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Effect of Ginger Extracts on Palm Olein Quality during Frying and Impact of Fried Oils on Some Biological Parameters of Albino *Wistar* Rats

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Abstract

The objective of this work was to evaluate the effect of ginger root extracts on the oxidative stability of palm olein during frying and to determine the impact of fried oils on some biochemical parameters of albino *Wistar* rats. The extracts were added to palm olein at concentrations 1000, 1400 and 1800 ppm. A sample containing 200 ppm of butylhydroxytoluene (BHT) served as positive control and another without additives was used as negative one. All oil samples were subjected to 15 frying cycles with samples collected at 0, 1, 5, 8, 10 and 15 cycles. Peroxide, anisidine and total oxidation values were performed to assess the oxidative stability of oils samples. Only samples taken at 0, 5, 10 and 15 cycles were used for the *in vivo* tests. One hundred and five rats divided into twenty-one groups including a neutral control group fed only with the staple food and twenty test groups were given the different oil samples (2 ml/100g of food) daily for thirty days. Results showed that the effectiveness of the extracts was concentration dependent, and that at 1800 ppm, they delayed the oxidation of palm olein better than BHT. It was also observed that consumption of the oils previously enriched with plant extracts resulted in an improvement in the biochemical parameters of the rats compared to those of rats fed with oils enriched with BHT and free from additives. These extracts can be used as natural source of antioxydant to stabilize palm olein.

Keywords: ginger root extract, frying, oxidative tests, biochemical parameters

1. Introduction

Frying is a culinary method used in households and restaurants in the world. Fried foods are part of the dietary habits of many populations and are quite popular. Indeed, during frying, texturing, oil impregnation, starch gelatinisation and Maillard reactions take place, offering to the consumer products with highly appreciated organoleptic characteristics (Bordin, Kunitake & Aracava, 2013). At the same time, there is oxidation, polymerisation and isomerisation reactions that cause oil alteration through the formation of radical and non-radical compounds, especially hydroperoxides, *Trans* fatty acids, polymers and aldehydes (Patsioura, Ziaiifar, Smith, Menzel & Vitrac, 2017; Perumella & Subramanyam, 2016). These alterations lead to the loss of the nutritional value and can affect the health upon ingestion through fried foods (Wu et al., 2019).

In fact, the reactive oxygen species formed in oil during frying are responsible of the destruction of organs, especially the liver, kidneys, heart and intestinal mucosa (Alaam, Yasin, Hafez & Mohammed, 2012; Boniface, Ejimofor & Ezissi, 2014). Moreover, there is a correlation between the consumption of fried oils and the

alteration of the lipid profile, which is reflected in a decrease in HDL cholesterol, followed by an increase in LDL cholesterol, total cholesterol, triglycerides and the atherogenecity index (Badr El Said, Nahed, & Reham, 2015; Hammad, Pu & Jones, 2016; Zeb & Khan, 2019), all of which lead to the occurrence of cardiovascular diseases. In the same line, Mesembe, Ibanga, & Osim, (2005), Chacko & Rajamohan, (2011) and Ani , Nna, Obi, & Udobong, (2015) showed that administration of thermooxidised vegetable oils to rats causes an alteration of their haematological parameters through leucocytosis, thrombocytopenia and anaemia.

The use of antioxidants remains an indispensable means of limiting the formation of free radicals during frying. However, the use of such compounds should take into account their nature, their safety, their effectiveness in limiting oxidation and their thermal resistance. Therefore, synthetic antioxidants such as butylated hydroxytoluene, butylated hydroxyanisole and *ter*-butyl hydroquinone are increasingly abandoned in favor of natural sources of antioxidants extracted from plants (Anwar , Jamil, Iqbal, & Sheikh, 2006; Womeni et al., 2016, Djikeng et al., 2017, Teboukeu, Djikeng, Klang, Karuna & Womeni, 2018). Investigation carried out by Djikeng *et al*. (2017) showed that methanolic extracts of ginger roots contain a significant amount of phenolic compounds (34.63 mg EAG/g of extracts), among which they detected the presence of ferulic acid and 6-gingerol, which have high antioxidant potential. They also demonstrated that the methanolic extract of ginger roots are as effective as BHT in delaying the oxidation of palm olein during storage in an oven at 70 ℃ (Schaal test) for 30 days. However, vegetable oils are mainly used during culinary processes such as frying, and there is no information related to the effect of this extract during this process. Consumer health being at the heart of society's concern, it's also important to investigate the impact of enriched oil on the vitals organs responsible for the balance and well-being of humans. This study was therefore conducted in order to evaluate the effect of ginger root extracts on the oxidative stability of palm olein during frying and to determine the impact of fried oils on some biochemical parameters of albino Wistar rats.

2. Material and Methods

2.1 Material

Ginger roots and unripe plantain (*Musa spp.*) used for frying were purchased at the central market of Bafoussam. Palm olein without additives was purchased from SCS/RAFCA (Société de Raffinerie du Cameroun) in Bafoussam, West Cameroon. The animals were purchased from the animal house of the Biochemistry Department, University of Dschang. All reagents and chemicals used were of analytical grade.

2.2 Methods

2.2.1 Preparation of Methanolic Extracts of Ginger Roots

The preparation of the methanolic extracts of ginger roots followed the method of Djikeng et al. (2017) with slight modifications. The fresh roots were cleaned and dried in an oven at 50 ℃ for 48 h. Then they were ground and sieved. 250 g of the obtained powder was macerated at room temperature in 1 L of methanol with regular stirring for 48 h. The solution was then filtered using Whatman papers No. 1. The filtrates obtained were subjected to rotary evaporation at 40 ℃ under reduced pressure using a "Buchi" evaporator, to eliminate the solvent. The solvent residue was removed by drying the extract in an oven at 45 ℃.

2.2.2 Enrichment of Palm Olein with Extract

The method used for the incorporation of extracts into palm olein was that of Djikeng et al. (2017) with some modifications. The concentrated extract was dissolved in 5 mL of methanol and added individually into 1.5 Kg of preheated palm olein (50 ℃ for 3 h) at different concentrations (1000, 1400 and 1800 ppm). Butylhydroxytoluene was used at its recommended concentration (200 ppm) (Duh & Yen, 1997) and served as a positive control to compare the stabilising effect of the extracts. Palm olein without additives was prepared as previously described and served as a negative control. Subsequently, the oil samples were shaken manually and vigorously for three hours before being placed without cover in an oven at 45 ℃ for 48 h as described by Djikeng et al (2017), to reduce the amount of methanol added. There were a total of five (05) different oil samples, all of which were used for the rest of the experiment.

2.2.3 Frying Plantain Chips

Frying was carried out according to the protocol of Leong, Mustafa, Das, & Jaarin, (2010) with slight modifications. 100 g of fresh oil from each sample was collected prior to the start of frying. A Rowenta electric fryer was used to fry 50 g of unripe plantain that were previously cleaned and sliced into small pieces. Frying took place for 3 min at 180 ℃, and the plantain chips were removed from the oil. The hot oil was left to cool at room temperature for 5 hours. 100 g of oil sample was collected after each cycle. The pre-cooled oil was used to fry another batch of plantain without adding new oil. Oil samples used in quality analysis were those collected

after 0, 1, 5, 8, 10 and 15 frying cycles. All oil samples (stabilised and non-stabilised) were processed similarly. The oxidative tests were performed on all the oil samples collected while only the samples collected at 0, 5, 10 and 15 frying cycles were used during feeding of the rats. The in vivo test consisted of administering the different oil samples obtained at 0, 5, 10 and 15 cycles to rats by dietary supplementation for 30 days (paragraph 2.2.5).

