Pak. J. Agri. Sci., Vol. 58(3), 977-984;2021
ISSN (Print) 0552-9034, ISSN (Online) 2076-0906
DOI: 10.21162/PAKJAS/21.529
http://www.pakjas.com.pk

EFFECT OF ADDING POLYPHENOLIC FRACTIONS ON THE ACROLEIN AND *TRANS* FATTY ACID CONTENTS DURING DEEP FRYING AND HEATING OF CORN OIL

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Easily accessible, natural polyphenols were used to stabilize the corn oil degradation during repeated use of deep frying and heating. Degradation of edible oil quality is directly related to loss of nutritional value, taste and flavor of products. Effect of crude extract (CE), neutral fraction (NF) and acidic fraction (AF) of *Camellia sinensis* (green tea), gallic acid (GA), catechin (Ctch.) and quercetin (Qt.) were analyzed during repeated frying and heating of corn oil at 180˚C up to 24 hours (144 batches) on oil stability index (OSI), acrolein (ACR) and *trans* fatty acids (TFA). All polyphenols reduced the formation of ACR and TFA in comparison with the control sample measured at 4 hours interval. The oil stability index (OSI) was also elevated by all polyphenols. The order of the highest activity as an antioxidant in OSI and ACR was CE > GA > Ctch.> *NF* > Qt.>AF. The sequence of activity as anti-isomer (reduce *trans* isomerization) was NF>Qt.>GA>CE>AF. Potatoes accelerated the formation of ACR and TFA during deep frying of corn oil when compared with oven heated corn oil.
Keywords: Acrolein, *trans* fatty acid, oil stability index, *Camelia sinensis*, repeated frying.

INTRODUCTION

The chemical reactions followed by loss in nutritional quality and flavor during the frying of oils were studied extensively (Choe and Min, 2006). It was reported that the rate and extent of hydrolysis, isomerization (Gercar and Smidovnik, 2002), oxidation and polymerization reactions during frying are dependent upon the frying time, temperature, mode of frying (continuous or intermittent), the composition of the frying oil and fried stuff, chemical structure and concentration of antioxidants (Totani *et al.*, 2012; Koh and Surh, 2015; Lolos *et al.*, 1999; Ponginebbi *et al*., 2000; Milic *et al*., 1998; Malheiro *et al*., 2009; Houhoula *et al*., 2003). Numerous degradation products of these reactions were identified as a consequence of heating of oil which includes lipid hydroperoxides, free fatty acids, polymerized glycerides and products from further degradation of hydroperoxides for example aldehydes, ketones, acids, alcohols, esters and short-chain hydrocarbons (Esterbauer *et al*., 1991; Miyata *et al.*, 2000; Choe and Min, 2006). These products were caused by quality loss and loss of nutritional value (Zbikowska, 2010, Aladedunye and Przybylski, 2008). Among all the degradation products, formation and monitoring of acrolein (ACR) (an unsaturated aldehyde) and *trans* fatty acids (TFA) were taken seriously as both were reported as health hazardous (Ascherio and Willet, 1997).

Acrolein was reported as a potent carcinogen. Acrolein displayed high toxicity because of its passive diffusion in the cell membrane (Burcham *et al*., 2008) and covalent replacement of proteins, DNA and phospholipids (biomolecules) (Ellis *et al*., 2007; Zarkovic, 2003; Uchida, 2000). ACR caused oxidative stress leading to Alzheimer's disease (Williams *et al*., 2006; Uchida *et al*., 1998; Poli, 2008) and disturbed redox potentials in the body (Uchida and Stadtman, 1992; Falletti *et al*., 2007; Petersen and Doorn, 2004) and even caused cancer, atherosclerosis, traumatic spinal cord injury and diabetes (Calingasan *et al*., 1999; LoPachin *et al*., 2008; Yong *et al*., 2010; Hamann and Shi, 2009; Lovell *et al*., 2001 and Ellis, 2007). Glycerol and fatty acids were formed when the oil heated beyond the smoke point due to hydrolysis of oil and then free glycerol oxidized to produce acrolein and water. Hirayama *et al.* (1989) examined methyl linoleate for acrolein production through autoxidation and the result found positive with the quantification of acrolein (2258 µg/g). It was observed that deep-frying of chips in oil having a high quantity of linolenic acid leads to high acrolein formation (Ewert *et al*., 2012).

It was reported that a high concentration of TFA promoted coronary heart diseases (Martin *et al*., 2007; Willet, 2006; Stender *et al*., 2006; Oomen *et al*., 2001). TFA not only increased the low-density lipoprotein cholesterol in the body but also decreased the HDL cholesterol (Mensink and Katan, 1990, Zock and Katan, 1997). Diabetes and insulin resistance were reported due to TFA consumption (Mozaffarian, 2006; Riserus, 2006 and Bendsen *et al*., 2011). During heating at a temperature above 180˚C or above smoke point, the *cis* double bond in fatty acids shifted to adjacent bonds, forming trans *fatty* acids (Yang *et al*., 2012, Przybylski and Aladeunye, 2012).

