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RESEARCH ARTICLE

Effect of Repeated Deep Frying on Oxidative Stability and Quality of Unrefined Bulk Coconut Oil

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Highlights

- Coconut oil reduced its physicochemical quality and oxidative stability with increasing frying cycles
- Negative quality and safety aspects of coconut oil should be considered when the oil is reused for deep frying
- Coconut oil can be reused for a maximum of 2-4 frying cycles, maintaining its quality and safety requirements specified by the Codex and Sri Lankan food standards

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Effect of Repeated Deep Frying on Oxidative Stability and Quality of Unrefined Bulk Coconut Oil

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Abstract

Although repeated use of edible oils for deep frying can adversely alter their physicochemical properties, detailed studies examining these effects on coconut oil remain limited. In this study, the effect of repeated deep frying of unrefined copra coconut oil sold in bulk was evaluated for its selected quality parameters. For this purpose, coconut oil was heated at 180±5°C for 20 min with food (*vadai*) as a frying treatment, or without food as a frying control, for ten repeated frying cycles. After each frying cycle, the oil samples were analyzed for oxidative stability, physicochemical properties, and structural alterations using standard methods. The levels of free fatty acids and thiobarbituric acid reactive substances (TBARS) in fried coconut oil samples increased significantly (p<0.05), while their saponification and iodine values decreased significantly (p<0.05), with increasing number of frying cycles. Moreover, the peroxide value of coconut oil heated without food showed a significant (p<0.05) increment over all successive frying cycles tested. However, it increased significantly (p<0.05) only up to the sixth frying cycle and a significant (p<0.05) reduction was observed thereafter. The mean FFA content of coconut oil samples heated repeatedly with food was significantly (p<0.05) higher than that of samples heated without food. This indicates that repeated heating of unrefined coconut oil has a significant (p<0.05) adverse effect on its oxidative stability and physicochemical properties, compared to the accepted limits. However, unrefined coconut oil can be used for a maximum of 2-4 repeated deep frying cycles, while maintaining its physicochemical parameters within the limits specified by the Codex and Sri Lankan food standards.

Keywords: Deep frying; Hydrolysis; Oxidation; Peroxidation; Unrefined coconut oil

INTRODUCTION

Fried foods are widely popular among the most frequently consumed foods worldwide due to their appealing flavor, color, and crispy texture (Dangal et al., 2024). Deep frying has been one of the most commonly used methods of food processing, since it develops unique sensory characteristics (Rani et al., 2023). During deep frying, food is fully immersed in heated oil at temperatures ranging from 150 to 190°C, allowing interactions among oil, air, and the food (Asokapandian et al., 2020; Dehghannya & Ngadi, 2021). During deep frying, the frying oil deteriorates as a result of the migration of water vapor and various compounds from the food into the oil, together with prolonged exposure to high frying temperatures (Valle et al., 2024).

The key deteriorative chemical reactions that occur in repeatedly used frying oils include hydrolysis, thermal oxidation, isomerization, and polymerization (Dodoo et al., 2022; Sivaranjani et al., 2024). These changes contribute to the loss of essential fatty acids and the development of undesirable flavors, colors, and odors in both the oil and the fried foods, thereby reducing their palatability (Shahidi & Hossain, 2022). Oxidative reactions are responsible for the most significant changes that occur in frying oils, leading to the formation of a variety of primary and secondary oxidation products, including peroxides, hydroperoxides, aldehydes, ketones, acids, and alcohols (Geng et al., 2023a; Musakhanian et al., 2022). These compounds not only negatively affect the sensory quality of fried foods but also pose potential hazards to human health and have adverse nutritional implications (Vieira et al., 2017; Wang et al., 2023).

Sri Lanka is the eighth-largest coconut producer in the world, with coconut cultivation covering approximately 455,000 hectares (Annual Report, 2016). Moreover, Sri Lanka has the highest per capita coconut consumption in the world, with an average annual consumption of approximately 110 coconuts per person. According to previous studies, coconut and coconut oil are the primary dietary sources of fat for most Sri Lankans, contributing approximately 22% of the

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total daily energy intake (Jayasekara, 2004). The country had produced around 31,000 MT of coconut oil in 2024, based on combined data from the Coconut Development Authority and the Department of Census and Statistics of Sri Lanka (Central Bank of Sri Lanka, 2025). The abundant availability of coconuts has made coconut oil the most widely consumed edible oil in Sri Lanka. Unrefined coconut oil sold in bulk is the main type of coconut oil consumed by the majority of households in Sri Lanka, which account for approximately 70% of the country's population. The use of unrefined coconut oil for deep frying has gained increasing popularity as an alternative to oils containing trans-fatty acids because of growing concerns regarding their adverse health effects. Coconut oil is regarded as one of the most stable edible oils for deep frying due to its high content of saturated fatty acids, which enhances its resistance to oxidation and thermal degradation compared with many other edible oils (Habarakada et al., 2021).

In Sri Lanka, *vadai,* a deep-fried snack made out of soaked, ground lentils mixed with some condiments, is one of the most popular street foods among local people and visitors. It is also frequently prepared in many households in Sri Lanka. For the preparation of *vadai* and other fried foods, many street vendors use unrefined coconut oil sold in bulk. To minimize operating costs, this oil is often used repeatedly, particularly in response to the rising cost of edible oils. In addition, some food industries like fast food restaurants, hotels, *etc.,* and many households also use frying oils repeatedly. This has become a common practice due to the low level of awareness among the public about the negative health and nutritional effects caused by the compounds generated during repeated deep frying.

There are shreds of evidence which emphasize the negative effects of reusing frying oils on the sensory, nutritional quality of fried food and the safety concerns affecting public health. Nevertheless, there are no well-established guidelines specifying the maximum number of frying cycles for which coconut oil can be safely reused, taking into account the chemical changes that take place in the oil. Therefore, it is essential to investigate the physicochemical changes that occur during repeated deep frying to monitor oil quality and ensure the safety of fried foods. Such information will also help increase awareness among the public, street vendors, and the food industry regarding the adverse effects of repeatedly reused frying oil on food quality and human health. Furthermore, identifying the optimal number of frying cycles will facilitate the efficient use of coconut oil while maintaining the safety and quality of fried foods.

To the best of our knowledge, no recent studies have systematically evaluated the physicochemical changes occurring in unrefined coconut oil during repeated heating in the absence and presence of food. As the composition of the fried food influences the rate and extent of oil deterioration, it is an important factor that should be considered when evaluating changes in frying oils. Given the widespread use of unrefined coconut oil for deep frying in Sri Lanka and the high consumption of *vadai* as a popular street food, investigating the physicochemical changes occurring in coconut oil during repeated frying is of considerable importance. Therefore, the present study aimed to investigate the physicochemical changes in unrefined coconut oil obtained from five oil-processing mills in the Western and North Western Provinces of Sri Lanka during repeated heating without food and repeated frying of *vadai*. In addition, the study sought to determine the maximum number of frying cycles for which unrefined coconut oil can be safely reused for frying *vadai* while maintaining acceptable food quality and safety. This research provides novel insights into the quality, stability, and safety of unrefined coconut oil under repeated deep frying conditions in both the absence and presence of food, thereby generating evidence to support recommendations for its safe and efficient use in domestic, street-vending, and commercial food preparation.

