Food Technology Research Journal, Vol. 2, special issue 2, 37-54, 2023

FTRJ

Influence of psyllium, mustard, and flax seeds water extracts as fat replacers on cake quality and hyperlipidemic rats

*[1]Ayat, E. Rizk, [2]Zahrat EL-OLA, M. Mohamed, [3]Dalia, B. Othman & [1]Ebtehal, A. El-kholany

[1]Special Food and Nutrition Department, [2]Crops Technology Research Department, [3]Bread and Pasta Research Department,

Food Technology Research Institute, Agricultural Research Center ,Giza, Egypt

Original Article

Article information

Received 20/9/2023

Revised 13/10/2023

Accepted 9/11/2023

Published 27/11/2023

Available online 1/12/2023

Keywords:

Plantago ovata, Sinapis alba and Linum usitatissimum seeds, fat replacer, cake, sensory assessment and high fat diet.

ABSTRACT

In recent years, healthy and low- caloric bakery products have been widely demanded. The purpose of this study was to produce low-fat cake using water extracts of psyllium, mustard, and flax seeds. Fat was replaced in cake with water extracts of psyllium, mustard, and flax seeds at 50 and 100%. It was examined how the amount of fat replacers affected the cakes' physicochemical, sensory, and shelf life characteristics. The biological impact of these extracts on hyperlipidemic rats was also investigated. The produced cakes exhibited a higher moisture content when the amount of fat replacer was increased. In comparison to the control, the value of energy in the cake samples decreased significantly as the amount of fat replacer increased. Increasing the amount of fat replacement resulted in a decrease in the specific volume of cakes. The control and cake samples with 50% fat replacement showed higher values for all of the sensory parameters, with significant differences ($p \geq 0.05$) in taste, texture and overall acceptability compared with the other samples. Fat replacement in cake processing led to a prolonged shelf-life of the cakes compared with the control sample with respect to the microbiological assay. For biological evaluation, psyllium and flax seed extracts showed a significant ($p \geq 0.05$) decrease in total cholesterol and LDL- cholesterol. Psyllium, mustard, and flax seeds extracts caused a reduction in the activities of serum enzymatic transeferases (ALT and AST). So, the water extract of psyllium, mustard, and flax seeds produced an acceptable cake, thus allowing the production of potentially healthier food items. It may also be used to manage the accumulation of lipids, which affects body weight.

1. Introduction

Consuming excessive amounts of fat is said to be one of the main nutritional issues facing people today since it is associated with type-2 diabetes, obesity, hyperlipidemia cardiovascular disease, hypertension, colorectal and colon malignancies, LDL cholesterol, and other conditions. (Peng and Yao, 2017). A prolonged high-fat diet results in a disturbance of lipid metabolism and hyperlipidemia by reducing the activity of hepatic lipase, lipoprotein lipase, and other lipid metabolic enzymes (Li et al., 2011). For a healthy diet, the World Health Organization (WHO) advises that the consumption of saturated fat ought to be no more than 10% of total calories and that the consumption of fat should not exceed 30% of total calories (WHO, 2018). The need for reduced, low, or nonfat foods, as well as reduced-saturated and trans-fat foods, was brought about by these facts, government actions, and the growing impact of nutrition claims on consumers' food purchase intentions (Oostenbach et al., 2019). This was especially true for bakery products, which frequently use solid fats like butter and shortenings (Lim et al., 2017). Recent studies have focused on replacing saturated fats, including partly hydrogenated oil, because of their high trans-fat