Several studies based on development, mechanism, monitoring and possible reduction of *trans* fatty acids (Hou *et al.,* 2011) and acrolein in frying oil were designed (Beaucham *et al*., 1985; Zhu *et al*., 2011; Zhu *et al*., 2009; Steven and Maier, 2008). Gamel *et al*. (1999) found that TFA contents were increased with frying time while Tsuzuki *et al*. (2010) found that TFA formation was related to the increase in temperature. Weber *et al*. (2008) detected TFA when silver catfish was fried in hydrogenated vegetable oil. Simple heating generated less *trans*-fat in canola oil as compared to the oil used for frying. C18:2t formation was observed higher in corn oil when heated at 180˚C for 2 and 4 hours in comparison to canola oil, rice bran oil, safflower oil and sesame oil (Tsuzuki *et al.,* 2010). C18:2t was found higher in oil extracted from French fries, fried in margarine at temperature 180˚C up to 31 minutes in comparison to mixed oil and sunflower oil (Yildirim *et al*., 2015). TFA formation was observed in corn oil during heating at above 180˚C and increased with heating time and temperature both (Yang *et al*., 2012). Heating and frying of corn oil at 170˚C had no effects on TFA formation even fish fillet did not possess any significant amount of TFA during frying (Yang *et al*., 2014). TFA formation was 6.17 times higher in pressed soya bean oil when French fries were fried at 180 to 185˚C in comparison to first and third grades solvent extracted soya bean oils (Hou *et al*., 2011).

Many of the researchers highlighted the role of natural antioxidants in the control of these deleterious compounds. In Spain, phytosterols, alpha-tocopherol and beta carotene were used in croissants and muffins to control the TFA in products. The results were found satisfactory and recommended for other bakery items (Quilez *et al*., 2006). Lutein (0.1g/kg) reduced the TFA up to 1.43% while rosemary extract (Rosmarinus officinalis L.) (1g/kg) reduced the TFA contents to 1.55% (Filip *et al*., 2011). Methanolic phenolic extract of dry rosemary alone and in the combination of BHA were examined for TFA formation during frying in olive and sunflower oil. A decreased in TFA contents was observed (Gamel *et al*., 1999)

Number of synthetic and natural antioxidants was used as trapping agents for ACR. The high radical scavenging activity and antioxidation capacity were shown by the quercetin rich flavonoid extract from *Sophora jabonica* flower buds during the chicken (as real food system) cooking using lard and sunflower (Mihalova and Schalow, 2013).

The catechin flavonols in leaves of tea (green and black) were identified as a major scavenger of ACR on account of 77-82% and 47-58% anti-oxygen activity respectively (Gardner *et al*., 1998). Gramza *et al*., (2006) found higher total polyphenols in green tea than black tea, depending upon the extent of extractability that was related to the nature of the solvent, type of the leaves and method of extraction.

This study is based on the extraction of polyphenolic fractions from *Camellia sinensis* (green) and determination of their effect on the oil stability index, acrolein and *trans* fatty acid contents of frying and heating corn oil along with some pure polyphenols.

MATERIALS AND METHODS

Refined corn oil, green tea, potatoes and chicken were bought from the local market. Sigma Aldrich's analytical grade gallic acid, quercetin, catechin and The BIO-RAD's polypropylene columns and OASIS (Waters) Lichroprep RP 18 column were used for experiments. Analytical grade chemicals and solvents were purchased from MERCK. Perkin Elmer UV-Visible Spectrophotometer, ANNEX deep fryer was used during the study. The samples were stored at 4˚C until further used.

***Extraction, fractionation and quantification of Polyphenol in Green tea*:**

The polyphenols extracted from the green leaves of *Camellia sinensis* were separated into acidic and neutral polyphenol and total phenols were then determined into these fractions according to the procedure described by Sheikh *et al*., (2016).

***Sample Preparation*:** One thousand milligrams of each of the three extracts (crude extract of green tea (CE), acidic fraction (AF) and neutral fraction (NF) in triplicate) were added to 10 L of corn oil in nine separate containers. In nine other separate containers, 100 mg of each of the three antioxidants, catechin (Ctch), gallic acid (GA), quercetin (Qt) in triplicate were added to 10 L of corn oil. Three 10 L corn oil containers without any antioxidants were used as blank, so there were 21 samples altogether. The samples were stored in dark and air-tight bottles. A volumetric flask was used for measuring oil and containers were airtight and kept in a cool and dark place till further use.

***Effect of adding polyphenols on Oil Stability Index*:** The samples prepared above were analyzed for oil stability index according to the method of (AOCS, 1996. Official methods Cd 12b-92).

Effect of adding polyphenols on acrolein and TFA in frying corn oil

Deep frying: Peeled potatoes were cut into 10 x10 x 90 mm cubical bars and fried at 180˚C in 2.5 L of 21 samples of oil individually in a deep fryer. The batch size was kept fifty grams and frying time was ten minutes per batch. At the end of frying of the first batch the second batch of 50 g peeled potatoes were fried in the same oil for 10 minutes and in this way, 144 batches were fried at the rate of 8 hours per day for 24 hours in total. Frying oil samples were drawn from the fryer after every 4 hours so there were six test samples for each of the 21 samples prepared above and 126 samples overall for TFA and acrolein analysis. The oil samples were filtered after frying and cooling and stored in the dark at 4˚C until further analysis.

***Estimation of Acrolein in deep-fried oil*:** The method of Cohen and Altschuller, 1961 was adopted for Acrolein

estimation. Ethyl alcohol (10 ml) was added to 10 ml of the oil sample taken in a test tube. Then 4-hexylresorcinol solution (0.5 ml) containing 1 ml of the $HgCl_2$ solution and 3.5 ml trichloroacetic acid solution were poured into the same tube. Similarly, a blank was prepared (without oil sample) in a separate test tube. Test tubes were heated at 60˚C, cooled for 15 to 20 minutes and the absorbance was recorded by using a spectrophotometer at 605 nm.

Estimation of TFA in deep-fried oil*: *Preparation of methyl ester

Methyl esters were prepared from 250 mg of oil sample using the method of AOAC 969.33 (1997) and analyzed by gas chromatography after dilution to 10%.

***Estimation of trans fatty acids*:** Estimation of *trans* fatty acid (TFA) was carried out on Schimatzo, 2010 by AOAC 994.15 (1995) using standards of methyl esters for comparison.