MATERIALS AND METHODS

A total of five coconut oil (copra oil) samples without any adulteration, of each 5 L, were directly purchased from five different coconut oil processing mills located in the Western and North Western provinces of Sri Lanka. All the ingredients needed to prepare *vadai* batter were purchased from a local market. All chemicals used in this study were of analytical grade with the highest purity available (> 99.5 %) and obtained from Sigma-Aldrich, USA.

Preparation of *vadai* batter

A standard formulation of *vadai* batter was maintained for the repeated deep frying in all coconut oil samples. Every day, 1 kg of chickpeas were taken and soaked in 1.5 L of water for 2 h. Then the water was drained off, and the chickpeas were washed well. The chickpeas were ground into small particles using a grinder for about 3 min. Then 200 g of chopped onion, 30 g of dried red chilies, 15 g of cumin seeds, and 20 g of curry leaves were ground together using a grinder for 1 min. This ground mixture and 10 g of salt were added to the ground chickpeas and mixed well. This batter was then used for preparation of *vadai* of approximately uniform size, each weighing about 15±0.5 g.

Heating of oil

Five samples of unrefined copra coconut oil sold in bulk (S1, S2, S3, S4 and S5) were collected from the local market and divided into two equal volumes of 2.5 L. The deep-frying experiment was done as two parts. First was heating of oil without food and the second was heating of oil with food (*vadai*). In both, repeated heating of coconut oil was carried out in a stainless steel frying pan. A constant temperature was maintained throughout the heating process after heating the oil to a temperature of 180±5°C and heating was continued exactly for 20 min. When heating the oil with food, the prepared *vadai* batter was introduced into each coconut oil samples after bringing

the temperature to 180±5°C and the ratio of oil to *vadai* batter was maintained as 10:3 (V/W).

All coconut oil samples were reused for nine more frying cycles (ten frying cycles in total), with intervals of 10-14 h between cycles. Following each heating cycle, the oil was allowed to cool to room temperature and was stored in airtight glass bottles with a headspace of 2-3 cm. Triplicate samples (150 mL) were drawn at the end of each second, fourth, sixth and tenth cycle of repeated heating of all coconut oil samples without and with food in glass bottles. The sample bottles were tightly sealed and stored in a refrigerator at 4 °C for five days until all frying cycles were completed, prior to chemical analysis. A control sample (150 mL) devoid any heating or frying process was also collected as triplicate and it was used for comparative purpose.

The oxidative and physicochemical changes in the coconut oil after each frying cycle was assessed by determining free fatty acid content (FFA), saponification value (SV), iodine value (IV), peroxide value (PV), thiobarbituric acid reactive substances (TBARS), trans-fats, and oil color.

Determination of free fatty acid (FFA) content

The content of FFA was determined according to the AACC Method 58-15 (AACC 2000; Manchanda & Passi, 2016). Each coconut oil sample of 5-6 g was measured into a 250 mL Erlenmeyer flask. Amount of 50 mL neutralized ethanol was added and boiled on water bath (WNB14, Schwabach FRG, Germany) for 5 min. Approximately 1 mL phenolphthalein indicator was added and this solution was titrated with standard alkali (0.1 N, NaOH) while shaking vigorously until the endpoint had reached. The end point was indicated by a slight pink color that had persisted for 30 s. The volume of the sodium hydroxide used for the titration was recorded. The content of FFA was expressed as, g lauric acid per 100 g of oil sample. Free fatty acid content was calculated by using the equation (1).

$$\text{FFA (g lauric acid/100g)} = \frac{\text{V x N x 200.3 x 100}}{\text{W}} \quad (1)$$

Where, V: volume of NaOH titrant (mL), N: normality of NaOH titrant (mol/1000 mL), 200.3: molecular weight of lauric acid (g/mol) and W: sample mass (g)

Determination of saponification value (SV)

Saponification value of oil was measured according to the AOAC Official Method 920.160 with slight modification (AOAC, 2010a). Each oil sample was accurately weighed (5 g) into the 250 mL round bottom flask. Accurately, 50 mL of ethanolic potassium hydroxide was added into the flask. Next, pumice beads were added into the flask and the flask was connected to a condenser. This solution was boiled gently but steadily on a sand bath for 30-60 min until the sample was clear and homogenous indicating complete saponification. The sample was allowed to cool and the inside of the condenser was washed down with a jet of distilled water. The flask was disconnected from the condenser and the sample was allowed to cool down to room temperature. Then 1 mL of phenolphthalein was added to sample and this solution was titrated with 0.5 M, hydrochloric acid until the pink color just disappeared. The volume of titrant used was recorded. Blank determination was conducted along with that on test portion, using the same pipet for measuring potassium hydroxide solution and refluxing for the same time as used for the sample. Saponification value was expressed as milligrams of potassium hydroxide required to saponify 1 g fat and it was calculated using the equation (2).

$$\text{Saponification value} = \frac{(\text{B} - \text{S})\text{x N x } 56.1}{\text{W}} \quad (2)$$

Where, B: volume of the titrant (mL) for blank, S: volume of titrant (mL) for sample, N: normality of HCl (mmol/mL), 56.1: molecular weight of KOH (mg/mmol) and W: sample mass (g).

Determination of iodine value (IV)

The iodine value was determined according to Diop et al, (2014), which was based on ISO 3961, (2009). Each oil sample was filtered through filter paper to remove impurities. The sample of approximately 2.5 g was weighed into a 500 mL glass stoppered flask. Chloroform (10 mL) was added into the flask to dissolve the oil and the blank was prepared by adding 10 mL chloroform into 500 mL glass stoppered flask. Accurately 25 mL Wijs solution (Techno Pharmchem, India) was pipetted into the flask and the flask was allowed to stand for 30 min in the dark. After incubation period in the dark, 20 mL of potassium iodide solution (15%) was added into the flask and shaken vigorously. Then 100 mL freshly boiled and cooled water was added into the flask washing down the free iodine on the stopper. The iodine in the flask was titrated with standard sodium thiosulphate (0.1N) by adding gradually with constant and vigorous shaking until the color turned to pale yellow. Approximately 1-2 mL of starch indicator was added and the titration was continued until the blue color disappeared. The flask was stoppered and shaken vigorously toward the end of the titration so that any iodine remaining in chloroform could be taken up by the potassium iodide solution. The volume of titrant used was recorded and the blank titration was done with the solvent devoid of oil sample. Iodine value was expressed as the number of grams of iodine absorbed by 100 g of oil and it was calculated by using the equation (3).

$$\text{Iodine value} = \frac{(\text{B} - \text{S})\text{x M x } 126.9 \text{ x } 100}{\text{W}} \quad (3)$$

Where, B: volume of titrant (mL) for blank, S: volume of titrant (mL) for sample N: normality of $Na_2S_2O_3$, 126.9: molecular weight of iodine and W: sample mass (g).