*Corresponding Author

Email: layat72@yahoo.com

content, rather than lowering the overall amount of fat in diets. It has been demonstrated that consuming saturated fats increases levels of both low-density lipoprotein and total cholesterol. Additionally, increased concentrations of these substances have been connected to heart conditions (Patel et al., 2016**).** According to a number of previous studies, the qualities and traits of bread, cake, biscuits, and cookies can be affected by adding or removing fat using a range of hydrocolloids (Bavaro et al., 2021 and Rios et al., 2018). Since they are water soluble and hydrophilic by nature, hydrocolloids based on carbohydrates may find application as fat substitutes. These consist of inulin, fibers, and starches. gums, polydextrose, maltodextrins, fibers, and inulin (Colla et al., 2018 and Mousa, 2021). Their capacity to bind water molecules makes them valuable for replacing fat in processed foods (Saha and Bhattacharya, 2010). With fewer fat calories and better health outcomes due to increased dietary fiber, enhanced dietary system and prebiotic activity, and decreased calorie intake, carbohydrate-based hydrocolloid structure can perform some or all of the functions of fat (Li & Nie, 2016 and Kumar, 2021). Additionally, fat droplets can combine with polysaccharide hydrocolloids to form reduced-fat products with physico-chemical and sensory qualities equivalent to full-fat products (Murtaza et al., 2017). *Plantago ovata*, or psyllium, is a significant therapeutic herb. The husk and seed of *Plantago ovata* are used in common home folk medicine in the Pak-Indo subcontinent; according to Jabbar et al. (2020), the husk's alcoholic extract has higher antioxidant activity than the plant's seed, making it beneficial for diseases of the liver. *Plantago ovata* is abundant in bioactive substances, including fatty acids, amino acids, polyphenols, alkaloids, terpenoids, and vitamin C. Flavonoids and polyphenols exhibit significant antioxidant activity. These substances exhibit strong anti-inflammatory and antioxidant properties (Rafiee et al., 2022).

The seed of the flax plant, *Linum usitatissimum*, which is a genus of the Linaceae family, is mostly recognized for its nutritional value as a "functional food." In addition, it lowers the risk of atherosclerosis, cancer, cardiovascular disease, insulin-dependent diabetes mellitus, and hyperlipoproteinemia (Buckner et al., 2019). Several of flaxseed's physiologically active phytochemicals have been linked to health benefits. According to (Imran et al., 2015), it has a high concentration of plant lignans, alpha-linoleic acid (ALA), and dietary fiber. Mustard is valued for its pungent and spicy dried seeds, and it is a member of the Brassicaceae family. Black mustard, Brassica nigra (L.) W. D. J. Koch, brown mustard, Brassica juncea (L.) Czerniak, Brassica rugosa Hort., Sinapis juncea L., white mustard, and Brassica hirta Moench are a few of the well-known species of mustard (Rahman et al., 2018). In soils with a sandy type and little rainfall, mustards thrive. While tropical and subtropical areas are also suitable for its cultivation, temperate climates are the most common growing conditions for it. Grown extensively in Asia, North Africa, and Europe, it is regarded as one of the earliest cultivated crops (Divakaran and Babu, 2016). In the food industry, mustard plants are commonly used. The main purpose of white mustard is to flavor meals, but brown and black mustards are usually used for their fragrance. Herbal medicine experts also utilize mustard plants, like B. alba and B. juncea, to treat diabetes, arthritis, colds, coughs, sore throats, and muscle aches. Rahman and colleagues, 2018). Omega-3 fatty acids and glucosinolates (GSLs) are two of the bioactive substances found in mustard seeds (Melrose, 2019). GSLs consist of three compartments: variable aglycone side chain produced from an α-amino acid, thiohydroximate-O-sulfonate, and β-thioglucose (Ladak et al., 2021). Their well-known health benefits include lowering the risk of cancer and heart disease as well as inflammation (Gammone et al., 2019). Moreover, humans that consume glucose inoleates have favorable effects on their bodies and possess anticarcinogenic qualities, which include enhancing the bioactivity of mustard seed oil (Grygier, 2015).The present study aims to utilize the water extracts of Psyllum, mustard, and flax seeds in cake preparation and to evaluate the effect of replacing fat with these extracts on physical, ,texture and sensory characteristics as well

as the shelf life of produced cake. In addition, the biological effect of these extracts on hyperlipidemic rats was evaluated.