2.2.4 Determination of Oxidative Parameters of Oil Samples

The determination of the peroxide value of oil samples was done according to the standard spectrophotometric method of IDF, 74A: 1991 (IDF, 1991). The anisidine value was assessed according to the procedure of the official American Oil Chemists' Society method CD 18-90 (AOAC, 2003). The total oxidation value (TOTOX) were calculated using the following equation: TOTOX=2PV+AV according to Shahidi & Wanasundara, (2008).

2.2.5 Treatment of the Animals and Preparation of Their Feed

One hundred and five (105) rats aged between 7 and 9 weeks and weighing between 150 and 170 g, were randomly divided into 21 groups of 5 animals each as shown in Table 1. The tests were carried out according to the protocol described by OECD (2008). The animals randomly divided into different groups underwent a 7-day of acclimatation during which all consumed the staple food and abundant water. This feed consisted of maize meal (68%), soybean meal (20%), fish meal (10%), bone meal (1%), table salt (0.8%) and vitamin complex (0.1%). The different oil samples were administered to the animals by dietary supplementation at a rate of 2 mL of oil in 100 g of food. All animals had unlimited access to food and water, and the food formulation was done on a daily basis to avoid fermentation.

Table 1: Food composition of the different animal groups

Groups	Codes	Diet
1	Neutral control	staple food (SF)
2	PO	SF + Palm Olein without additives at 0 frying time
3	PO+BHT	SF + Palm Olein enriched with 200 ppm BHT at 0 frying time
4	PO+ERG1000	SF + Palm Olein enriched with 1000 ppm ginger root extract at 0 frying time
5	PO+ ERG1400	SF + Palm Olein enriched with 1400 ppm ginger root extract at 0 frying time
6	PO+ ERG1800	SF + Palm Olein enriched with 1800 ppm ginger root extract at 0 frying time
7	5PO	SF + Palm Olein without additives at 5 frying time
8	5PO+BHT	SF + Palm Olein enriched with 200 ppm BHT at 5 frying time
9	5PO+ ERG1000	SF + Palm Olein enriched with 1000 ppm ginger root extract at 5 frying time
10	5PO+ ERG1400	SF + Palm Olein enriched with 1400 ppm ginger root extract at 5 frying time
11	5PO+ ERG1800	SF + Palm Olein enriched with 1800 ppm ginger root extract at 5 frying time
12	10PO	SF + Palm Olein without additives at 10 frying time
13	10PO+BHT	SF + Palm Olein enriched with 200 ppm BHT at 10 frying time
14	10PO+ ERG1000	SF + Palm Olein enriched with 1000 ppm ginger root extract at 10 frying time
15	10PO+ ERG1400	SF + Palm Olein enriched with 1400 ppm ginger root extract at 10 frying time
16	10PO+ ERG1800	SF + Palm Olein enriched with 1800 ppm ginger root extract at 10 frying time
17	15PO	SF + Palm Olein without additives at 15 frying time
18	15PO+BHT	SF + Palm Olein enriched with 200 ppm BHT at 15 frying time
19	15PO+ ERG1000	SF + Palm Olein enriched 1000 ppm ginger root extract at 15 frying time
20	15PO+ ERG1400	SF + Palm Olein enriched with 1400 ppm ginger root extract at 15 frying time
21	15PO+ ERG1800	SF + Palm Olein enriched with 1800 ppm ginger root extract at 15 frying time

PO: palm olein; BHT: butylated hydroxytoluene; ERG: root ginger extract; 5, 10, and 15: number of frying time; SF: staple food.

The experiment lasted for 30 days during which all animals were weighed weekly. On the last day of the test, the rats were weighed and then, anaesthetised with chloroform vapour and the blood collected by cardiac puncture in two tubes. The tubes with anticoagulant (EDTA) were used to determine the haematological parameters while those without anticoagulant were used for the determination of the biochemical parameters. The organs (liver and kidneys) were also collected and the mass gains and relative masses of the organs of each rat calculated as follow.

$$Gm=Mf-Mi \quad (1)$$

Where, Gm= mass gain, Mf=final mass on the day of sacrifice and Mi=initial mass at the start of the experiment.

$$Rm\ (\%) = Mf/Mo \times 100 \quad (2)$$

Where, Rm=relative mass of the organ, Mf=mass of the animal on the day of sacrifice and Mo=mass of the organ.

2.2.6 Determination of Biochemical Parameters

2.2.6.1 Determination of Haematological Parameters

Hematological analyses were performed on blood samples taken in Ethylene Diamine Tetraacetic Acid (EDTA) tubes by a blood count using an impedance hematology automate “SFRI H18 LIGHT”.

2.2.6.2 Determination of Biochemical Parameters

The blood samples taken from the dry tubes were centrifuged for 15 minutes at 3000 rpm. The supernatant (serum) was then collected to determine the biochemical parameters. The biochemical parameters assessed were serum transaminase levels (ALAT/ASAT), creatinine, protein levels, total cholesterol and triglyceride levels, LDL-cholesterol and HDL-cholesterol levels and atherogenicity index. With the exception of the total protein level which was determined with the BIOLABO kit, the determination of all other parameters was done with the SPINREACT kits. The LDL-cholesterol level of each rat was calculated according to the Friedman formula:

$$\text{Chol LDL}=\text{Cholt}-\text{Chol HDL}-(\text{Trig}/5) \quad (3)$$

Where, Chol LDL=Low density lipoprotein, Cholt=Total cholesterol, CholHDL=High density lipoprotein and Trig=Total triglycerides.

For the determination of the atherogenicity index (AI) of each rat, the calculation was done as follows:

$$\text{AI}=(\text{Total cholesterol})/(\text{HDL cholesterol}) \quad (4)$$

2.2.7 Statistical Analysis

All results were subjected to the analysis of variance (ANOVA) test using SPSS software version 23.0. The Waller-Duncan test was used to calculate the means and standard deviations for each parameter. The results were considered statistically significant for values of $p<0.05$.

3. Results and Discussion

3.1 Results

3.1.1 Oxydative Parameters of Oil Samples

Table 2 shows the variations in oxidative parameters of different oil samples during frying. In general, the amount of hydroperoxide increased significantly in all oil samples between the initial (T0) and the end of frying (T15). The peroxide values of oils samples supplemented with the plant extract increased significantly ($p<0.05$) between the beginning (T0) and the fifth (5^{th}) frying cycle, then decreased significantly ($p<0.05$) from the 8^{th} to the 15^{th} cycle where these hydroperoxide values were significantly lower compared to the positive control (palm olein enriched with BHT). Concerning the anisidine value, this parameter increased significantly ($p<0.05$) in all oil samples with the number of frying cycles. However, the increase in anisidine value in the oils enriched with the extract was significantly lower ($p<0.05$) than that of the negative control (palm olein without antioxidant). Generally, a significant increase ($p<0.05$) in total oxidation value of different oil samples was observed with the number of frying cycles. The increase in total oxidation values in the enriched oil samples was significantly ($p<0.05$) lower compared to the negative control. On the other hand, the oils enriched with extract showed low and comparable ($p\geq0.05$) total oxidation value compared to the positive control. Furthermore, the values of this parameter significantly ($p<0.05$) decreased in oil samples with the increase of the concentration of extract. At concentration 1800 ppm, ginger root extract was best in delaying palm olein oxidation compared to the BHT.