Determination of peroxide value (PV)

Peroxide value of oil was measured according to the AOAC Official Method 965.33 (AOAC, 2010b). Coconut

oil (5±0.05 g) was transferred into a 250 mL stoppered Erlenmeyer flask. Amount of 20 mL acetic acid: chloroform solution (3:2) was added and the flask was swirled until the oil sample was completely dissolved. Then 0.5 mL of saturated potassium iodide solution was added using a pipette. The flask was stoppered, shaken gently and kept in the dark for 1 min with occasional shaking. Then 30 mL of distilled water was added, stoppered and shaken. The liberated iodine was slowly titrated with 0.1 N sodium thiosulphate solution with vigorous shaking until the yellow color was almost gone. About 0.5 mL of starch indicator was added and the titration was continued while shaking vigorously to release all iodine from chloroform layer until blue color disappeared. The blank determination also conducted. Peroxide value was expressed as milli equivalent of peroxide oxygen per kg sample (meq/kg). The peroxide value was calculated by using the equation (4).

$$\text{Peroxide value} = \frac{(S - B)x\ N\ x\ 1000}{W} \quad (4)$$

Where, S: volume of titrant (mL) for sample, B: volume of titrant (mL) for blank, N: normality of $Na_2S_2O_3$ solution (mEq/mL), W: sample mass (g) and 1000: conversion of units (g/kg).

Quantification of 2-Thiobarbituric acid reactive substances (TBARS)

The assay was conducted according to the method described by Kampa et al, (2022) with some modifications. Each oil sample of 2±0.05 g was weighed into a centrifuge tube and 5 mL of 10% (W/V) trichloro acetic acid (TCA, Sigma-Aldrich, USA) was added. This solution was vortex (Super-Mixer, USA) at highest speed for 2 min. Amount of 5 mL of 0.02 M aqueous solution of 2-thiobarbituric acid (TBA, HiMedia Laboratories, India) was added and vortexed (Super-Mixer, USA) for 30 s. This solution was centrifuged (Himac CT 4D, Hitachi, Japan) at 3000x*g* for 10 min. The supernatant was filtered through a Whatman no.3 filter paper and the filtrate was placed in a boiling water bath (WNB14, Schwabach FRG, Germany) for 45 min. Then it was cooled to room temperature on ice and the absorbance at 532 nm of the filtrate was measured using UV-Visible spectrophotometer (Multiskan GO, Thermo Fisher Scientific, Finland).

Standard curve was prepared using 1, 1, 3, 3 - Tetramethoxy propane (TMP) which is the precursor of malondialdehyde (MDA). First 25 µM of TMP was prepared by adding 41.1 µL of the concentrated TMP to 10 mL distilled water. Then a concentration series was prepared by using 25 µM of TMP, 10% TCA and 0.02 M TBA and the same procedure was followed for the oil samples. After measuring the absorbance standard curve was drawn for absorbance against the concentration of samples and the slope of the standard curve was estimated. Then TBARS values of coconut oil samples were calculated by using the equation (5).

$$\text{MDA in sample} = \left(\frac{\text{Absorbance of sample}}{\beta}\right) x \left(\frac{\text{Total volume of sample}}{\text{Sample weight (g)}}\right) \quad (5)$$

Where, β: slope of the standard curve and total volume of sample = volume of sample + volume of TCA (5 mL).

Quantification of trans-fats

Trans-fat content was analyzed according to the AOCS Official Method Ce 1j-07 (AOCS, 2017a). Fatty acid methyl esters in the oil samples were prepared following the AOCS Official Method Ce 2-66 (AOCS, 2017b). Test portion of oil sample (approximately 0.1 g) was accurately measured into a stoppered glass centrifuge vial. Then 0.5 mL of methanolic potassium hydroxide (2 N) and 7 mL of heptanes (assay >99%, ROMIL LTD, UK) were added respectively. The vial was closed and put on a vortex machine (2 min, 2500 rpm). An amount of 0.5 mL of TAG 13 (Internal standard) was added into it and put on vortex machine (30 s, 2500 rpm). The vial with content was allowed to stand for 30 min and 0.5 mL of upper layer was transferred into a labeled stoppered glass vial. The labeled, stoppered glass vials were placed on the respective cells of the Gas Liquid chromatogram (GLC 14A, Shimadsu, Japan) and chromatogram was run using the software.

Determination of color of the oil

The color of the oil was measured using spectrophotometric method as described by Elisaveta and Pavel, 2014), which was based on AOCS Official Method Cc 13c (AOCS, 1989). The oil samples were brought to 25-30°C and placed in a microplate. The absorbance of the oil was read at the wave lengths of 460, 550, 620 and 670 nm using spectrophotometer (Multiskan GO, Thermo Fisher Scientific, Finland). Then the photometric color index of oil samples was calculated by using the equation (6).

$$\text{Photometric color index} = 1.29(A_{460}) + 69.7(A_{550}) + 41.2(A_{620}) - 56.4(A_{670}) \quad (6)$$

Where, (A_{460}) : absorbance at 460 nm, (A_{550}): absorbance at 550 nm, (A_{620}): absorbance at 620 nm and (A_{670}): absorbance at 670 nm.

Statistical analysis

The data were analyzed using MS Office 2007 (Excel) and SAS software version 9.0. All measurements were performed in triplicate and results are expressed as mean ± SD. The ANOVA tables were constructed using nested design and PROC GLM procedure. Duncan's multiple range test was used to determine significant differences at 0.05 significant levels.

RESULTS AND DISCUSSION

Physicochemical changes taken place in the oil during heating

Heating of oil induces several physicochemical changes, especially when it is subjected to high temperatures for prolonged periods. The composition of the oil is changed when bonds are broken and new bonds are formed (Wang et al., 2021). Further, lipids easily undergo oxidation which leads to deterioration of the quality and oxidative stability of lipids (Dragoev, 2024). To analyze physicochemical changes in oils during heating a variety of analytical methods are used. In this study, free fatty acid content, saponification value, and iodine value were determined as chemical changes. The level of oxidation was determined by quantifying the primary and secondary oxidative products generated in the oil sample. In this research, peroxide value was used to estimate the primary oxidation products while determination of TBARS was used to estimate the secondary oxidative products.

Effect of frying on FFA content

The content of FFA reflects the amount of fatty acids removed from triacylglycerol structure through hydrolysis and it is one of the indicators of oil deterioration, which mainly measures the degree of hydrolysis. The Sri Lanka Standards (SLS 32:2002) and the Codex recommendations for the maximum allowable FFA content in quality coconut oil are 0.8% and 0.3% (g lauric acid per 100 g oil) respectively (CODEX, 2001; SLS, 2002). The initial FFA content of the coconut oil sample used in this study was 0.29±0.02%, thus it complies with these standards.

Figure 1 shows the changes in the FFA contents in the coconut oil samples over the repeated heating of oil without and with food. It was observed that the FFA contents in all coconut oil samples had increased significantly (p<0.05) with the number of frying cycles in both oils heated with food and without food. Similar results have been reported in previous studies observing accumulation of FFAs with increasing duration of deep-frying (Ben Hammouda et al., 2017; 2018). Further, a study has reported that the FFAs can be formed as secondary oxidation products by the decomposition of primary oxidation products of lipid oxidation, mainly the hydroperoxides (Han et al., 2023). Therefore, the formation of FFAs in the oil heated without food is primarily attributed to oxidative degradation, as there is no opportunity for water to be introduced into the oil to facilitate hydrolytic reactions. However, hydrolytic reactions may still occur due to the moisture generated during other oil deterioration reactions and the inherent moisture present in unrefined coconut oil. In contrast, when oil is heated in the presence of food, hydrolysis of triacylglycerols is further promoted by the water released from the food, resulting in the formation of FFAs in addition to those produced through oxidative degradation. Sample 5 (S5) had a significantly (p<0.05) higher FFA content than the other oil samples tested. This may be attributed to excessive heat treatment during processing or the retention of excess moisture in the oil as a result of improper processing.