Effect of adding polyphenols on Acrolein and TFA in oven heated corn oil

***Oven Heating*:** One hundred milliliter of each of the 21 samples and control were poured in 250 ml beakers separately. The samples were heated in an oven (Binder B34, 7200 Tuttlingen, Germany) at 180˚C for 24 hours (8 hours per day). Ten-milliliter oil was drawn from each of the samples in the oven after every 4 hours so there were 126 samples collected and stored at 4˚C till further analysis of TFA and acrolein.

***Estimation of Acrolein and TFA in oven heated oil*:** Estimation of acrolein and TFA in oven heated oil samples were carried out as described in procedure 2.2.4.2 and 2.2.4.3.

***Statistical Studies*:** All analyses were performed in triplicate and the results were analyzed by two-way ANOVA (SPSS version 17.0 Inc, Chicago, USA) and reported as mean of triplicate + standard deviation. Significance was measured at $p<0.05$

RESULTS AND DISCUSSION

***Total phenols*:** Total phenols in CE, AF and NF ($n=3$, $p<0.05$) were found to be 14500±330 mg GAE/100g, 1538±49.0 mg GAE/100g and 11800±158.0 mg GAE/100g respectively (Fig. 1). These results are concomitant to the results reported by Aman *et al*., (2013).

***Oil Stability Index (OSI)*:** The oil stability index is the accelerated procedure for determining oil oxidation stability (Coppin and Pike, 2001). There was a significant increase ($p<0.05$) of OSI in corn oil with extract and pure antioxidants compared to reference corn oil (Fig. 2). The order of OSI was found to be CE > GA > Ctch.> *NF* > Qt.>AF.

Among the crude extract and its fractions, the crude extract showed the highest OSI owing to the highest concentration and variety of polyphenols. Apak *et al*. (2006) demonstrated the antioxidant capacity of polyphenols in *C. Sinensis*. Gardner (1998) showed the effectivity of catechin flavanols in control of lipid oxidation. Caldwell (2001) reported the antioxidant potential of green tea due to epigallocatechin-3-gallate (EGCG) and epicatechin gallate (ECG).

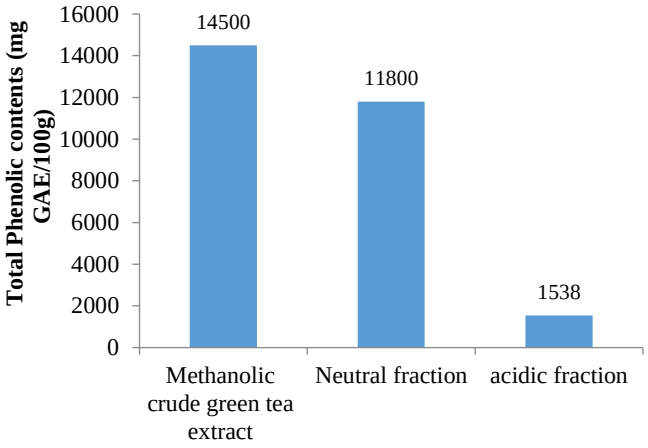

Figure 1. Phenol contents of methanolic crude extract of *Camilla Sinensis* leaves (green tea) and its fractions.

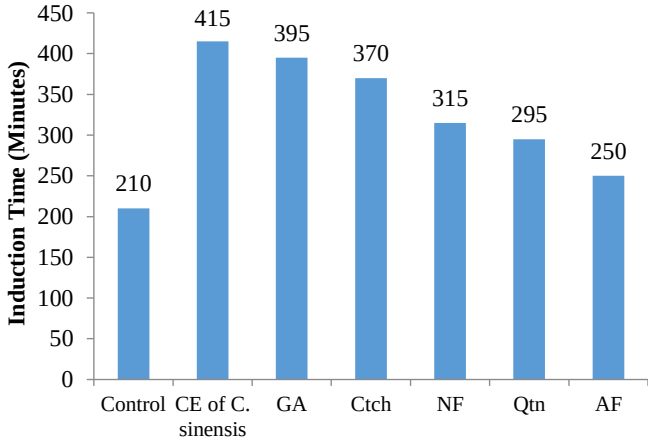

Figure 2. Oil stability index of corn oil at 120˚C in the presence of Crude extract of *Camellia sinensis*, acidic fraction and neutral fractions, gallic acid, catechin and quercetin

Among pure polyphenols, GA showed a higher oil stability index as it contains three adjacent OH groups which enable it to inhibit oil oxidation more strongly than other molecules. Phenolic antioxidant (R-OH) reduced lipid oxidation by scavenging alkoxyl radical depending upon the amount and location of –OH group (Millic *et al*., 1998, Sroka and Cisowski, 2003). The target points of phenolic acids are reactive oxygen species (ROS) including $O_2^{\bullet-}$, $^{\bullet}OH$, $NO^{\bullet}$, $RO^{\bullet}$, $ROO^{\bullet}$) while flavonoids are known for their metal chelating activity (Potapovich and Kostyuk, 2003)

$$LO^{\bullet} + R\text{-}OH \rightarrow LOH + RO^{\bullet}$$

Alkoxyl radical — *Hydrogen gain*

Phenolic antioxidant *Hydrogen donation*

Catechin showed higher antioxidant ability than quercetin due to the presence of *ortho* dihydroxy benzene (catechol) group at the ring-B of catechin and *meta* dihydroxy benzene (resorcinol) group at the ring-A of catechin. It was reported earlier that –OH group having ortho position on a ring- B and meta position on the ring-A of flavonoids possess higher antiradical (oxygen anion radical) activity (Potapovich and Kostyuk, 2003). The reduction potential of the polyphenols is lesser than the reduction potential of concerning free radicals that may cause the transfer of hydrogen to free radicals. The reduction potential of gallic acid (-1.04 V at pH 7.53) (Abbasi *et al.*, 2011) is lesser than catechin (0.11 V at pH 8) (Janeiro & Brett, 2004) and catechin reduction potential is still less as compared to quercetin (0.33 V at pH 7) (Jovanovic *et al.*, 1996).