Table 2. Effect of frying on the quality of different oil samples

Parameters	Samples	Number of frying cycles					
		0	1	5	8	10	15
Peroxide (meq O_2/Kg)	PO	$2.20 \pm 0.03^{e}_{A}$	$4.02 \pm 0.02^{e}_{B}$	$4.24 \pm 0.05^{b}_{BC}$	$5.60 \pm 0.02^{e}_{E}$	$5.27 \pm 0.20^{cd}_{D}$	$4.44 \pm 0.13^{bc}_{C}$
	PO+BHT	$1.15 \pm 0.00^{d}_{A}$	$1.63 \pm 0.00^{b}_{C}$	$1.55 \pm 0.02^{a}_{B}$	$3.73 \pm 0.02^{c}_{D}$	$4.97 \pm 0.01^{b}_{E}$	$7.03 \pm 0.03^{d}_{F}$
	PO+GRE1000	$0.83 \pm 0.01^{b}_{A}$	$1.82 \pm 0.02^{c}_{B}$	$7.08 \pm 0.46^{d}_{D}$	$5.16 \pm 0.00^{d}_{C}$	$5.05 \pm 0.03^{bc}_{C}$	$4.75 \pm 0.16^{c}_{C}$
	PO+GRE1400	$0.56 \pm 0.01^{a}_{A}$	$2.28 \pm 0.00^{d}_{B}$	$6.06 \pm 0.08^{c}_{F}$	$2.86 \pm 0.04^{b}_{C}$	$5.39 \pm 0.08^{d}_{E}$	$4.20 \pm 0.13^{b}_{D}$
	PO+GRE1800	$0.90 \pm 0.01^{c}_{A}$	$1.46 \pm 0.04^{a}_{B}$	$6.04 \pm 0.04^{c}_{F}$	$2.76 \pm 0.00^{a}_{D}$	$2.29 \pm 0.14^{a}_{C}$	$3.25 \pm 0.28^{a}_{E}$
p-anisidine	PO	$9.31 \pm 0.32^{d}_{A}$	$15.15 \pm 0.07^{d}_{B}$	$24.55 \pm 0.33^{d}_{D}$	$21.05 \pm 0.04^{d}_{C}$	$25.42 \pm 0.17^{d}_{E}$	$33.81 \pm 0.13^{d}_{F}$
	PO+BHT	$5.78 \pm 0.24^{b}_{A}$	$8.68 \pm 0.17^{b}_{B}$	$12.21 \pm 0.26^{c}_{C}$	$12.14 \pm 0.07^{b}_{C}$	$13.17 \pm 0.02^{a}_{D}$	$14.16 \pm 0.07^{a}_{E}$
	PO+GRE1000	$6.73 \pm 0.13^{c}_{A}$	$9.22 \pm 0.31^{c}_{B}$	$8.83 \pm 0.12^{b}_{B}$	$14.67 \pm 0.15^{c}_{C}$	$15.73 \pm 0.16^{c}_{D}$	$16.55 \pm 0.59^{b}_{E}$
	PO+GRE1400	$6.91 \pm 0.08^{c}_{B}$	$6.12 \pm 0.16^{a}_{A}$	$8.81 \pm 0.05^{b}_{C}$	$11.43 \pm 0.14^{a}_{D}$	$14.36 \pm 0.02^{b}_{E}$	$17.58 \pm 0.22^{c}_{F}$
	PO+GRE1800	$2.42 \pm 0.11^{a}_{A}$	$9.27 \pm 0.00^{c}_{C}$	$7.76 \pm 0.08^{a}_{B}$	$11.44 \pm 0.56^{a}_{D}$	$15.31 \pm 0.43^{c}_{E}$	$17.47 \pm 0.33^{c}_{F}$
TOTOX	PO	$13.71 \pm 0.24^{c}_{A}$	$23.19 \pm 0.03^{d}_{B}$	$33.04 \pm 0.43^{d}_{D}$	$32.25 \pm 0.10^{d}_{C}$	$35.98 \pm 0.23^{d}_{E}$	$42.71 \pm 0.40^{c}_{F}$
	PO+BHT	$8.08 \pm 0.25^{b}_{A}$	$11.94 \pm 0.19^{b}_{B}$	$15.31 \pm 0.21^{a}_{C}$	$19.60 \pm 0.11^{b}_{D}$	$23.12 \pm 0.00^{b}_{E}$	$28.24 \pm 0.00^{b}_{F}$
	PO+GRE1000	$8.39 \pm 0.16^{b}_{A}$	$12.86 \pm 0.36^{c}_{B}$	$22.99 \pm 0.81^{c}_{C}$	$24.99 \pm 0.16^{c}_{D}$	$25.83 \pm 0.10^{c}_{D}$	$27.09 \pm 0.10^{b}_{E}$
	PO+GRE1400	$8.03 \pm 0.04^{b}_{A}$	$10.69 \pm 0.14^{a}_{B}$	$20.94 \pm 0.22^{b}_{D}$	$17.16 \pm 0.06^{a}_{C}$	$25.14 \pm 0.20^{c}_{E}$	$24.95 \pm 0.86^{a}_{E}$
	PO+GRE1800	$4.23 \pm 0.15^{a}_{A}$	$12.19 \pm 0.08^{b}_{B}$	$19.84 \pm 0.17^{b}_{D}$	$16.96 \pm 0.54^{a}_{C}$	$19.90 \pm 0.71^{a}_{E}$	$23.97 \pm 0.91^{a}_{E}$

Data are presented as mean (± SD) (n = 2) (a-d) Means within each column for each parameter with different superscripts are significantly (p<0.05) different. (A-F) Means within each line for each parameter with different superscripts are significantly (p<0.05) different. PO: palm olein without antioxidant; PO+BHT 200ppm: palm olein containing BHT (butylated hydroxytoluene) as antioxidant at concentration of 200 ppm; PO+ERG1000= palm olein enriched with ginger root extract at 1000 ppm; PO+ERG1400= palm olein enriched with ginger root extract at 1400 ppm; PO+ERG1800= palm olein enriched with ginger root extract at 1800 ppm; TOTOX: total oxidation value.

3.1.2 Effect of the Different Oil Samples on the Mass Gains of the Animals and the Relative Masses of Their Organs

The body masses of the animals increased significantly between the beginning and the end of the experiment (table 3). However, the mass gains in groups fed with different frying oil samples were significantly ($p<0.05$) lower compared to those of their counterparts fed with oil samples free from additives. As a result, rats fed with oils supplemented with ginger extract and subjected to 15 frying cycles showed significantly higher ($p<0.05$) mass gain compared to rats fed with palm olein without additive after 15 frying cycles. Regardless of the type of oil sample consumed, no significant ($p>0.05$) difference was observed between the relative organ masses of the test animals and the neutral control.