The comparison of the mean FFA contents in the coconut oil samples (excluding the significantly different sample S5) over repeated frying cycles with and without food is shown in the Figure 2. The mean FFA content of coconut oil samples heated repeatedly with food was significantly higher (p<0.05) than that of samples heated without food (*vadai*). This may be attributed to the enhanced hydrolytic degradation of triacylglycerols in the presence of water released from the food, together with the accelerated oxidative degradation of the oil during repeated heating, resulting in a greater accumulation of FFAs. A previous study further explained that water acts as a weak nucleophile, causing the cleavage of the ester bonds in triacylglycerols and resulting in the formation of free fatty acids, along with monoacylglycerols and diacylglycerols (Soodoo et al., 2023).

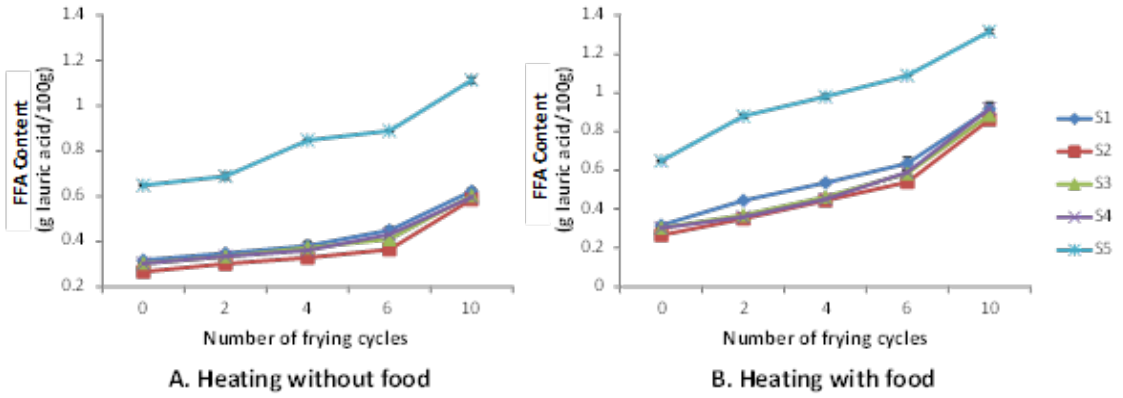


Figure 1: Changes in the FFA content of tested coconut oil samples during repeated heating without food (A) and with food (B). Five coconut oil samples (S1, S2, S3, S4 and S5) were repeatedly heated (at 180±5°C for 20 min) without food and with food (*vadai*) for ten cycles, allowing cool down to room temperature after each frying. The FFA content in the oil was determined after selected frying cycles (0, 2, 4, 6 and 10). The FFA contents in all coconut oil samples had significantly (p<0.05) increased with the increasing number of frying cycles. Sample 5 had significantly (p<0.05) higher FFA content compared to all other samples throughout

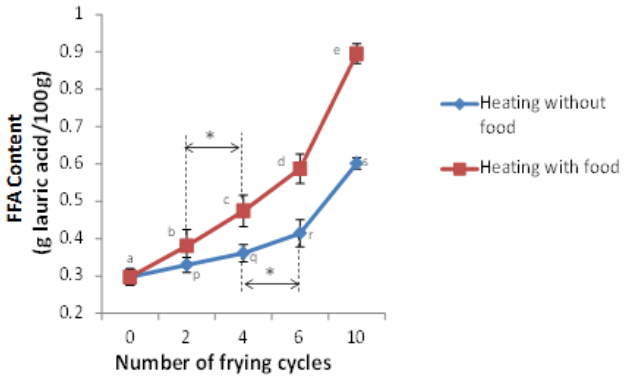


Figure 2: Comparison of mean FFA content in the coconut oil samples during repeated heating with and without food. The mean±SD FFA contents of coconut oil samples (S1, S2, S3 and S4) excluding the significantly different sample S5 were calculated at the selected number of frying cycles (0, 2, 4, 6 and 10). The mean±SD value of FFA content of coconut oil samples heated with food was significantly (p<0.05) higher than that of samples heated without food. Different letters within a series show significant difference at (p<0.05). The mean±SD values also had significantly (p<0.05) increased with the number of frying cycles in both samples heated with and without food

Effect of frying on SV

The SV is the number of milligrams of potassium hydroxide required to saponify 1 g of oil or fat. Because oils containing shorter-chain fatty acids have a greater number of ester bonds per unit mass, the SV is inversely proportional to the average molecular weight of the triacylglycerols and the average chain length of their constituent fatty acids (Jain et al., 2020; Doris, 2024). The SLS (SLS 32:2002) and the Codex recommended standards for SV of coconut oil are 248-264 and 248-265 mg KOH/g respectively (SLS, 2002; CODEX, 2001). The mean SV of coconut oil samples evaluated in this study was 253.46±8.23mg KOH/g, and it was within the acceptable range as per the quality standards.

According to the Figure 3, it was observed that the SV in each coconut oil sample had decreased significantly (p<0.05) with the increasing number of frying cycles in the presence or absence of food. Overall, there were no significant (p>0.05) difference among the SVs of five tested coconut oil samples after each frying cycle heated with or without food. Since the SV is an indirect measure of the average molecular weight of triacylglycerols in oil, the observed decrease in SV during repeated heating of coconut oil with and without food may be attributed to the hydrolytic and oxidative degradation of triacylglycerols and also formation of higher-molecular-weight polymerization products.

Figure 4 illustrates the comparison of the mean SV of all five coconut oil samples over repeated heating, in the presence and absence of the food. The mean SV of oil heated with food was lesser than that of oil heated without food because when heating the oil with food, both hydrolysis and oxidation contribute to the breakage of the ester bond. However, this difference was not significant (p> 0.05), suggesting that the contribution of hydrolysis to the cleavage of ester bonds in the presence of moisture released from the food was relatively small compared with that of oxidative degradation. Moreover, the polymerization of fatty acids, together with oxidative and hydrolytic reactions during repeated heating, may have contributed to the differential changes in the SV of oils heated with and without food.

Effect of frying on IV

The IV is a measure of the degree of unsaturation or the average number of double bonds in fats and oils (Samanta et al., 2023). The SLS (SLS 32:2002) and Codex recommendations for the IV of quality coconut oil are 7.5-9.5 and 6.3-10.6 g/100g I_2 respectively (CODEX, 2001; SLS, 2002). The average initial IV of the coconut oil samples used in this study was 7.68±0.05 g/100g I_2, and complied with the quality standards.