2. Materials and Methods

Materials

Psyllum *(Plantago ovata*), and mustard (*Sinapis alba* L) were obtained from a local market in Cairo, Egypt. Flaxseeds ((*Linum usitatissimum* L.) were obtained from the from Fiber Institute, Agricultural Research Center , Giza ,Egypt. Wheat flour of 72% extraction rate was obtained from South Cairo Mills Company Sugar, baking powder, corn oil, fresh eggs, Skimmed milk and vanillin powder,butter oil were purchased from the local market, Giza, Egypt. All chemicals were of the analytical reagent grade, Sigma Company. Analytical kits for LDL-cholesterol, HDL- cholesterol, total cholesterol, and triglycerides were obtained from Randox laboratories Ltd., Diamond Road, Crumlin, Co., Antrim, United Kingdom. BT294QY

Methods

Water Extract Preparation

With a few modifications, the water extract is prepared in the same method that these plants are traditionally administered, following (Bhatty's 1993) methodology. There, 5 g of the powdered psyllium, mustard, and flax seeds were extracted using 100 mL of distilled water and stirred for 20 minutes at 100°C, then passed through folded muslin.The process was repeated twice. The obtained water extract was mixed and stored in dark bottles immediately.

Cake Preparation

Cakes were prepared using the Bennion and Bamford (1997) formula. Table 1. contains the cake formula. Vegetable oil (in the reference formulation, RF) was replaced by the above extracts at 50 and 100% levels of substitution. The cake was prepared by creaming the sugar and oil together in a kitchen-aid mixer for three minutes on speed 5. After adding the eggs, they mixed for two minutes at the same speed. Following the addition of the flour, baking powder, and skim milk, the batter was mixed for four minutes at speed 2. filling each pan with cake batter and baking in an oven at 180ºC for 30 min. The cakes were removed from the pans after they were baked, allowed to cool, then wrapped in polyethylene bags and stored for a period of 21 days at room temperature. After being taken out of the pans, samples were collected for analysis within an hour and frequently once a week.

Chemical Analysis

Moisture, protein, fat, ash, and crude fiber contents of raw materials and bread samples were determined according to (AOAC 2019). Carbohydrate content was calculated on a dry weight basis by the difference:

[Carbohydrates=100 - (protein + fat + ash + crude fibers)].

Energy (Kcal) was calculated by the formula of James, 1995 as following equation:

Energy (Kcal) = Fat × 9 + Protein × 4 + Carbohydrates x 4

Total dietary fiber (TDF), soluble dietary fiber (SDF), and insoluble dietary fiber (IDF) were determined according to the enzymatic gravimetric method (Prosky et al., 1988). According to (Singleton and Rossi 1965), the Folin-Ciocalteu method was used to determine the total phenol content of aqueous extracts. To prepare the standard curve, gallic acid was chosen as the standard. According to (Brand-Williams et al., 1995), the antioxidant activity of water extracts was determined by their capacity to scavenge radicals when they reacted with a stable DPPH-free radical using extract of methanol. In summary, 3.90 ml of the DPPH solution (2.40mg of DPPH in 100 ml of methanol)was added to 0.10 ml of sample extract. The mixture was shaken vigorously using a tube shaker for a few seconds and allowed to stand at room temperature for half an hour while kept out of the light. At 515 nm, the absorbance was then measured. Using the following equation, the radical scavenging percentage (DPPH) was calculated

$$Radical\ scavenging\ (\%) = \left[\frac{A0 - A1}{A0}\right] x\ 100$$

A_0 = absorbance of the control reaction (containing all reagents except the test compounds)

A1 = absorbance in the presence of the tested extracts after 30 min

Table 1. Cakes formula prepared with fat replacer extracts at different levels of replacement

Samples Ingredients (g)	Control	P 50%	P 100%	M 50%	M 100%	F 50%	F 100%
Wheat flour 72% ext.	100	100	100	100	100	100	100
Sugar	85	85	85	85	85	85	85
Corn oil	65	32.5	-	32.5	-	32.5	-
Psyllium seed water extract as a fat replacer ratio	-	32.5	65	-	-	-	-
Mustard seed water extract as a fat replacer ratio	-	-	-	32.5	65	-	-
Flax seed water extract as a fat replacer ratio	-	-	-	-	-	32.5	65
Whole egg	85	85	85	85	85	85	85
Skimmed milk	3.0	3.0	3.0	3.0	3.0	3.0	3.0
Vanillin	0.6	0.6	0.6	0.6	0.6	0.6	0.6
Baking powder	3.8	3.8	3.8	3.8	3.8	3.8	3.8

P: Psyllium seed water extract; F: Flax seed water extrac ; M: Mustard seed water extract ; 50 &100: percentages of replacement. Water was added as need.

