/s11746-009-1435-y.

[33] Maszewska M, Florowska A, Dłużewska E, Wroniak M, Marciniak-Lukasiak K, Zbikowska A. 2018. Oxidative stability of selected edible oils. Molecules. 23(1746): 1–12. doi: 10.3390/molecules23071746.