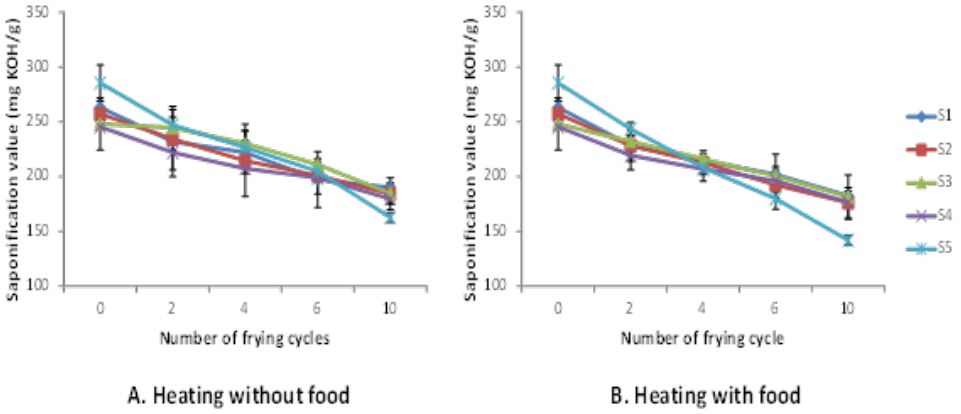


Figure 3: Changes in the SA in the coconut oil samples during repeated heating of oil without food (A) and with food (B). Five coconut oil samples (S1, S2, S3, S4 and S5) were repeatedly heated (at 180±5°C for 20 min) without food and with food for ten cycles allowing cool down to room temperature after each frying. Saponification value of the oil was determined after selected frying cycles (0, 2, 4, 6 and 10). The SV of all coconut oil samples had significantly (p<0.05) decreased with the number of frying cycles. No significant (p>0.05) difference was observed in the SVs among all coconut oil samples throughout

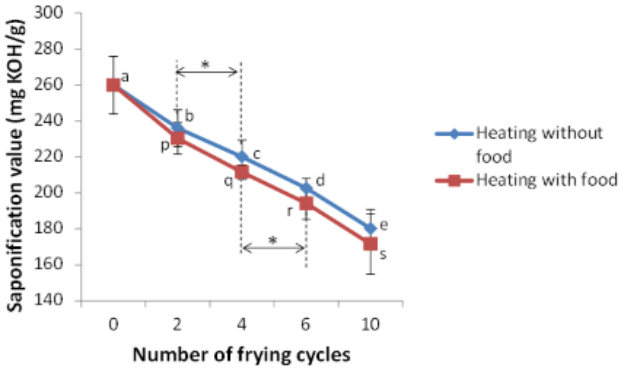


Figure 4: Comparison of mean saponification value in the coconut oil samples over repeated heating with and without food. The mean±SD of saponification values of coconut oil samples (S1, S2, S3, S4 and S5) were evaluated at the selected number of frying cycles (0, 2, 4, 6 and 10). Even though mean±SD values of coconut oil samples heated with food is lower than that of samples heated without food, it was not significant (p>0.05). Different letters within a series indicate significant differences at (p<0.05). The mean±SD SV also had significantly (p<0.05) decreased with the number of frying cycles in both oil samples heated with and without food

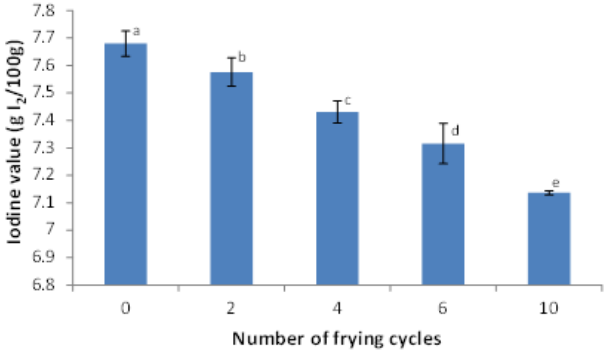


Figure 5: Changes in the IV in the coconut oil samples over repeated frying of food. Five coconut oil samples (S1, S2, S3, S4 and S5) were repeatedly heated (at 180±5°C for 20 min) with food (*vadai*) for ten cycles allowing cool down to room temperature after each frying. Iodine values in the oil were determined after selected frying cycles (0, 2, 4, 6 and 10). The mean±SD of IVs of samples (S1, S2, S3 and S4) excluding the significantly different sample S5, were evaluated at each selected number of frying cycles. Different letters indicate significant difference at p<0.05. Mean±SD of IVs of all coconut oil samples had significantly (p<0.05) decreased with the increasing number of frying cycles

The change in the IV of coconut oil during repeated deep frying of *vadai* is shown in Figure 5. It was observed that there was a significant (p<0.05) reduction of IV in coconut oil with the increasing number of repeated frying cycles. The IV of the coconut oil sample before any frying process and after the next immediate cycle of frying were not significantly (p>0.05) different, indicating the longer induction period of coconut oil. The IV after the fourth frying cycle was significantly (p<0.05) lower than that after the second frying cycle. The IV value after fourth and sixth cycle of frying were not significantly (p>0.05) different. However, the IV after the tenth cycle was significantly (p<0.05) lower than that after the sixth cycle of repeated frying.

The reduction in the IV of coconut oil during repeated heating can be explained by the decrease in the degree of unsaturation, or the number of double bonds in fatty acids, resulting from the oxidation and polymerization of the oil. In previous studies similar observations have been reported due to oxidation of oil, (Choudhary et al., 2015; Otache et al., 2022; Geng et al., 2023b). Furthermore, the decrease in the IV observed during repeated frying can be attributed to a reduction in the degree of unsaturation of fatty acids caused by complex physicochemical reactions, other than oxidation and polymerization, that occur during thermal processing (Jurid et al., 2020).

Effect of frying on PV

During deep frying, lipid oxidation initially leads to the formation of lipid hydroperoxides, commonly quantified as PV. These hydroperoxides are the primary oxidation products, formed through the oxidation of unsaturated fatty acid moieties in triglycerides, and are inherently unstable, readily decomposing into secondary oxidation products. (Longanesi et al., 2022). The Codex recommended standard for PV of coconut oil is not more than 15 mEq per kg (CODEX, 2001). The initial average PV of the coconut oil samples used in this study was 3.36±0.95 mEq per kg, and it is within the accepted level of Codex standards.

Figure 6 illustrates the changes in the peroxide value of the coconut oil samples over repeated frying without and with food. Results revealed that PV had increased significantly (p<0.05) with the increasing number of frying cycles in all coconut oil samples heated without food, indicating a gradual oxidation of oils with time. Moreover, the PV of coconut oil samples S2 and S4 had also increased significantly (p<0.05) with the increasing number of frying cycles in the presence of food. The increase in the PV over the number of frying cycles could be due to the formation of hydroperoxides as primary oxidation products. In contrast, the PV of coconut oil samples S1, S3, and S5 increased significantly (p<0.05) up to the sixth frying cycle and subsequently decreased significantly. This observation is consistent with previous reports and can be attributed to the thermal decomposition of unstable lipid hydroperoxides, the primary oxidation products, into secondary oxidation products such as aldehydes, ketones, and alcohols during prolonged frying (Atamaleki et al., 2022; Choe & Min, 2007).