Table 3. Mass gains of the animals and the relative masses of their organs (liver and kidneys)

Groups	Parameters				
	Mi (g)	Mf (g)	Gm (g)	RmL (%)	RmK (%)
Neutral control	159.80 ± 3.89^{a}	228.20 ± 5.63^{cdef}	68.40 ± 5.50^{bcdefg}	3.35 ± 0.14^{a}	0.32 ± 0.03^{a}
PO	160.80 ± 5.11^{a}	238.40 ± 5.17^{fgh}	77.60 ± 6.06^{fghij}	3.38 ± 0.34^{a}	0.31 ± 0.02^{a}
PO+BHT	155.00 ± 3.60^{a}	244.60 ± 3.57^{h}	89.60 ± 6.22^{j}	3.32 ± 0.33^{a}	0.30 ± 0.02^{a}
PO+GRE1000	156.20 ± 1.78^{a}	241.20 ± 4.60^{gh}	85.00 ± 5.74^{hij}	3.45 ± 0.40^{a}	0.32 ± 0.03^{a}
PO+ GRE1400	155.80 ± 6.14^{a}	231.40 ± 4.03^{defg}	75.60 ± 8.61^{efghi}	3.54 ± 0.33^{a}	0.32 ± 0.04^{a}
PO+ GRE1800	159.40 ± 5.41^{a}	244.20 ± 7.98^{h}	84.80 ± 12.25^{hij}	3.68 ± 0.27^{a}	0.32 ± 0.01^{a}
5PO	155.40 ± 3.84^{a}	219.00 ± 7.84^{bc}	63.60 ± 11.14^{bcde}	3.50 ± 0.28^{a}	0.32 ± 0.02^{a}
5PO+BHT	154.20 ± 3.34^{a}	241.40 ± 4.39^{gh}	87.20 ± 6.87^{ij}	3.29 ± 0.18^{a}	0.29 ± 0.00^{a}
5PO+ GRE1000	162.20 ± 5.21^{a}	224.40 ± 4.21^{bcd}	62.20 ± 6.30^{bc}	3.88 ± 0.45^{a}	0.31 ± 0.04^{a}
5PO+ GRE1400	156.00 ± 3.93^{a}	235.60 ± 8.96^{efgh}	79.60 ± 7.79^{ghij}	3.41 ± 0.41^{a}	0.30 ± 0.04^{a}
5PO+ GRE1800	158.60 ± 3.78^{a}	234.60 ± 7.63^{defgh}	76.00 ± 10.55^{efghi}	3.34 ± 0.28^{a}	0.30 ± 0.03^{a}
10PO	159.80 ± 3.19^{a}	226.40 ± 4.56^{bcde}	66.60 ± 5.50^{bcdef}	3.38 ± 0.11^{a}	0.31 ± 0.03^{a}
10PO+BHT	155.40 ± 2.50^{a}	217.60 ± 9.39^{b}	62.20 ± 11.21^{bc}	3.42 ± 0.25^{a}	0.32 ± 0.03^{a}
10PO+ GRE1000	158.40 ± 8.67^{a}	230.20 ± 9.47^{def}	71.80 ± 6.30^{cdefg}	3.37 ± 0.15^{a}	0.30 ± 0.01^{a}
10PO+ GRE1400	155.80 ± 3.49^{a}	230.80 ± 4.54^{def}	75.00 ± 4.18^{defghi}	3.27 ± 0.12^{a}	0.31 ± 0.01^{a}
10PO+ GRE1800	166.00 ± 3.93^{a}	232.60 ± 6.14^{defg}	66.60 ± 4.82^{bcdef}	3.18 ± 0.09^{a}	0.30 ± 0.00^{a}
15PO	159.20 ± 3.27^{a}	198.60 ± 4.21^{a}	39.40 ± 4.33^{a}	4.15 ± 0.29^{a}	0.39 ± 0.02^{a}
15PO+BHT	159.80 ± 3.56^{a}	232.80 ± 5.97^{defg}	73.00 ± 8.30^{cdefgh}	3.47 ± 0.10^{a}	0.33 ± 0.01^{a}
15PO+ GRE1000	155.00 ± 5.95^{a}	217.80 ± 6.37^{b}	62.80 ± 10.91^{bcd}	3.68 ± 0.15^{a}	0.32 ± 0.03^{a}
15PO+ GRE1400	160.60 ± 3.04^{a}	217.00 ± 8.68^{b}	56.40 ± 8.01^{b}	3.38 ± 0.46^{a}	0.35 ± 0.02^{a}
15PO+ GRE1800	157.80 ± 1.30^{a}	225.20 ± 3.56^{bcd}	67.40 ± 2.60^{bcdefg}	3.22 ± 0.26^{a}	0.32 ± 0.04^{a}

Data are expressed as mean ±SD. n=5 (a-j) Values for a given group in a column followed by a different letter (a-f) are significantly different according to Waller–Duncan's multiple comparison test (p<0.05). PO: palm olein; BHT: butylated hydroxytoluene; GRE: ginger root extract; 5; 10; and 15: number of frying cycles Gm= mass gain, Mf=final mass on the day of sacrifice and Mi=initial mass at the start of the experiment, RmK=relative mass of the kidney, RmL= relative mass of the liver.

3.1.3 Effect of Different oil Samples on the Haematological Profile or Wistar Rats

The haematological profile differed between the groups depending on the oil sample consumed (table 4). Animals fed with different oil samples (fresh and fried) without antioxidants showed a significant increase ($p<0.05$) in white blood cell count, mean corpuscular volume, and mean corpuscular haemoglobin concentration. In contrast, consumption of oils supplemented with the plant extract (fresh and fried) resulted in a significant ($p<0.05$) decrease in these parameters. Regarding the number of blood platelets, its values decreased significantly ($p<0.05$) with the number of frying cycles. Therefore, animals fed with oils enriched with plant extracts have significantly ($p<0.05$) higher blood platelet concentrations compared to animals fed with different oil samples without antioxidants. Furthermore, the highest blood platelet concentrations (491.60 ± 15.04 and 480.60 ± 16.10 10^3/µL) were recorded with PO+GRE1800 and 5PO+GRE1800 groups.

Table 4. Effect of different oil samples on the changes in white blood cells, red blood cells and some figurative elements in the blood of rats