***Measurement of acrolein*:** Acrolein was analyzed in 126 samples of corn oil (collected during frying as described in 2.2.4). Oil during deep-frying at 180˚C was estimated for acrolein and changes are illustrated in Table 1. The control sample exhibited the highest ACRs during the entire period of the experiment. Adding *CE* to the corn oil, prevented the development of acrolein through lipid hydrolysis progressions. Similar to the OSI results, the effects of natural and synthetic antioxidants on delaying the formation of ACR were observed through the frying process and the order of inhibitory effects of natural and synthetic antioxidants was similar to OSI i.e CE > GA > Ctch.> *NF* > Qt.>AF. The reason for the relatively high antioxidant potential of gallic acid has already been explained in section 3.2.

All polyphenols crude extract (CE), neutral fraction (NF) and acidic fraction (AF) of *Camellia sinensis* (green tea), gallic acid (GA), catechin (Ctch.) and quercetin (Qt.) reduced ACR in comparison with control sample but with time ACR formation was increased. An increase in ACR could be due to the degradation of antioxidants as they are converted into volatiles at high temperature and activity was decreased with heating time. Cheng *et al.* (2014) stated the decomposition of five phenols (catechol, protocatechualdehyde, salvianic acid A, protocatechuic acid and ferulaic acid) at high temperature and heating during long hours. Salvianic A, protocatechuic and ferulaic acids were highly decomposed with increasing temperature. The stability of acids was decreased with a prolonged time of heating.

Changes in acrolein of corn oil samples during oven heating at 180˚C is illustrated in Table 2. The same pattern of antioxidant activity was observed in the oven heated oil without food as in deep-frying oil i.e., CE > GA > Ctch.> *NF* > Qt.>AF. The lower ACR values were may be due to the absence of food moisture (same air exposure), which initiated a series of interrelated reactions. Triacylglycerol was hydrolyzed into di or monoacylglycerol in the presence of moisture and resulted in the formation of free fatty acid and

Table 1. Effect of adding different polyphenols / extract as antioxidants on the acrolein formation in corn oil, deep fried at 180˚C for 24 hours.

Samples	Acrolein mg/L (deep fried)					
	4 hours	8 hours	12 hours	16 hours	20 hours	24 hours
Oil without antioxidant	37.0±2.0	38.5±1.4	39.0±1.0	40.0±1.8	40.0±2.0	42.0±1.5
Crude extract	18.0±2.0	19.0±1.0	19.5±1.0	20.0±0.8	20.5±1.0	21.5±1.5
Gallic acid	21.6±1.7	22.0±1.0	22.0±1.2	22.5±1.0	22.5±1.1	23.0±1.6
Catechin	24.0±1.3	24.5±1.8	25.0±0.9	25.9±1.2	26.6±1.9	27.4±2.0
Neutral fraction	26.5±1.0	27.0±2.0	29.5±0.5	31.8±1.0	33.8±1.5	35.9±1.6
Quercetin	29.5±1.0	30.0±2.0	32.0±0.2	33.9±1.0	35.4±1.2	37.0±0.8
Acidic	33.0±1.0	33.5±05	36.0±0.5	38.2±1.0	39.7±1.8	41.4±2.0

Note: Estimates are mean of triplicate ± SD. Numbers in the similar column were significantly dissimilar at p<0.05.

Table 2. Effect of adding different polyphenols / extract on acrolein formation in corn oil heated in an oven at 180˚C for 24 hours.

Samples	Acrolein mg/L (heated in the oven)					
	4 hours	8 hours	12 hours	16 hours	20 hours	24 hours
Oil without antioxidant	24.0±2.0	25.5±1.4	27.0±1.0	28.5±2.0	30.0±2.0	31.0±1.5
Crude extract	6.0±1.2	7.5±1.0	9.0±1.0	10.5±1.0	11.0±2.0	12.0±1.0
Gallic acid	8.7±0.6	9.5±1.0	11.0±1.0	12.8±1.0	13.6±1.7	14.5±1.0
Catechin	10.7±0.6	11.7±1.0	13.6±1.0	15.8±1.8	16.5±2.0	18.2±0.7
Neutral fraction	13.0±0.7	14.5±1.0	16.6±1.0	18.5±1.2	20.0±2.0	21.8±0.5
Quercetin	15.0±1.1	16.5±0.0	19.2±0.6	21.5±1.4	23.0±2.0	25.0±0.5
Acidic	17.8±0.5	20.5±1.0	23.6±1.0	25.5±1.0	27.0±1.2	28.6±0.5

Note: Estimates are mean of triplicate ± SD. Numbers in the similar column were significantly dissimilar at p<0.05.

glycerol. Free glycerol then oxidizes to produce acrolein and water (Hirayama *et al*., 1989). Houhoula *et al.* (2003) examined cottonseed oil at 185˚C and reported that oxidized triglycerides, diglycerides, monoglycerides and free fatty acids contents are lower in heating than frying with sliced potatoes. So, hydrolysis and oxidation are limited in heated oil than frying oil.