The comparison of the mean PV of all coconut oil samples over repeated heating cycles with and without food is shown in Figure 7. By comparing the mean PV of oil heated with and without food, it was found that the means were not significantly (p>0.05) different. The mean PV of oil samples heated with food was comparatively higher

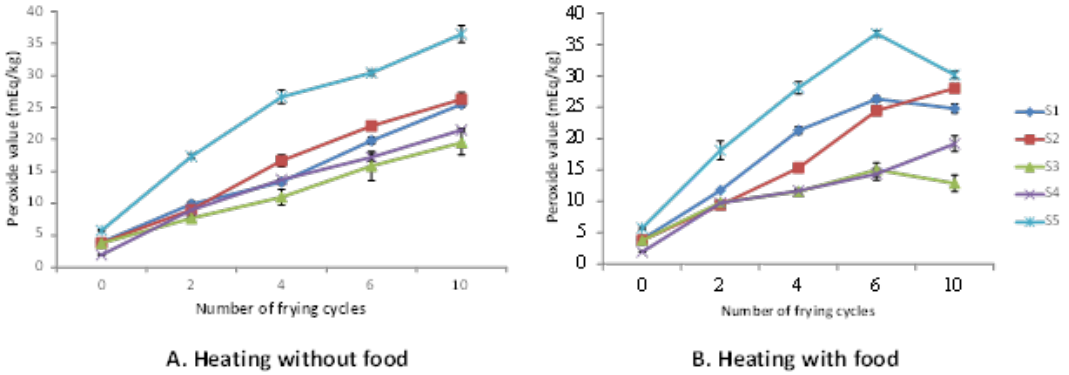


Figure 6: Changes in PV of the coconut oil samples over repeated heating of oil without food (A) and with food (B). Five coconut oil samples (S1, S2, S3, S4 and S5) were repeatedly heated (at 180±5°C for 20 min) without food and with food (*vadai*) for ten cycles allowing cool down to room temperature after each frying. The PV of the oil was determined after selected frying cycles (0, 2, 4, 6 and 10), and the oil samples heated without food had significantly (p<0.05) increased with the number of frying cycles. However, when the oil is heated with food, PV of samples S1, S3 and S5 had significantly (p<0.05) increased only up to the sixth frying cycle, and a significant (p<0.05) reduction was observed thereafter. Sample 5 had a significantly (p<0.05) higher peroxide value compared to all the other samples throughout

than that of oil samples heated without food until the fourth number of frying cycle. However, after the fourth cycle the mean PV of oil samples heated with food gradually became lower than that of oil heated without food. This could be due to the decomposition of peroxides into secondary oxidation products when the oil is heated with food, which can be observed in Figure 6, where the PV of three coconut oil samples increased during the first period of frying until reached the peak and then started to decrease.

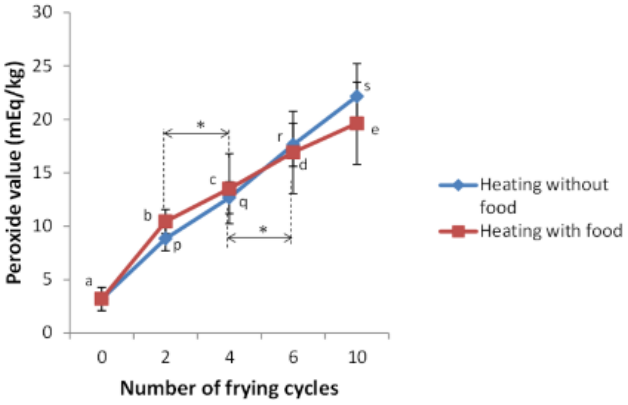


Figure 7: Comparison of mean PV in the coconut oil samples over repeated heating with and without food. The mean±SD PVs of coconut oil samples (S1, S2, S3 and S4) excluding the significantly different sample S5 were calculated at the selected number of frying cycles (0, 2, 4, 6 and 10). The mean±SD PVs had significantly (p<0.05) increased with the number of frying cycles in both samples heated with and without food (different letters within a series indicate a significant difference at p<0.05).The mean±SD PVs of coconut oil samples heated with and without food after each respective frying cycle were not significantly (p>0.05) different. However, after the fourth frying cycle, the mean peroxide value (PV) of oil samples heated in the presence of food gradually became lower than that of oil samples heated without food

Previous studies have reported that intermittent repeated heating results in greater oil deterioration than continuous heating.This increased deterioration is attributed to the formation of fresh lipid hydroperoxides during the cooling and storage intervals between heating cycles, followed by their thermal decomposition into secondary oxidation products during subsequent reheating, thereby promoting further oxidative degradation of the oil (Le Gresley et al., 2019).

Effect of frying on TBARS

The TBARS assay is one of the most widely used methods for assessing secondary lipid oxidation and has been extensively applied for more than four decades (De Leon & Borges, 2020). The assay primarily quantifies malondialdehyde (MDA) and other thiobarbituric acid-reactive carbonyl compounds formed during the decomposition of lipid hydroperoxides. Among these compounds, MDA is one of the major aldehydic products generated during secondary lipid oxidation and is widely recognized as a reliable biomarker of oxidative deterioration. The accumulation of these secondary oxidation products is associated with the development of undesirable off-flavours and rancidity in oxidized oils (Abeyrathne et al., 2021). Furthermore, the TBARS assay has been reported to provide a reliable estimate of MDA content in cooking oils, supporting its suitability for evaluating oxidative deterioration during thermal processing (Piranavatharsan et al., 2023).

The changes in the TBA value of the coconut oil samples over the repeated heating of oil with and without food are shown in the Figure 8. It was observed that values of TBA had significantly (p<0.05) increased with number of frying cycle in all coconut oil samples heated with and without food, indicating a gradual formation of secondary oxidative products in the coconut oil with time.

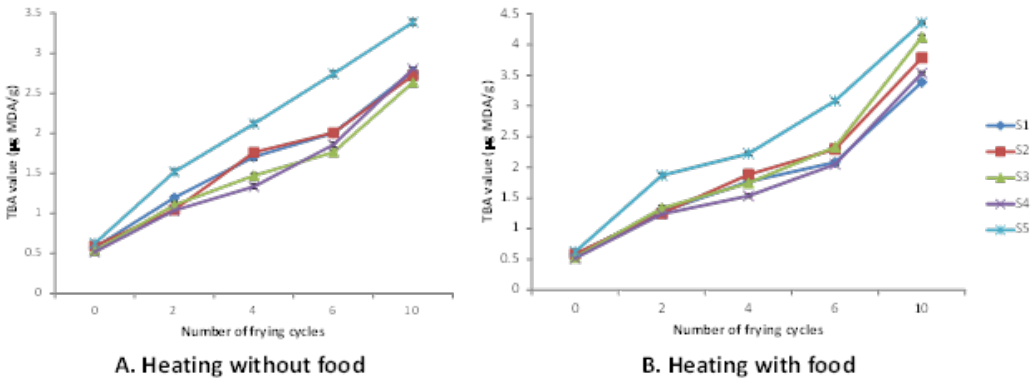


Figure 8: Changes in the TBA values of the coconut oil samples over repeated heating of oil without food (A) and with food (B). Five coconut oil samples (S1, S2, S3, S4 and S5) were repeatedly heated (at 180±5°C for 20 min) without food and with food (vadai) for ten cycles allowing cool down to room temperature after each frying. The TBA values of the oil were determined after selected frying cycles (0, 2, 4, 6 and 10), and they had significantly ($p<0.05$) increased with the number of frying cycles. Sample 5 had a significantly ($p<0.05$) higher TBA value compared to all the other samples throughout

Figure 9 shows the comparison of the mean TBA value of all coconut oil samples excluding the most deviated sample (sample 5) over repeated heating with and without food. It was observed that even though the mean TBA values of oil heated without food were lower than that of oil heated with food, there was no significant ($p>0.05$) difference between them. Thermal oxidation occurring during repeated frying, both in the presence and absence of food, leads to the formation and accumulation of secondary lipid oxidation products, as reflected by the progressive increase in TBA values with increasing frying cycles. When oil is heated with food, water in the food leaches to the oil inducing hydrolysis of glycerides and releasing free fatty acids. These free fatty acids easily undergo thermal oxidation and produce more primary and secondary oxidation products which leads to the high TBA values observed in the oil. However, there was no significant ($p>0.05$) difference between oil heated with and without food because the formed secondary products could be absorbed by the food and the amount of TBARS formed through the oxidation of free fatty acids might be comparatively low.