Viscosity of psyllium, mustard, and flax seeds water extracts

The viscosity of different water extracts was measured using the Brookfield Engineering Labs DV-III Ultra Rheometer in accordance with the (Brookfield Manual, 1998). The sample was placed in a small adaptor, and the appropriate temperature was kept constant using a water bath. The rpm range for the viscometer was 10 to 60. Viscosity was obtained directly from the instrument, and at room temperature (25 ^{o}C ± 1 ^{o}C), the SC4-21 spindle was selected for the measurement. The measurement of each sample was averaged from 3replicates.

Physical Properties of Cakes

The rapeseed displacement method was used to calculate the cake volume (in cm^3), according (AACC 2002). By dividing the volume by the weight, the specific volume (cm^3g^{-1}) of the cake was calculated. Weight divided by volume got the density (g cm^{-3}) value. A micrometer was used to measure the height of the cake to the nearest millimeter. The Physical measurement of each sample was averaged from 3 replicates.

Texture Profile Analysis (TPA) of Cake Samples

A texture profile analyzer (TPA) was used to measure the cake's firmness (N), cohesiveness and springiness were calculated from TPA graphic in accordance with (Baixaulieta et al., 2008). A universal testing machine was used to determine the parameters of the cake texture (Brook Field Engineering Lab, Inc., Middleboro, MA 02346-1031, USA). A TPA was utilized with a cylindrical probe with a 25 mm diameter and a 2 mm/s speed. The texture measurement of each sample was averaged from 3 replicates.

Color of Cakes

The technique that (McGurie 1992) described was used for evaluating the color of cake samples. Using a hand-held tristimulus reflectance Colorimeter (model CR-400, Konica Minolta, Japan), the color of the cake (crust and crumb) was measured. *L* (lightness, with *L* = 100 for lightness and *L* = 0 for darkness), *a* [chromaticity on green (−) to red (+)], and *b* [chromaticity on blue (−) to yellow (+)] were provided by the apparatus. The color measurement of each sample was averaged from 3 replicates.

Sensory Evaluation of Cake Samples

After backing, cake samples were allowed to cool (25°C±2) for four hours before being tested for

four hours before being tested for organoleptic characteristics. Ten well trained panelists - five men and five women-from Food Technology Research Institute personnel were given a slice of each cake sample on white, odorless, disposable plates. Taste, odor, texture, appearance, and total score on a 9-hedonic scale from one (dislike extremely) to nine (like extremely) were all assigned to the samples. The evaluation was completed in accordance with (Bennion and Bamford 1997).

Microbiological analyses

The microbiological quality of stored cake for 3 weeks at an ambient temperature was evaluated by determining total fungal count (1g sample) using malt yeast agar media to be as a good tool to estimate the shelf life according to (Mislivec et al., 1992) and aerobic plate count using total count media (Swanson et al., 1992).

Biological evaluation

The experimental animals design

In The Experimental Animal House of Food Technology Research Institute, Agriculture Research Center, Giza, Egypt, thirty adult male albino rats weighing between 180±20 g. The animals were housed in plastic cages and Fed on a basal diet of (g/100g) were consisted of 10 g protein as casein,1g vitamin,4 g mineral.10 corn oil .5g cellulose and the remaining of 100 g of the diet was completed by starch .that was based on AIN-formulation with some modifications (Reeves et al., 1993) and provided water and libitum for one week as an adaptation period. The animal room temperature was maintained at 22ºC±2 with timed lighting of 12 hours and a relative air humidity of 40% to 60%.after the adaptation period the animals were divided into two groups :first group 5 rats fed on basal diet along of experimented and the second group contain 25 rats were fed a high-fat diet according to (Woods et al., 2003) with some modificatin that 25g fat (butter oil was added to 100 g basal diet and rats were fed this diet until the end of the experiment. After three week five rats were randomly selected to get lipid profile analyses examinations to confirm the rats had become hyperlipidemic. then ,the rats of the second group (20 rats) were divided to 4 groups each group contain 5 rats. as treated hyperlipidemic groups (G3,G4,G5 groups to administrate water extracts by stomach tube for other 7 weeks according to the following scheme.