***Measurement of TFA*:** The TFA value is an indicator of the isomerization of fatty acid. The outcomes of the TFA analysis of the corn oil samples in deep frying at 180˚C are mentioned in Table 3. An increasing trend of TFA was observed in the control sample of corn oil. CE, GA, *NF*, Qt. and AF reduced the development of *trans* fatty acid when all oil samples were analyzed during frying. The sequence of higher antioxidant activity is NF > camellia sinensis CE >AF while Qtn. > Ctch. (Fig. 1 and 2). This indicates the good capacity of NF to inhibit the isomerization process. Isomerization also occurs when a double bond is broken down by metals. Cheng *et al.* (2018) described the mechanism of *trans* oleic acid formation which includes breakage of π bond of C=C and C-H bond. The activation energy required for C-H bond was 362.62KJ/mol which can be obtained by light, metal ions or enzymes. Flavonoids are good metal chelators than phenolic acid. Khokhar & Apenten (2003) reported the higher iron-binding capacity of the compounds containing catechol group (flavonoids) than galloyol groups (tannic acid and gallic acid). Neutral fraction also contained higher flavonoids (catechin and derivatives of catechin) than an acidic fraction (gallic acid). That's why NF showed higher anti-isomerization activity. Yilmaz and Toledo in 2004 reported that quercetin possesses five –OH groups and the number of protons for antioxidation donation is 4.0 N while gallic acid possesses four –OH groups and the number of protons for antioxidation donation is 3.2 N.

All polyphenols reduced the TFA formation during heating of corn oil in a similar order as during deep frying but TFA was increased with the increase of time of heating and frying (discussed in section 3.3).

Changes in TFA values of corn oil during oven heating as illustrated in Table 4. Yang *et al.* reported in 2012 that heating of corn oil without food at 180˚C above for two hours heating gave rise the formation of TFA. TFA formed was estimated lesser in oven heating of corn oil without food than that of deep-fried oil at 180˚C for up to 24 hours. It is clearly indicated that potato is an accelerating factor for TFA formation. On contrary, Yang *et al.* (2014) reported that neither heating nor frying under 170˚C induced the formation of TFAs. This discrepancy may be due to a lesser temperature than 180˚C. Song *et al.* (2015) found no difference in TFA formation due to baking and frying of corn oil. TFA formation in the presence of quercetin after 24 hours of heating at 180˚C without food was found 0.35% while this value was achieved by potato fried corn oil in the presence of quercetin within 12 hours. Similarly, TFA in heated corn oil in the presence of neutral fraction was found to be 0.33% after 24 hours at 180˚C while this value was achieved by deep-fried corn oil before 20 hours. The lower values of TFA in heated oil were associated with the absence of any food material which could

Table 3. Effect of adding different polyphenols / extract as antioxidants on the TFA formation in corn oil, deep fried at 180˚C for 24 hours.

Samples	TFA (%) Deep fried					
	4 hours	8 hours	12 hours	16 hours	20 hours	24 hours
Oil without antioxidant	0.40±0.05	0.60±0.03	0.66±0.02	0.69±0.01	0.72±0.01	0.82±0.02
Crude extract	0.30±0.03	0.50±0.01	0.53±0.01	0.58±0.03	0.63±0.02	0.66±0.02
Gallic acid	0.24±0.01	0.25±0.01	0.36±0.01	0.40±0.01	0.60±0.01	0.62±0.04
Neutral fraction	0.21±0.01	0.25±0.02	0.29±0.02	0.31±0.02	0.34±0.03	0.40±0.01
Quercetin	0.22±0.03	0.23±0.01	0.34±0.01	0.40±0.01	0.56±0.01	0.58±0.01
Acidic	0.35±0.02	0.53±0.02	0.60±0.03	0.62±0.02	0.69±0.02	0.80±0.05

Note: Estimates are mean of triplicate ± SD. Numbers in the similar column were significantly dissimilar at $p<0.05$.

Table 4. Effect of adding different polyphenols / extract as antioxidants on the TFA formation in corn oil heated in an oven at 180˚C for 24 hours.

Samples	TFA (%) (heated in oven)					
	4 hours	8 hours	12 hours	16 hours	20 hours	24 hours
Oil without antioxidant	0.25±0.02	0.27±0.01	0.55±0.03	0.63±0.02	0.69±0.03	0.80±0.05
Crude extract	0.19±0.01	0.21±0.01	0.38±0.03	0.49±0.02	0.54±0.04	0.60±0.03
Neutral fraction	0.10±0.01	0.13±0.03	0.25±0.01	0.29±0.02	0.31±0.01	0.33±0.01
Acidic	0.21±0.02	0.23±0.02	0.45±0.03	0.51±0.04	0.60±0.03	0.73±0.03
Gallic acid	0.15±0.03	0.20±0.01	0.32±0.01	0.40±0.02	0.45±0.03	0.45±0.03
Quercetin	0.21±0.01	0.21±0.02	0.25±0.02	0.30±0.03	0.34±0.05	0.35±0.04

Note: Estimates are mean of triplicate ± SD of triplicate analyzes. Numbers in the similar column were significantly dissimilar at $p<0.05$.

release moisture or itself become a source of fatty acids. Tsuzuki *et al*., (2010) found smaller TFA formation during heating than frying of oil at three temperatures (160, 180 and 200˚C) in six types (cooking oil, canola, corn, rice bran, safflower and sesame).

***Conclusions*:** Oil degradation and formation of ACR and TFA in oil is more pronounced due to deep frying oil in comparison to simple heating due to the presence of potatoes (food). The addition of polyphenols extracted from green tea leaves decreased the oxidation in oven heated oil as well as deep-fried oil. Crude tea extract showed maximum antioxidant activity, while the acidic fraction showed the least activity with respect to OSI and ACR formation while as anti-isomerization Qt. was most effective. It may be concluded that in order to control or minimize the formation of ACR and TFA in deep frying oil, the use of polyphenols (Crude extract, acidic and neutral fractions of *Camellia sinensis)* from relatively cheap and easily available sources could be beneficial as well as pure compounds (catechin, quercetin and gallic acid).