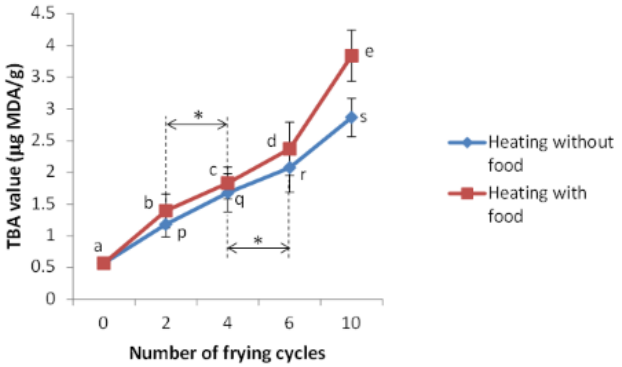


Figure 9: Comparison of mean TBA value in the coconut oil samples over repeated heating with and without food. The mean±SD TBA values of coconut oil samples (S1, S2, S3 and S4) excluding the significantly different sample S5 were calculated at the selected number of frying cycles (0, 2, 4, 6 and 10). The mean±SD TBA values in oils significantly ($p<0.05$) increased with the number of frying cycles in both samples heated with and without food (different letters within a series indicate significant difference at ($p<0.05$). The mean±SD TBA values of coconut oil samples heated with and without food after each frying cycle tested were not significantly ($p>0.05$) different

Structural alteration of fatty acids

Repeated frying accelerates the thermo-oxidative degradation of fatty acids, leading to the generation of primary and secondary oxidation products as well as polymerization compounds. These chemical changes not only impair the sensory and nutritional quality of fried foods but also raise food safety concerns. Consumption of thermally abused oils has been reported to reduce fat digestibility and may induce gastrointestinal disorders, such as diarrhoea (Chen et al., 2021). Thermal oxidation results in structural modifications of fatty acids, mainly through *cis–trans* isomerization and positional shifts of double bonds. As these changes may have implications for food safety, the *trans*-fatty acid content was determined in the present study as a marker of thermally induced structural alteration in repeatedly heated coconut oil.

Trans-fatty acids are referred to as unsaturated fatty acids with at least one or more double bonds in the *trans* position (Guo et al., 2023). During deep fat frying, the *cis* configuration of fatty acids can be changes into *trans* configuration due to the breakage, shift and formation of C-C bonds (Zhang et al., 2015). There are safety issues for human health contributed by high contents of *trans*-fats in fried foods since they promote coronary heart diseases, increase the LDL cholesterol and triacylglycerol levels and decrease the HDL cholesterol in the blood. There is some evidence that *trans*-fats can cause systemic inflammation and sudden cardiac death (Pipoyan et al., 2021). High temperature (above 190°C) can increase the amount of *trans*-isomers during deep-frying (Valle et al., 2024).

The results showed that *trans*-fat content in coconut oil was slightly changed with the number of frying cycles when the oil is heated with food (Figure 10). However, the formation of *trans*-fats was relatively low since coconut oil has about 92% of saturated fatty acids. Thus, there was a less chance for the shifting of double bond to *trans*-configuration and formation of *trans*-fats.

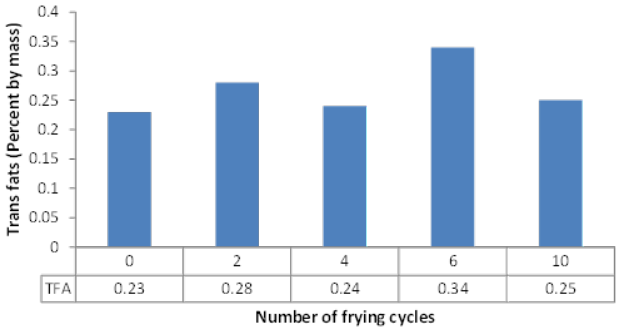


Figure 10: Changes in the *trans*-fat content of the coconut oil samples over repeated heating with food. Five coconut oil samples (S1, S2, S3, S4 and S5) were repeatedly heated (at 180±5°C for 20 min) with food (*vadai*) for ten cycles allowing cool down to room temperature after each frying. *Trans*-fat content in the oil was determined after selected frying cycles (0, 2, 4, 6 and 10), and it showed a fluctuating change with the number of frying cycles. TFA denotes *trans*-fats

Effect of frying on oil color

Colour is an important quality attribute of edible oils, influencing both the refining process and consumer acceptance. Changes in oil colour during frying reflect physicochemical alterations associated with thermal degradation and, therefore, can serve as a useful indicator of oil deterioration (Yang et al., 2022). The colour of edible oils is primarily determined by naturally occurring pigments, including carotenoids and polyphenolic compounds. Each of these pigments exhibits characteristic light absorption at specific wavelengths, collectively contributing to the overall colour of the oil.

As shown in Figure 11, the colour intensity of coconut oil sample S4 increased up to the second frying cycle during repeated heating in the absence of food, after which it gradually decreased until the tenth cycle. In contrast, when the oil was heated in the presence of food, the colour intensity increased progressively up to the fourth frying cycle before gradually declining during the subsequent frying cycles. Overall, oil heated in the presence of food exhibited greater colour darkening than oil heated without food, suggesting that food components enhanced colour formation during repeated frying.

Table 1 shows the photometric color index (PCI) of coconut oil samples over repeated heating of oil without food. It was revealed that there were no significant (p>0.05) differences in PCI between the coconut oil samples when heated without food, however sample 1 (S1) had a slightly lower mean±SD value of PCI than the other four samples. In each sample, the PCI values showed a significant (p<0.05) difference over the number of heating cycles. Most of the samples showed maximum PCI at the second cycle of repeated heating and the PCI had increased significantly until the second cycle of frying however after that, the PCI had significantly reduced until the tenth cycle in all samples.