G1: Negative control contend fed on basal diet all experimental

G2: positive control group (Hyperlipidemic rats received high fat diet

G3:Hyperlipidemic rats received high fat diet and orally administrated (2ml/rat/day)of water extracts flaxseed

G4 :Hyperlipidemic rats received high fat diet and orally administrated (2ml/rat/day)of water extracts Mustard

G5 : Hyperlipidemic rats received high fat diet and orally administrated (2ml/rat/day)of water extracts psyllium

The blood samples were collected from eye plexuses after slight anesthesia in rats and immediately into glass tubes centrifuged at 30000 rpm for 20 min at 4 C to obtain the serum, which was kept frozen at -20 C until analysis as described by Shermer (1967).

Body weight ,final weight and BWG according to Essam and Maha 2012 the following equation

Body weight gain =final weight -initial weight / initial weight ×100

Biochemical Analysis

According to (Stein 1986), serum glucose, total cholesterol, total triglycerides, and high-density lipoproteins (HDL) were measured. Calculations were used to determine low-density lipoprotein (LDL) as follows: According to Wallach (1992), very low-density lipoproteins (VLDL) were computed using the following equation: LDL=TC-(HDL+VLDL) as reported by (Varley et al., 1980). Aspartate aminotransferase (AST) was measured using the Reitman and Frankel (1957) method, and VLDL=(TG/5). The serum's levels of creatinine and urea were measured in accordance with Patton and Crouch (1977).

Histopathological Examination

Tissue specimens were collected from the liver and kidney of rats, fixed in 10% neutral buffered formalin according to (Bancroft and Gamble 2008), dehydrated in different grades of alcohol, cleared in xylene, embedded in paraffin, sectioned with a microtome at 5µ thickness, and finally stained with hematoxylin and eosin (H&E) and masson`strichrome (MTC) according to Drury and (Wallington 1980).

Statistical Analysis

The data from this study were statistically analyzed using the Costat statistical software for means and standard deviations (Steel *et al*., 1997). The data were subjected to one-way analysis of variance (ANOVA) using one way, followed by Duncan's multiple range tests (at $p<0.05$) to assess differences between sample means.

3. Results and Discussions

Table 2. displays the chemical composition of Psyllium, Mustard, and Flax whole seed powder. The results indicate that psyllium seeds had a lower fat content (6.76%) than flax seed and mustard seed flour (30.24 and 31.33%, respectively). Mustard seed powder has the highest protein content (32.46%). The highest total dietary fiber content was found in psyllium seeds (26.49%), followed by mustard flour (5.88%) and flaxseeds (24.5%). (Romero-Baranzini et al., 2006) stated that when compared to cereals and legumes, the nutritional quality of psyllium whole grains is favorable.

Table 2. Chemical composition of psyllium, mustard and flax whole seeds powder*

Composition (g/ 100)	Psyllium seeds	Flax seeds	Mustard seeds
Moisture	$7.1^{a}\pm 0.06$	$7.30^{a}\pm 0.14$	$5.02^{b}\pm 0.05$
Ash	$4.43^{a}\pm 0.04$	$3.79^{b}\pm 0.04$	$3.95^{b}\pm 0.18$
Protein	$15.53^{c}\pm 0.11$	$21.30^{b}\pm 0.14$	$32.46^{a}\pm 0.38$
Fat	$6.76^{b}\pm 0.06$	$30.24^{a}\pm 0.15$	$31.33^{a}\pm 0.04$
Dietary fiber	$26.49^{a}\pm 0.1$	$24.50^{b}\pm 0.42$	$5.88^{c}\pm 0.056$
Soluble	$8.57^{a}\pm 0.17$	$5.25^{b}\pm 0.21$	$0.55^{c}\pm 0.021$
Insoluble	$17.92^{b}\pm 0.28$	$19.25^{a}\pm 0.21$	$5.33^{c}\pm 0.07$

*Chemical composition was calculated based on the dry weight basis (% on dwb)
Data are presented as means ± SDM (n=3) & Means within a column with different letters are significantly different at $P \leq 0.05$.