***Acknowledgments*:** This research received no specific grant from any funding agency in the public, commercial, or not-for-profit sectors.

Conflict of interest: None declared.

REFERENCES

Abbasi, S., A. Bahiraei and A. Farmany. 2010. Quantification of Sub-Nanomolar Levels of Aluminum by Adsorptive Stripping Voltammetry Using Rubeanic Acid as a Selective Chelating Agent. Electroanalysis. 22:1889-1893.

Aladedunye, F. A. and R. Przybylski. 2008. Degradation and Nutritional Quality Changes of Oil during Frying. J. Am. Oil Chem. Soci. 86:149-156.

Aman, S., A. Naim, R. Siddiqi and S. Naz. 2013. Antimicrobial polyphenols from small tropical fruits, tea and spice oilseeds. Food Sci. Tech. Interl. 20:241-251.

AOAC. 1995. Official Method 994.15. Total cis - and trans-Octadecenoic isomers and general fatty acids composition in hydrogenated Vegetable oils and Animal fats. Ass. Off. Anal. Chem. 41.1.35A

AOAC. 1997. Official Method 969.33. Preparation of methyl esters. Ass. Off. Anal. Chem. 41.1.28.

AOCS. 1996. Official methods and recommended practices of the American oil chemist society. Am. Oil Chem. Soc. Champaign, III.

Apak, R., K. Güçlü, M. Özyürek, S. Esin Karademir and E. Erçağ. 2006. The cupric ion reducing antioxidant capacity and polyphenolic content of some herbal teas. Int. J. Food Sci. Nut. 57:292-304.

A. Ascherio and W. C. Willett. 1997. Health effects of *trans* fatty acids. Am. J. Clinical Nut. 66: 1006S-1010S.

Beauchamp, R. O., D. A. Andjelkovich, A. D. Kligerman, K. T. Morgan, H. d'A. Heck and V. J. Feron. 1985. A critical review of the literature on acrolein toxicity. Crit. Rev. Toxicol. 14:309-380.

Bendsen, N. T., S. B. Haugaard, T. M. Larsen, E. Chabanova, S. Stender, A. Astrup. 2011. Effect of trans-fatty acid intake on insulin sensitivity and intramuscular lipids—a randomized trial in overweight postmenopausal women. Metabolism. 60:906-913.

Burcham, P., L. Kaminskas, D. Tan, S. Pyke. 2008. Carbonyl-Scavenging Drugs & Protection Against Carbonyl Stress-Associated Cell Injury. Mini-Rev. Med. Chem. 8:319-330.

Caldwell, C. R. 2001. Oxygen Radical Absorbance Capacity of the Phenolic Compounds in Plant Extracts Fractionated by High-Performance Liquid Chromatography. Anal. Biochem. 293:232-238.

Calingasan, N. Y., K. Uchida and G. E. Gibson. 1999. Protein-Bound Acrolein. J. Neurochem. 72: 751-756.

Cheng, N., J. Zhang, J. Yin and S. Li. 2018. Computational and experimental research on mechanism of cis/trans isomerization of oleic acid. Heliyon.4:e00768.

Cheng, Y., Q. Xu, J. Liu, C. Zhao, F. Xue and Y. Zhao. 2014. Decomposition of Five Phenolic Compounds in High Temperature Water. J. Brazil. Chem. Soc. 25:2102-2107.

Choe, E. and D. B. Min. 2006. Mechanisms and Factors for Edible Oil Oxidation. Compr. Rev. Food Sci. Food Saf. 5:169-186.

Cohen, I. R. and A. P. Altschuller. 1961. The spectrophotometric method for the determination of Acrolein in combustion gases and in the atmosphere. Anal. Chem. 33:726-733.

Coppin, E. A. and O. A. Pike. 2001. Oil stability index correlated with sensory determination of oxidative stability in light-exposed soybean oil. J Amer Oil Chem. Soc.78:13-18.

Ellis, E. M. 2007. Reactive carbonyls and oxidative stress: Potential for therapeutic intervention. J. Pharm. Therap. 115:13-24.

Esterbauer, H., R. J. Schaur and H. Zollner.1991. Chemistry and biochemistry of 4-hydroxynonenal, malonaldehyde and related aldehydes. Free Radic Biol Med. 11:81-128.

Ewert, A., M. Granvogl and P. Schieberle. 2012. Comparative Studies on the Generation of Acrolein as Well as of Aroma-Active Compounds during Deep-Frying with Different Edible Vegetable Fats and Oils. ACS Symposium Series. 1098.129-136.

Falletti, O., J. Cadet, A. Favier and T. Douki. 2007. Trapping of 4-hydroxynonenal by glutathione efficiently prevents formation of DNA adducts in human cells. Free Rad. Bio. Med. 42: 1258-1269.

Filip, S., J. Hribar and R. Vidrih. 2011. Influence of natural

antioxidants on the formation of *trans*-fatty-acid isomers during heat treatment of sunflower oil. Euro. J. Lipid Sci. Tech. 113: 224-230.

Gamel, T., A. Kiritsakis and C. Petrakis. 1999. Effect of phenolic extracts on *trans* fatty acid formation during frying. Grasas y Aceites.50:421-425.

Gardner, P. T., D. B. McPhail and G. G. Duthie. 1998. Electron spin resonance spectroscopic assessment of the antioxidant potential of teas in aqueous and organic media. J. Sci. Food Agri. 76:257-262.

Gerčar, N. and A. Šmidovnik. 2002. Kinetics of geometrical isomerization of unsaturated FA in soybean oil. J. Am. Oil Chem. Soc. 79:495-500.