Groups	**Parameters**						
	WBC (10^3/µL)	**RBC (10^6/µL)**	**HGB (g/dL)**	**HCT (%)**	**MCV (FL)**	**MCHC (g/dL)**	**PLT (10^3/µL)**
Neutral control	3.04 ± 1.11^{a}	8.96 ± 0.37^{a}	16.02 ± 0.76^{a}	52.48 ± 1.75^{a}	62.86 ± 4.02^{bcd}	34.26 ± 3.28^{abcde}	476.40 ± 12.34^{jk}
PO	4.26 ± 1.86^{ab}	7.50 ± 0.83^{a}	15.52 ± 0.59^{a}	46.94 ± 2.09^{a}	61.92 ± 1.22^{abc}	34.18 ± 1.72^{abcde}	468.60 ± 11.45^{jk}
PO+BHT	4.26 ± 2.01^{ab}	7.29 ± 0.85^{a}	15.46 ± 0.87^{a}	46.22 ± 4.46^{a}	63.08 ± 2.89^{bcd}	33.84 ± 2.55^{abcd}	506.60 ± 20.15^{l}
PO+GRE1000	4.00 ± 1.08^{ab}	7.82 ± 0.46^{a}	15.82 ± 0.72^{a}	49.04 ± 2.72^{a}	61.82 ± 1.49^{abc}	33.82 ± 3.29^{abcd}	476.20 ± 11.36^{jk}
PO+ GRE1400	4.04 ± 1.16^{ab}	7.94 ± 0.55^{a}	15.92 ± 0.42^{a}	47.04 ± 3.43^{a}	62.30 ± 1.00^{abcd}	32.82 ± 2.35^{ab}	452.60 ± 10.40^{hi}
PO+ GRE1800	4.86 ± 1.26^{abc}	7.58 ± 0.67^{a}	16.76 ± 0.95^{a}	47.12 ± 3.14^{a}	63.16 ± 3.40^{bcd}	34.14 ± 2.78^{abcde}	491.60 ± 15.04^{kl}
5PO	5.24 ± 1.89^{abcd}	7.33 ± 1.00^{a}	15.40 ± 1.61^{a}	45.84 ± 2.03^{a}	64.38 ± 2.84^{cd}	34.46 ± 1.31^{abcde}	402.60 ± 13.01^{ef}
5PO+BHT	3.90 ± 1.45^{ab}	7.42 ± 0.96^{a}	15.64 ± 1.23^{a}	45.42 ± 4.58^{a}	60.92 ± 2.48^{abc}	33.20 ± 0.89^{abc}	433.40 ± 19.60^{ghi}
5PO+ GRE1000	4.02 ± 1.27^{ab}	7.69 ± 0.76^{a}	16.20 ± 0.73^{a}	49.72 ± 0.98^{a}	61.30 ± 1.94^{abc}	34.00 ± 2.58^{abcd}	436.00 ± 16.83^{gh}
5PO+ GRE1400	5.54 ± 0.96^{bcde}	7.76 ± 0.91^{a}	16.86 ± 0.35^{a}	49.98 ± 3.81^{a}	63.16 ± 1.26^{bcd}	33.88 ± 2.99^{abcd}	442.20 ± 17.81^{h}
5PO+ GRE1800	3.06 ± 0.87^{a}	6.64 ± 1.80^{a}	15.10 ± 1.08^{a}	43.40 ± 8.54^{a}	57.58 ± 4.91^{a}	30.68 ± 1.86^{a}	480.60 ± 16.10^{kl}
10PO	7.42 ± 2.69^{defg}	6.78 ± 0.82^{a}	15.80 ± 0.68^{a}	47.42 ± 2.04^{a}	66.96 ± 5.32^{de}	36.88 ± 2.51^{cdef}	360.00 ± 19.88^{cd}
10PO+BHT	7.32 ± 1.40^{def}	8.30 ± 0.49^{a}	17.72 ± 1.56^{a}	53.20 ± 4.65^{a}	60.86 ± 2.95^{abc}	35.76 ± 2.16^{bcdef}	403.40 ± 15.05^{ef}
10PO+ GRE1000	5.96 ± 0.9^{bcdef}	6.86 ± 1.20^{a}	15.28 ± 1.74^{a}	45.88 ± 5.23^{a}	62.92 ± 4.28^{bcd}	37.60 ± 5.54^{def}	386.20 ± 22.86^{de}
10PO+ GRE1400	7.60 ± 0.85^{defg}	8.30 ± 0.88^{a}	15.92 ± 2.45^{a}	47.80 ± 7.40^{a}	60.64 ± 4.79^{abc}	34.84 ± 3.57^{bcde}	407.80 ± 15.44^{efg}
10PO+ GRE1800	4.08 ± 1.17^{ab}	7.71 ± 1.45^{a}	15.90 ± 2.53^{a}	47.70 ± 7.61^{a}	59.20 ± 2.08^{ab}	34.64 ± 1.37^{abcde}	413.80 ± 13.82^{fgh}
15PO	9.80 ± 2.96^{g}	6.02 ± 2.00^{a}	13.02 ± 2.84^{a}	41.40 ± 3.69^{a}	70.44 ± 3.80^{e}	39.46 ± 1.75^{f}	303.40 ± 22.64^{a}
15PO+BHT	8.24 ± 1.14^{eg}	7.74 ± 1.20^{a}	15.14 ± 2.47^{a}	45.50 ± 7.51^{a}	58.34 ± 2.92^{ab}	36.30 ± 1.68^{bcdef}	348.20 ± 13.49^{bc}
15PO+ GRE1000	7.04 ± 1.00^{cdef}	8.16 ± 0.86^{a}	16.42 ± 2.03^{a}	49.36 ± 6.14^{a}	58.88 ± 2.49^{ab}	38.06 ± 1.71^{ef}	322.60 ± 17.38^{ab}
15PO+ GRE1400	7.26 ± 0.75^{cdef}	8.22 ± 0.59^{a}	17.22 ± 2.88^{a}	51.70 ± 8.61^{a}	59.30 ± 3.01^{ab}	37.40 ± 2.32^{def}	386.60 ± 16.47^{e}
15PO+ GRE1800	7.68 ± 1.25^{efg}	7.74 ± 0.39^{a}	16.44 ± 1.02^{a}	48.86 ± 2.19^{a}	58.86 ± 1.95^{ab}	38.08 ± 0.94^{ef}	401.00 ± 10.51^{ef}

Data are expressed as mean ±SD. n=5. Values for a given group in a column followed by a different letter (a-f) are significantly different according to Waller–Duncan's multiple comparison test ($p<0.05$). PO: palm olein; BHT: butylated hydroxytoluene; GRE: ginger root extract; 5; 10; and 15: number of frying cycles; WBC: white blood cell count. RBC: red blood cell count; HGB: hemoglobin; HCT: hematocrit; MCV: mean corpuscular hemoglobin; MCHC: mean corpuscular hemoglobin concentration; PLT: platelet count.

3.1.4 Effect of the Different Oil Samples on the Lipid Profile of Wistar Rats

Table 5 shows the changes in lipid profile of the animals. Globally, there was a significant increase ($p<0.05$) in serum triglyceride, total cholesterol and LDL-cholesterol concentrations, followed by a decrease in HDL-cholesterol of the animals according to the rancidity level of the oil consumed. Nevertheless, the groups of animals fed with fried oils previously enriched with plant extracts showed a significant ($p<0.05$) decrease in total cholesterol, LDL-cholesterol and triglyceride concentrations followed by a significant ($p<0.05$) increase in HDL-cholesterol compared to the groups of animals fed with different oil samples, fresh and fried without antioxidants. In addition, the HDL-cholesterol concentrations of the groups that consumed oil samples supplemented with plant extracts at 0 and 5 frying cycles respectively, were high and comparable ($p\geq0.05$) to that of the neutral control group. The highest values (69.29 ± 3.38 and 68.46 ± 3.91 mg/dL) were observed in the PO+GRE1800 and 5PO+GRE1800 groups. The atherogenecity index of animals fed the plant extract-enriched oil samples was significantly ($p<0.05$) lower compared to those fed with the oil without antioxidants and the oils containing BHT. Furthermore, consumption of oils enriched with plant extracts in their fresh and fried state after 5 frying cycles led to a low atherogenecity index, comparable ($p\geq0.05$) to that of the neutral control group.