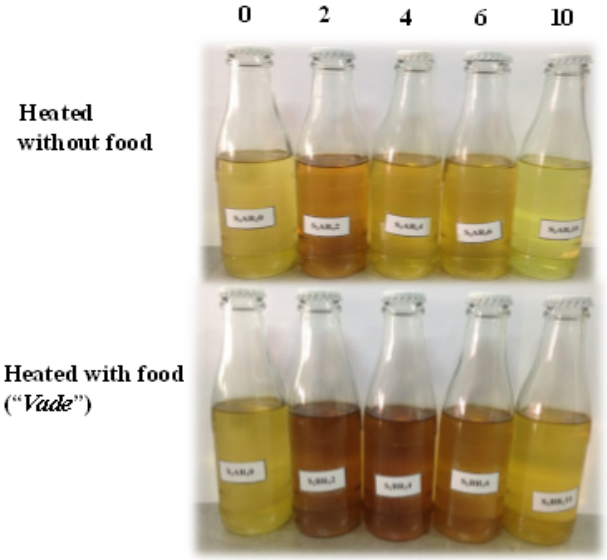


Figure 11: The changes in the visual color of the coconut oil sample (S2) over repeated heating of oil with and without food. Five coconut oil samples (S1, S2, S3, S4 and S5) were repeatedly heated (at 180±5°C for 20 min) with food (*vadai*) and without food for ten cycles allowing cooling down to room temperature after each frying. The color of the samples was visually inspected after selected frying cycles (0, 2, 4, 6 and 10). The color had changed with the number of frying cycles in oil heated with and without food. The oil heated with food showed a higher color development than that of oil heated without food

As shown in Table 2, PCI of the coconut oil samples changed significantly (p<0.05) with increasing frying cycles during repeated heating in the presence of food. No significant differences (p>0.05) in PCI were observed among the five coconut oil samples; however, samples S2 and S4 exhibited slightly higher mean ± SD PCI values than the remaining samples. In most samples, PCI increased significantly (p<0.05) up to the fourth frying cycle, reaching its maximum value at this stage. Thereafter, PCI decreased significantly (p<0.05) in all samples during the subsequent frying cycles.

The PCI was used as an indicator of the extent of oil degradation during repeated frying. The increase in PCI observed during repeated heating is likely attributable to the combined effects of lipid oxidation and polymerization of unsaturated fatty acids in the frying medium. Compared with many other edible oils, coconut oil exhibited relatively low colour development, which may be associated with its

low content of unsaturated fatty acids and, consequently, its greater oxidative stability. Under both frying conditions (with and without food), the highest PCI values were observed at the second and fourth frying cycles, respectively, suggesting the greatest extent of oxidative deterioration at these stages. The increase in PCI is likely associated with the accumulation of non-volatile degradation products, including oxidized triacylglycerols, free fatty acids, and other high-molecular-weight oxidation products formed during frying (Chen et al., 2022).

By comparing the heating of oil with food and without food, it was observed that samples S2, S3, S4 and S5 had a significantly (p<0.05) high mean value of PCI when heated with food that that of oil heated without food, while the sample S1 did not show any significant (p>0.05) difference between the oil heated with and without food. Solubilization of unsaturated carbonyl compounds and non-polar compounds from foodstuff into the oil or Maillard browning reaction and caramelization at the high frying temperature as food containing sugar could be the reason for more darkening of oil when heated with food than that of oil heated without food (Huang et al., 2024). The decrease in the color after a certain cycle of repeated frying could be due to the accumulation and deposition of non-volatile compounds at the bottom of the frying pan which contributed to the color development in the oil during frying.

CONCLUSION

In conclusion, repeated deep frying of food in the tested unrefined copra coconut oils significantly (p<0.05) accelerated physicochemical deterioration and oxidative degradation compared with repeated heating in the absence of food. The presence of food promoted hydrolytic and oxidative reactions, resulting in significantly (p<0.05) higher FFA contents and progressive deterioration of oil quality with increasing frying cycles. This deterioration was evidenced by significant increases in FFA content, PV, TBARS, and PCI, together with significant reductions in SV and IV. Collectively, these findings demonstrate that repeated frying progressively compromises the physicochemical quality and oxidative stability of unrefined copra coconut oil, with the extent of deterioration increasing as the number of frying cycles increases.

From a food quality and safety perspective, the deterioration associated with repeated frying should be considered when coconut oil is used by street vendors and households for deep frying. Based on the physicochemical quality criteria specified by the Codex Alimentarius and Sri Lanka Standards, including FFA content, PV, PV, and IV, the tested coconut oils remained within the acceptable limits for up to two to four frying cycles, depending on the quality parameter evaluated. Therefore, under the experimental conditions employed in this study, repeated use of unrefined

Table 1: Changes in the PCI of the coconut oil samples over repeated heating without food

Number of frying cycles	Coconut oil Sample Number				
	S1	S2	S3	S4	S5
0	2.90 ± 0.45^{a}	2.93 ± 0.08^{c}	3.22 ± 0.19^{b}	3.51 ± 0.02^{b}	2.65 ± 0.03^{c}
2	2.63 ± 0.05^{a}	3.74 ± 0.16^{a}	3.98 ± 0.27^{a}	3.90 ± 0.24^{a}	3.54 ± 0.09^{a}
4	2.79 ± 0.38^{a}	3.26 ± 0.06^{b}	3.38 ± 0.04^{b}	2.99 ± 0.16^{c}	3.35 ± 0.19^{a}
6	2.46 ± 0.09^{a}	2.98 ± 0.01^{c}	$2.98 \pm 0.06^{b,c}$	2.86 ± 0.02^{c}	3.32 ± 0.06^{a}
10	2.47 ± 0.19^{a}	2.87 ± 0.17^{c}	2.81 ± 0.05^{c}	2.94 ± 0.04^{c}	3.04 ± 0.04^{b}

Mean± S.D value with the different letters within same column are significantly different at p<0.05

Table 2: Changes in PCI of the coconut oil samples over repeated heating with food

Number of frying cycles	Coconut oil Sample Number				
	S1	S2	S3	S4	S5
0	2.90 ± 0.45^{a}	2.93 ± 0.08^{d}	2.22 ± 0.19^{a}	2.51 ± 0.02^{b}	2.65 ± 0.03^{b}
2	3.11 ± 0.10^{a}	4.54 ± 0.15^{a}	2.84 ± 0.28^{a}	3.12 ± 0.14^{a}	3.24 ± 0.43^{a}
4	2.98 ± 0.08^{a}	4.67 ± 0.05^{a}	2.89 ± 0.20^{a}	4.57 ± 0.15^{b}	3.46 ± 0.17^{b}
6	2.87 ± 0.11^{a}	3.83 ± 0.01^{b}	2.81 ± 0.09^{a}	$3.41 \pm 0.39^{b,c}$	$2.90 \pm 0.12^{a,b}$
10	2.61 ± 0.17^{a}	3.35 ± 0.17^{c}	2.77 ± 0.09^{a}	2.87 ± 0.15^{c}	$2.93 \pm 0.09^{a,b}$

Mean± S.D value with the different letters within same column are significantly different at p<0.05

copra coconut oil beyond two to four frying cycles may adversely affect its quality and oxidative stability.

Nevertheless, the applicability of these findings to commercial frying practices may be limited, as street vendors commonly replenish used frying oil with fresh oil during successive frying operations, thereby altering the rate and extent of oil deterioration. Future studies should investigate the physicochemical and oxidative changes in other commercially available coconut oils, including virgin coconut oil, white coconut oil produced from desiccated coconut, and refined coconut oil produced from copra. In addition, the influence of foods with different compositional characteristics on the deterioration of frying oil warrants further investigation to provide a more comprehensive assessment of coconut oil stability under practical frying conditions.

DECLARATION OF CONFLICT OF INTEREST

The authors declare no conflicts of interest.

FUNDING INFORMATION

No funding was received.

AUTHOR CONTRIBUTION

Conceptualization: **R.P.N.P.R.;** Data collection and statistical analysis: **S.S.;** data interpretation: **S.S., R.P.N.P.R., B.E.P.M.;** Writing the first draft: **S.S.**; Manuscript editing and reviewing: **R.P.N.P.R., B.E.P.M.**

DATA AVAILABILITY STATEMENT

All data generated or analyzed during this study are included in this published article**.**

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