Gramza, A., S. Khokhar, S. Yoko, A. Gliszczynska-Swiglo, M. Hes and J. Korczak. 2006. Antioxidant activity of tea extracts in lipids and correlation with polyphenol content. Euro. J. Lipid Sci. Tech. 108:351-362.

Hamann, K. and R. Shi. 2009. Acrolein scavenging: a potential novel mechanism of attenuating oxidative stress following spinal cord injury. J. Neurochem. 111:1348-1356.

Hirayama, T., M. Yamaguchi, T. Nakata, M. Okumura, T. Yamazaki, T. Watanabe and S. Fukui. 1989. Formation of acrolein by the autoxidation of unsaturated fatty acid methyl esters. Eisei kagaku. 35:303-306.

Hou, J. C., L. Z. Jiang and C. W. Zhang. 2011. Effects of frying on the *trans*-fatty acid formation in soybean oils. European J. Lipid Sci. Tech.114:287-293.

Houhoula, D. P., V. Oreopoulou and C. Tzia. 2003. The effect of process time and temperature on the accumulation of polar compounds in cottonseed oil during deep-fat frying. J Sci Food Agric. 83:314-319.

Janeiro, P. and A. M. Oliveira Brett. 2004. Catechin electrochemical oxidation mechanisms. Analy. Chimi. Acta. 518:109-115.

Jovanovic, S. V., S. Steenken, Y. Hara and M. G. Simic. 1996. Reduction potentials of flavonoid and model phenoxyl radicals. Which ring in flavonoids is responsible for antioxidant activity? J. Chem. Soc. Perkin Transactions.2:2497.

Khokhar, S. and R. K. Owusu Apentlen. 2003. Iron binding characteristics of phenolic compounds: some tentative structure–activity relations. Food Chem.81:133-140.

Koh, E. and J. Surh. 2015. Food types and frying frequency affect the lipid oxidation of deep frying oil for the preparation of school meals in Korea. Food Chem.174:467-472.

Lolos, M., V. Oreopoulou and C. Tzia. 1999. Oxidative stability of potato chips: effect of frying oil type, temperature and antioxidants. J. Sci. Food Agri.79:1524-1528.

LoPachin, R. M., D. S. Barber and T. Gavin. 2007. Molecular Mechanisms of the Conjugated α,β-Unsaturated Carbonyl Derivatives: Relevance to Neurotoxicity and Neurodegenerative Diseases. Toxic. Sci. 104:235-249.

Lovell, M. A., C. Xie and W. R. Markesbery. 2001. Acrolein is increased in Alzheimer's disease brain and is toxic to primary hippocampal cultures. Neurobio. Aging. 22:187-194.

Malheiro, R., I. Oliveira, M. Vilas-Boas, S. Falcão, A. Bento and J. A. Pereira. 2009. Effect of microwave heating with different exposure times on physical and chemical parameters of olive oil. Food Chem. Toxic. 47:92-97.

Martin, C. A., M. C. Milinsk, J. V. Visentainer, M. Matsushita and N. E. de-Souza. 2007. *Trans* fatty acid-forming processes in foods: a review. Anais da Academia Brasileira de Ciências. 79:343-350.

Mensink, R. P. and M. B. Katan. 1990. Effect of Dietary *trans* Fatty Acids on High-Density and Low-Density Lipoprotein Cholesterol Levels in Healthy Subjects. New England J. Med. 323:439-445.

Mihaylova, D. and S. Schalow. 2013. Antioxidant and Stabilization Activity of a Quercetin-Containing Flavonoid Extract Obtained from Bulgarian *Sophora japonica* L. Brazil. Arch. Bio. Tech. 56:431-438.

Milić, B. L., S. M. Djilas and J. M. Čanadanović-Brunet. 1998. Antioxidative activity of phenolic compounds on the metal-ion breakdown of lipid peroxidation system. Food Chem. 61: 443-447.

Miyata, T., K. Kurokawa and C. Van Ypersele De Strihou. 2000. Advanced glycation and lipoxidation end products: role of reactive carbonyl compounds generated during carbohydrates and lipid metabolism, J. Am. Soc. Neph. 11:1744-1752.

Mozaffarian, D. 2006. *Trans* fatty acids – Effects on systemic inflammation and endothelial function. Atherosclerosis Suppl.7:29-32.

Oomen, C. M., M. C. Ocké, E. J. Feskens, M. A. J. Erp-Baart, F. J. Kok and D. Kromhout. 2001. Association between *trans* fatty acid intake and 10-year risk of coronary heart disease in the Zutphen Elderly Study: a prospective population-based study. The Lancet. 357:746-751.

Petersen, D. R. and J. A. Doorn. 2004. Reactions of 4-hydroxynonenal with proteins and cellular targets. Free Rad. Bio. Med.37:937-945.

Ponginebbi, L., W. W. Nawar and P. Chinachoti. 2000. Effect of relative humidity on lipid oxidation in freeze dried emulsions. Grasas y Aceites.51:348-354.

Potapovich, A. I. and V. A. Kostyuk. 2003. Comparative Study of Antioxidant Properties and Cytoprotective Activity of Flavonoids. Biochem. (Moscow).68:514-519.

Poli, G., F. Biasi and G. Leonarduzzi. 2008. 4-Hydroxynonenalprotein adducts: A reliable biomarker of lipid oxidation in liver diseases. Mol. Aspects Med. 29:67-71.

Przybylski, R. and F. A. Aladedunye. 2012. Formation of *Trans* Fats: During Food Preparation. Can. J. Diet. Pract. Res. 73: 98-101.