Table 5. Effect of the different oil samples on the lipid profile of the animals

Groups	Parameters				
	TRIG (mg/dL)	T-CHOL (mg/dL)	LDL (mg/dL)	HDL (mg/dL)	AI
Neutral control	85.63 ±3.30^{a}	69.61 ±1.85^{a}	18.91 ±5.57^{a}	67.82 ±4.06^{hi}	1.02 ±0.07^{a}
PO	167.09 ±3.12^{f}	103.82 ±2.90^{f}	77.65 ±3.14^{g}	59.58 ±2.03^{g}	1.74 ±0.06^{bcde}
PO+BHT	144.78 ±2.73^{e}	91.20 ±3.74^{d}	53.52 ±8.90^{c}	66.63 ±6.47^{hi}	1.38 ±0.16^{abc}
PO+GRE1000	120.11 ±5.79^{b}	84.70 ±2.56^{c}	42.91 ±4.67^{b}	65.81 ±4.82^{ghi}	1.29 ±0.09^{abc}
PO+ GRE1400	116.61 ±4.12^{b}	82.76 ±2.52^{c}	40.64 ±3.60^{b}	65.44 ±4.26^{ghi}	1.26 ±0.06^{abc}
PO+ GRE1800	133.74 ±4.68^{c}	77.23 ±4.55^{b}	35.61 ±4.93^{b}	69.29 ±3.38^{i}	1.11 ±0.05^{ab}
5PO	183.09 ±2.67^{g}	109.20 ±1.13^{g}	95.57 ±4.95^{h}	50.25 ±5.20^{f}	2.19 ±0.22^{def}
5PO+BHT	142.30 ±2.667^{de}	97.25 ±1.87^{e}	63.56 ±3.69^{de}	62.15 ±2.88^{gh}	1.56 ±0.08^{abcd}
5PO+ GRE1000	144.67 ±4.35^{e}	107.26 ±2.86^{fg}	69.47 ±7.65^{ef}	66.72 ±4.86^{hi}	1.61 ±0.14^{abcd}
5PO+ GRE1400	136.67 ±3.32^{cd}	94.78 ±2.86^{de}	56.31 ±4.24^{cd}	65.81 ±3.25^{ghi}	1.44 ±0.08^{abc}
5PO+ GRE1800	123.04 ±3.57^{b}	79.85 ±2.69^{bc}	35.99 ±4.11^{b}	68.46 ±3.91^{hi}	1.16 ±0.07^{ab}
10PO	218.02 ±4.87^{i}	108.83 ±2.63^{g}	118.47 ±2.59^{i}	33.95 ±4.26^{cd}	3.23 ±0.34^{g}
10PO+BHT	182.08 ±4.35^{g}	106.51 ±3.17^{fg}	99.36 ±4.99^{h}	43.56 ±4.52^{e}	2.46 ±0.28^{f}
10PO+ GRE1000	207.09 ±7.18^{h}	108.08 ±2.08^{fg}	119.02 ±3.63^{i}	30.48 ±4.36^{bc}	3.60 ±0.50^{gh}
10PO+ GRE1400	202.70 ±5.23^{h}	95.98 ±48de	96.15 ±4.96^{h}	40.36 ±4.21^{de}	2.40 ±0.29^{ef}
10PO+ GRE1800	188.16 ±4.55^{g}	84.18 ±3.43^{c}	76.96 ±2.64^{fg}	44.85 ±4.02^{ef}	1.88 ±0.12^{cdef}
15PO	278.87 ±6.80^{m}	115.70 ±4.22^{h}	150.97 ±6.8^{l}	20.50 ±3.46^{a}	5.81 ±1.32^{i}
15PO+BHT	226.92 ±6.50^{j}	115.92 ±3.53^{h}	139.98 ±3.16^{k}	21.32 ±2.53^{a}	5.49 ±0.62^{i}
15PO+ GRE1000	256.78 ±5.63^{l}	115.03 ±2.75^{h}	143.41 ±6.95^{kl}	22.97 ±4.06^{a}	5.14 ±1.00^{i}
15PO+ GRE1400	243.60 ±4.75^{k}	106.06 ±4.10^{fg}	129.80 ±5.43^{j}	24.98 ±1.93^{ab}	4.26 ±0.39^{h}
15PO+ GRE1800	199.54 ±4.63^{h}	104.87 ±2.77^{fg}	114.57 ±2.50^{i}	30.20 ±2.87^{bc}	3.49 ±0.29^{g}

Data are expressed as mean ±SD, n=5. Values for a given group in a column followed by a different letter (a-m) are significantly different according to Waller–Duncan's multiple comparison test ($p<0.05$). PO: palm olein; BHT: butylated hydroxytoluene; GRE: ginger root extract; 5, 10, 15: number of frying cycles; TRIG: triglycerides; T-CHOL: total cholesterol LDL: low-density lipoprotein; HDL: high-density lipoprotein, AI: atherogenicity index

3.1.5 Effect of the Different Oil Samples on Liver and Kidney Parameters

In general, an increase in serum transaminases (ALAT/ASAT) and creatinine levels followed by a decrease in serum protein concentrations is observed in all groups of animals (table 6) proportionally to the oxidative state of the oil samples consumed. In particular, the groups of rats fed respectively with the diets containing fresh oil samples enriched with ginger extract of exhibited lower serum transaminase concentrations which was similar ($p\geq0.05$) to those of the neutral control group, and the lowest ASAT value (81.72 ± 3.19 UI/L) was observed in group PO+GRE1800. In contrast, the groups subjected to the diets supplemented with the fried oils without additives and oils enriched with BHT respectively, showed significantly ($p<0.05$) higher transaminase concentrations compared to the neutral control group.

The observations made in table 6 showed that the consumption of different oil samples enriched with plant extract in their fresh state (T_0), as well as after 5 and 10 frying cycles led to a non-significant ($p>0.05$) variation in serum total protein concentration in the different test groups compared to the neutral control group. It is also visible that, the consumption of all oil samples after 15 frying cycles led to a significant ($p<0.05$) decrease in serum protein in all test groups compared to the neutral control group. In addition, the PO+GRE1400, PO+GRE1800 and 5PO+1800GRE groups showed the best serum protein concentrations, namely: 63.22 ± 4.34; 63.53 ± 4.65; 63.53 ± 4.65 g/l respectively.

Creatinine (S-CREA) results (table 6) revealed that, compared to the neutral control group, the animals that consumed the fresh oil samples previously enriched with plant extracts presented significantly ($p<0.05$) lower values of S-CREA. On the other hand, a significant ($p<0.05$) decrease in creatinine was observed in animals groups fed with fried oils containing the extract compared to those that consumed the different samples of fried oils without antioxidant.

Table 6. Effect of different oil samples on ALAT/ASAT, total protein and creatinine concentration in serum animals

Groups	Parameters			
	ALAT (UI/L)	ASAT (UI/L)	S-PROT (g/L)	S-CREA (µmol/L)
Neutral control	23.45 ± 2.65^{a}	93.10 ± 3.53^{b}	60.35 ± 4.49^{fgh}	69.78 ± 4.65^{ef}
PO	39.72 ± 1.81^{h}	99.75 ± 4.94^{cd}	53.35 ± 3.03^{abcde}	65.13 ± 4.65^{de}
PO+BHT	31.67 ± 1.14^{def}	91.17 ± 3.23^{b}	61.95 ± 3.38^{fgh}	56.76 ± 2.08^{ab}
PO+GRE1000	29.75 ± 1.23^{cde}	93.10 ± 2.01^{b}	63.22 ± 4.34^{h}	54.90 ± 3.89^{a}
PO+ GRE1400	24.93 ± 1.75^{ab}	91.87 ± 4.24^{b}	62.70 ± 5.39^{gh}	53.97 ± 7.05^{a}
PO+ GRE1800	26.07 ± 1.56^{abc}	81.72 ± 3.19^{a}	63.53 ± 4.65^{h}	53.04 ± 2.54^{a}
5PO	52.15 ± 2.01^{j}	124.07 ± 1.89^{ij}	51.54 ± 4.89^{abc}	76.30 ± 2.54^{gh}
5PO+BHT	39.90 ± 2.66^{h}	105.35 ± 2.52^{de}	58.48 ± 5.08^{defgh}	61.41 ± 5.09^{bcd}
5PO+ GRE1000	33.25 ± 2.96^{ef}	108.50 ± 4.87^{ef}	52.56 ± 2.29^{abcd}	58.62 ± 2.54^{abc}
5PO+ GRE1400	23.45 ± 1.56^{a}	103.95 ± 3.46^{cde}	59.68 ± 5.61^{efgh}	$64.20 \pm \pm 2.08^{cde}$
5PO+ GRE1800	28.52 ± 1.91^{bcd}	99.22 ± 2.44^{c}	63.53 ± 4.65^{h}	56.76 ± 2.08^{ab}
10PO	61.07 ± 2.86^{k}	155.57 ± 4.76^{l}	48.42 ± 2.57^{a}	80.02 ± 5.09^{hi}
10PO+BHT	44.80 ± 0.73^{i}	113.75 ± 3.50^{fg}	57.04 ± 3.53^{cdefgh}	65.13 ± 4.65^{de}
10PO+ GRE1000	32.55 ± 4.34^{def}	121.45 ± 3.11^{hi}	55.52 ± 3.89^{bcdef}	84.67 ± 5.09^{ij}
10PO+ GRE1400	29.92 ± 4.34^{cde}	117.77 ± 5.72^{gh}	56.10 ± 2.28^{bcdefg}	78.16 ± 2.08^{gh}
10PO+ GRE1800	29.75 ± 4.41^{cde}	100.45 ± 3.11^{cd}	60.70 ± 3.80^{fgh}	64.20 ± 3.89^{cde}
15PO	75.08 ± 3.46^{l}	167.47 ± 2.28^{m}	48.01 ± 4.33^{a}	96.77 ± 2.08^{l}
15PO+BHT	49.87 ± 2.14^{j}	146.47 ± 2.80^{k}	51.17 ± 4.12^{abc}	69.78 ± 3.28^{ef}
15PO+ GRE1000	41.82 ± 2.99^{hi}	127.22 ± 4.35^{j}	49.66 ± 5.32^{ab}	87.46 ± 5.09^{jk}
15PO+ GRE1400	39.20 ± 4.92^{gh}	125.47 ± 2.19^{ij}	50.13 ± 6.05^{ab}	91.19 ± 2.54^{kl}
15PO+ GRE1800	35.00 ± 1.07^{fg}	116.90 ± 3.13^{gh}	52.40 ± 2.28^{abcd}	73.51 ± 2.08^{fg}