Quílez, J., J. A. Ruiz, G. Brufau and M. Rafecas. 2006. Bakery products enriched with phytosterols, α-tocopherol and β-carotene. Sensory evaluation and chemical comparison with market products. Food Chem.94:399-405.

Risérus, U. 2006. *Trans* fatty acids and insulin resistance. Atherosclerosis Suppl. 7:37-39.

Sheikh, H., S. A. Sayeed and S. Haider S. 2016. Reduction of oxidative stress by adding crude green tea extract and it's fraction to corn oil during oven heating and deep frying. World J. Pharm. Res. 5:214-232.

Song, J., J. Park, J. Jung, C. Lee, S. Y. Gim, H. Ka, B. Yi, M, J. Kim, C. Kim and J. Lee J. 2015. Analysis of Trans Fat in Edible Oils with Cooking Process. Toxic. Res. 31:307-312.

Sroka, Z. and W. Cisowski. 2003. Hydrogen peroxide scavenging, antioxidant and anti-radical activity of some phenolic acids. Food Chem. Toxic.41:753-758.

Stender, S., J. Dyerberg, A. Bysted, T. Leth and A. Astrup. 2006. A *trans* world journey. Atherosclerosis Suppl.7: 47-52.

Stevens, J. F. and C. S. Maier. 2008. Acrolein: Sources, metabolism and biomolecular interactions relevant to human health and disease. Mol. Nut. Food Res. 52:7-25.

Totani, N., S. Tateishi, T. Mori and E. G. Hammond. 2012. Oxidation of Frying Oils during Intermittent Usage. J. Oleo Sci. 61:601-607.

Tsuzuki, W., A. Matsuoka and K. Ushida. 2010. Formation of *trans* fatty acids in edible oils during the frying and heating process. Food Chem.123:976-982.

Uchida, K. and E. R. Stadtman. 1992. Selective cleavage of thioether linkage in proteins modified with 4-hydroxynonenal. Proceed. Nat. Acad. Sci. 89:5611-5615.

Uchida, K., M. Kanematsu, K. Sakai, T. Matsuda, N. Hattori, Y. Mizuno, D. Suzuki, T. Miyata, N. Noguchi, E. Niki and T. Osawa. 1998. Protein-bound acrolein: Potential markers for oxidative stress. *Proceed.* Nat. Acad. Sci. 95:4882-4887.

Uchida, K. 2000. Role of reactive aldehyde in cardiovascular diseases. Free Rad. Bio. Med. 28: 1685-1696.

Weber, J., V. C. Bochi, C. P. Ribeiro, M. Victório AdeM, T. Emanuelli. 2008. Effect of different cooking methods on the oxidation, proximate and fatty acid composition of silver catfish (Rhamdia quelen) fillets. Food Chem. 106:140-146.

Willett, W. C. 2006. Trans fatty acids and cardiovascular disease—epidemiological data. Atherosclerosis Suppl.7:5-8.

Williams, T. I., B. C. Lynn, W. R. Markesbery and M. A. Lovell MA. 2006. Increased levels of 4-hydroxynonenal and acrolein, neurotoxic markers of lipid peroxidation, in the brain in Mild Cognitive Impairment and early Alzheimer's disease. Neurobio. Aging. 27:1094-1099.

Yang, M., Y. Yang, S. Nie, M. Xie and F. Chen. 2012. Analysis and Formation of *trans* Fatty Acids in Corn Oil during the Heating Process. J. Am. Oil Chem. Soc. 89:859-867.

Yang, M., Y. Yang, S. Nie, M. Xie, F. Chen and P. G. Luo. 2014. Formation of *trans* fatty acids during the frying of chicken fillet in corn oil. Int. J. Food Sci. Nut. 65:306-310.

Yildirim, E., Ö. S. Toker, S. Karaman, A. Kayacier and M. Doğan. 2015. Investigation of fatty acid composition and *trans* fatty acid formation in extracted oils from French-fried potatoes and classification of samples using chemometric approaches. Turkish J. Agri. Forestry. 39:80-90.

Yilmaz, Y. and R. T. Toledo. 2004. Major Flavonoids in Grape Seeds and Skins: Antioxidant Capacity of Catechin, Epicatechin and Gallic Acid. J. Agri. Food Chem. 52:255-260.

Yong, P. H., H. Zong, R. J. Medina, G. A. Limb, K. Uchida, A. W. Stitt and T. M. Curtis. 2010. Evidence supporting a role for N-(3-formyl-3,4-dehydropiperidino) lysine accumulation in Müller glia dysfunction and death in diabetic retinopathy. Mol. Vision. 16:2524-2538.

Zarkovic, K. 2003. 4-Hydroxynonenal and neurodegenerative diseases. Mol. Aspects Med. 24: 293-303.

Żbikowska, A. 2010. Formation and properties of *trans* fatty acids - a review. Polish J. Food Nut. Sci. 60:107-114.

Zhu, Q., Z. Sun, Y. Jiang, F. Chen and M. Wang. 2011. Acrolein scavengers: Reactivity, mechanism and impact on health. Mol. Nut. Food Res. 55:1375-1390.

Zhu Q, Z. P. Zheng, K. W. Cheng, J. J. Wu, S. Zhang, Y. S. Tang, K. H. Sze, J. Chen, F. Chen and M. Wang. 2009. Natural Polyphenols as Direct Trapping Agents of Lipid Peroxidation-Derived Acrolein and 4-Hydroxy-trans-2-nonenal. Chem. Res. Tox. 22:1721-1727.

Zock, P. L. and Katan, M. B. 1997. Butter, margarine and serum lipoproteins. Atherosclerosis. 131:7-16.

[Received 07 May 2020; Accepted 04 Nov. 2020; Published (online) 25 Jun 2021]