Data are expressed as mean ±SD, n=5. Values for a given group in a column followed by a different letter (a-m) are significantly different according to Waller–Duncan's multiple comparison test ($p<0.05$). PO: palm olein; BHT: butylated hydroxytoluene; GRE: ginger root extract; 5, 10, 15: number of frying cycles; ALT: Alanine transaminase; AST: Aspartate transaminase; S-PROT: serum protein; S-CREA: serum creatinine.

3.2 Discussion

The peroxide value is an indicator of the primary oxidative state of fats. The increase of this parameter in oil samples reflects the increased production of hydroperoxides. Indeed, very high temperatures such as those experienced by the oil during frying facilitate the formation of these compounds in two stages: initiation and propagation (Wu et al., 2019). Under these conditions, unsaturated fatty acids rapidly lose a hydrogen atom at the α-position on their side chains with the formation of alkyl radicals which react with triplet oxygen to produce peroxyl radicals, the latter will in turn abstract a hydrogen atom from another fatty acid in the medium and then form hydroperoxides (Leong, Ng, Jaarin, & Mustafa 2015). Therefore, the significant increase in peroxide value recorded with the positive control (OP+200BHT) marks the increased formation of hydroperoxide in this oil sample. On the other hand, the small increase of this parameter in the oil samples enriched with plant extracts could be explained by the free radical scavenging activity of phenolic compounds present in these extracts (Djikeng et al., 2017). However, it should be noted that hydroperoxides are unstable at high temperatures and decompose very rapidly to give rise to secondary oxidation compounds (Nayak, Dash, Rayaguru, & Krishnan, 2015). Thereby, peroxide value is an insufficient parameter to determine the rancidity status of oil during frying. These results are similar to those of Houhoula, Oreopoulou, & Tzia, (2003) and Guo et al. (2016) whose respective investigations showed that, the addition of ouregan at 2000 ppm in cottonseed oil and rosemary extract in palm olein at 120 ppm significantly reduces the peroxide value of these oils during frying of chips.

The anisidine value is used to assess the secondary oxidation state of a fat by detecting the aldehyde compounds 2, 4-dienals and 2-alkenals (Anwar et al., 2006). The increase of these different compounds in all oil samples during frying could be the result of the decomposition of primary products into secondary oxidation products. In this regard, the terminal phase of oxidation of unsaturated fatty acids is marked by the breaking of adjacent double bonds of hydroperoxides followed by the formation of hydrocarbons, aldehydes, alcohols and ketones (Leong et al., 2015; Nayak et al., *2015*). The increased formation of oxidation by-products in the negative control (PO) could be a consequence of the absence of antioxidant in this sample. On the other hand, the small increase in these products observed in the oil samples enriched with plant extracts would testify to the thermal resistance of the latter as well as to the antiradical action of the phenolic compounds present in them. In fact, previous work (Djikeng et al., 2017) attests to the presence of phenolic compounds such as ferulic acid and 6-gingerol in these extracts. The latter would therefore have acted by yielding their labile hydrogen to alkyl

radicals and peroxides, thus transforming them into more stable non-radical products. These results are in agreement with those of Jaswir, Man, & Kitts, (2000) who found that the use of rosemary and sage extracts in palm olein at 4000 ppm resulted in a reduction in the formation of secondary oxidation compounds during frying of crisps. They are also in agreement with those of Nor, Mohamed, Idris & Ismail, (2008) and Li *et al.* (2020) whose respective work showed that the addition of *Pandanus amaryllifolius* leaf extracts at 2000 ppm in palm olein and the addition of rosemary extracts at 2% in soybean oil led to a small increase in the anisidine value during the production of Chips.

The overall oxidative state of oil can be taken into account by determining its total oxidation value. This parameter provides information on both the formation and decomposition of hydroperoxides, and gives a better estimate of the overall weathering state of the oil (Womeni et al., 2016). The high rancidity of the negative control (PO) could be attributed to the absence of antioxidants in this oil. Therefore, during frying, secondary oxidation compounds are formed at an exponential level affecting the oxidative, olfactory and taste quality of the oil (Choe & Min, 2006). On the other hand, the small increase in the total oxidation value observed in oils enriched with plant extracts would be related to the antioxidant action of the phenolic compounds present, since phenolic compounds offer good oxidative stability to the oil under frying conditions as reported by Wu et al. (2019). Indeed, ferulic acid and 6-gingerol present in ginger root extracts are classified as type I antioxidants, whose particularity lies in the inactivation of peroxyl and hydroxyl radicals. Moreover, the activity of these extracts is concentration-dependent because the total oxidation values decrease with increasing extract concentrations. Decreases in total oxidation values were also observed by Jamilah, Man & Ching, (1998) and Solati & Baharin (2014). Their work focused on the use of citrus peel extract at 2000 ppm in palm olein for fish frying and the use of *Nigella Sativa* extract in palm olein and sunflower oil for the production of Chips respectively.

The administration of palm olein without additives at 15 frying cycles resulted in a delay in growth in the animals concerned compared to those in the different test groups. This could be explained by the fact that the oil administered to them was highly oxidised and therefore the free radicals would have irritated the intestinal walls of these animals, reducing their capacity to absorb fats and certain nutrients such as essential fatty acids and vitamins (Badr El Said et al., 2015). Oxidation could also have led to a decrease in nutrient availability by complexing them with free radicals (Hochgraf, Cogan & Mokady, 2000). Previous work has also reported that the consumption of oxidised vegetable oils is responsible for stunted growth in animals (Badr El Said et al., 2015; Ambreen, Siddiq & Hussain, 2020).

The blood count is the primary biological test used to screen for most haemopathies. White blood cells are cells involved in the body's immune reactions. They seek out, invade and destroy pathogens (viruses, bacteria, fungi, etc.) on a daily basis. Their production is a normal process, however, a sudden increase in the concentration of white blood cells as observed with animals from the negative control groups (10PO and 15PO), could indicate an inflammation caused by a state of stress in one or more organs by oxidation products (Mesembe et al., 2005). These results are in agreement with those of Ani et al. (2015) who found that consumption of thermooxidised palm oil for 28 days resulted in increased white blood cell counts in rats. In contrast to the animals fed with oils enriched with plant extracts, whose showed MCV and MCHC similar to neutral control group, those fed with oil samples without additives at 10 and 15 frying cycles showed very high MCV and MCHC. This could reflect a macrocytic anaemia caused by vitamin B12 or folate deficiency (Elleuch, 2004). It is possible that free radicals in the oils consumed by these respective groups of animals caused inflammation of the distal ileum or jejunum in the small intestine, resulting in malabsorption of vitamin B12, folic acid and other nutrients. However, the significant decrease observed in the test groups would be due to the low formation of oxidation products in the oil samples consumed. These results are in agreement with those of Mesembe et al. (2005) who showed that consumption of oxidised palm oil lead to a deterioration of haematological parameters of rats. There are also in agreement with those of Zeb & Khan (2019) who showed that, administration of alpha-tocopherol in oxidised olive oil-induced toxicity in rats leads to an improvement in their haematological parameters. Platelets or thrombocytes are components of blood that form clots in the event of haemorrhage in order to stop bleeding, and their synthesis takes place in the bone marrow from stem cells (Twomey et al., 2019). The decrease in blood platelet levels in rats fed with oxidised oils is thought to be due to free radical damage to these stem cells. However, the high blood platelet concentrations observed in the PO+GRE1800 and 5PO+GRE1800 groups can be explain by the presence of phenolic compounds in these sample oils, which would have consequently delayed the oxidation of these oil samples. These results are in line with those of Chacko & Rajamohan, (2011). The latter had found that the consumption of thermoxidized vegetable oils led to an alteration in platelet function in rats. However, they contradict those of Hamam & Eldalo, (2018). They found that vitamin E supplementation

had no impact on the adverse effects caused by frying oil in rats.

The lipid profile is a test to determine the risk of cardiovascular disease, it may be prescribed for individuals on high-fat diets. The increase in triglyceride concentration after ingestion of fried oil could be due to the presence of abundant free fatty acids in these oils, and their availability as an esterification substrate in the formation of these molecules (Shastry, Ambalal, Himanshu & Aswathanarayana, 2011). These results are in agreement with those of Rueda-Clausen et al. (2007) who found that consumption of fried palm oil increased triglyceride levels in humans. Several studies (Adam et al., 2008; Zeb and Khan 2019) suggest that consumption of frying oils may have a negative influence on the lipid profile leading to an increase in total and LDL cholesterol followed by a decrease in HDL cholesterol as found in this work, this could be attributed to the ingestion of oxidised LDL through fried oil samples. Indeed, LDL is rich in polyunsaturated fatty acids and is therefore very sensitive to free radical attacks. The oxidation of these molecules generally leads to the formation of oxidised LDL which are taken up by macrophages in which they accumulate to form foam cells (Duriez, 2004; Favier, 2006). The accumulation of foam cells in the vascular subendothelium contributes to the development of atherosclerotic plaques and the onset of atherosclerosis (Duriez, 2004). Chemical characterisation tests showed that oils enriched with plant extracts are weakly oxidised compared to non-enriched oils. This could explain the improved lipid parameters of animals consuming enriched oils with extract compared to those consuming the non-enriched oils. These results are in agreement with those of Shafaeizadeh et al. (2011) who showed that pectin improve the lipid parameters of rats fed a diet containing thermooxidized sunflower oil.

Serum transaminase variations (ALAT/ASAT) provide information on the pathological state of the liver. The increase in serum transaminases in the different groups following the consumption of frying oils without antioxidant would be the consequence of lipid peroxidation reactions at the level of the hepatocyte membranes caused by the oxidation products present in these oils. Indeed, the highly reactive free radicals have as their preferred substrates the phospholipids of the membrane bilayer. An attack on these molecules results in a disorganisation of the membrane with modifications of its structure, flexibility, fluidity and permeability (Catalá, 2006; Repetto, Semprine & Boveris, 2012). The decrease in serum transaminases in groups fed with oils enriched with plant extracts could be explained by the low rancidity of these oil samples as previously observed with quality indices. Elevated transaminase levels in rats after consumption of fried oils had already been observed in a number of studies (Shastry et al., 2011; Badr El Said et al., 2015; Mboma et al., 2018). These results are also in agreement with Abdulaziz, Fasih, Saada, Khalid & Zarina, (2006), they showed that dietary supplementation with *Nigella sativa* limits the toxic effect of oxidised corn oil in rats by decreasing serum ALAT/ASAT concentrations. Similarly, Zeb & Khan (2019) found that administration of alpha-tocopherol in the diet of rats following oxidised olive oil-induced toxicity resulted in a decrease in serum ALAT concentration.

Proteins are biochemical macromolecules involved in the structural and biological functions of the organism and are also among the preferred targets of oxidation products. The decrease in protein levels observed is thought to be linked to the reduced digestibility of these molecules following their oxidation by free radicals. Indeed, free radicals react mainly at the sulfhydyl (SH) groups of amino acids such as cysteine, tyrosine and methionine contained in proteins, causing their oxidation (Therond, 2006). This phenomenon generally leads to the formation of protein aggregates, rendering them unusable by the body. On the other hand, the high concentrations recorded in the groups fed with enriched oil samples indicate the low oxidation of these oils, as previously observed in the chemical characterisation tests. These results are in agreement with those of Badr El Said et al. (2015) whose investigations resulted in the fact that the consumption of fried oils by dietary supplementation for three months leads to a decrease in serum protein concentration in rats.

Creatinine is a non-protein nitrogen compound synthesised from creatine in the muscles and is excreted primarily through the kidneys by glomerular filtration (Gowda et al., 2010), so an abnormally high level of creatinine in the blood would indicate kidney damage. The highest serum creatinine concentrations were recorded in the groups of animals that received the different samples of non-enriched oil. This increase could reflect inflammation of the kidneys in the glomeruli caused by the free radicals formed in these oils during frying. On the other hand, the low serum creatinine concentration observed in the groups of animals after consumption of palm olein enriched with plant extracts would be related to the low oxidative status of these oil samples. These results corroborate those of Amsalu *et al.* (2020) and Chew *et al.* (2019). Indeed, Amsalu, Wondimnew, Mateos, Fekadie & Bogale, (2020) showed that serum creatinine increased in rats following the consumption of fried palm oil, whereas the investigations of Chew et al. (2019) showed that the simultaneous consumption of citrus leaf extracts and thermoxidized palm olein resulted in a decrease in serum creatinine in rats.

4. Conclusion

It was found that ginger root extracts protect palm olein from oxidation during frying and that their effect is concentration dependent. With regard to total peroxide and oxidation values, ginger root at 1800 ppm was more effective than the BHT. The results of the *in vivo* test showed that the consumption of fried oils has an adverse effect on the biochemical parameters of rats while those supplemented with ginger root extract but not fried led to a clear improvement in these parameters, as the groups of animals consuming these oil samples showed lower or similar levels of serum transaminases, total protein, serum creatinine, HDL-cholesterol, atherogenicity index and haematological profiles compared to the neutral control group.

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