Phenol content, antioxidant activity, and viscosity of psyllium, mustard, and flaxseed extracts

Table 3. demonstrates that water extract of mustard seeds had higher phenols content and antioxidant activity % than water extract of psyllium and flax seed. (Aziz et al., 2020) reported that The antioxidant properties of mustard seeds were probably attributed to these rich phenolic profiles. Therefore, scientific evidence for the medical application of these plants may contribute to bettering the health of those who use them. while, highest viscosity was observed in water extract of psyllium seeds 126Cp. (Niu et al., 2012) repeated that depending on the concentration, this extremely hydrophilic substance will swell and form a gel or a highly viscous slurry by absorbing more than its weight in

Table 3. Total phenols content (mg/100g), antioxidant activity% and viscosity (CP) of psyllium, Mustard and flaxseed water extracts

Water extract of seeds	Total phenols, (mg/100g)	Antioxidant activity %	Viscosity (CP)
Psyllium seeds	$102^{c}\pm 1.41$	$18^{c}\pm 1.3$	$126^{a}\pm 0.00$
Mustard seeds	$438^{a}\pm 2.82$	$40^{a}\pm 1.40$	$1.28^{c}\pm 0.05$
Flaxseeds	$310.5^{b}\pm 0.76$	$32^{b}\pm 1.06$	$50^{b}\pm 1.0$

Data are presented as means ± SDM (n=3) & Means within a column with different letters are significantly different at $P \leq 0.05$.

Table 4. shows the approximate chemical composition and caloric value of a cake prepared with water extracts of, mustard, psyllium and flaxseed as a replacement for fat. The cakes' moisture content increased significantly as the replacement level got higher. Table 4 shows that protein content increased with increasing levels of fat replacement extracts compared with control; however, fat content decreased with increasing levels of water extract relative to control. A higher content of fat and energy was found in the control compared with the substituted cake samples. It is clear that all fat substitutes increased ash levels compared to control samples. Carbohydrates significantly ($P \leq 0.05$) increased with elevating the fat level replacement compared to the control samples. Concerning the energy value, a gradual, significant decrease was observed as the substitution level increased. It was reported byEl-Sayed *et al.* (2014)found that when the replacement level got higher, the cakes' moisture content increased significantly.

Physical Characteristics of Cakes

Volume is a very important quality for cakes, which strongly influences consumer preference and is directly related to the type and amount of shortening used. (Bennion and Bamford 1997) stated that fat provides several advantages to cakes, such as higher volume and flavor. (Borneo et al., 2010) explained this by the higher air incorporation during batter preparation and inhibition of gas bubble coalescence, leading to a finer and softer crumb structure.

Table 4. Chemical composition (g/100g) of cakes containing different levels of fat replacer extracts

Sample	Moisture g/100g	Ash g/100g	Protein g/100g	Crude fat g/100g	Crude fiber g/100g	Carbohydrate g/100g	Energy Kcal/100g
C	$1.82^{e}\pm 0.03$	$1.78^{c}\pm0.06$	$9.19^{b}\pm 0.25$	$15.76^{a}\pm 0.16$	$0.36^{cd}\pm 0$	$72.91^{e}\pm0.03$	$470.24^{a}\pm 0.59$
F50	$2.78^{c}\pm 0.04$	$1.84^{bc}\pm0.02$	$10.31^{a}\pm 0.03$	$8.86^{b}\pm 0.06$	$0.33^{d}\pm 0.04$	$75.66^{d}\pm .047$	$435.62^{b}\pm 0.2$
F100	$3.22^{b}\pm 0.01$	$1.94^{ab}\pm0.05$	$10.65^{a}\pm 0.03$	$2.88^{d}\pm 0.04$	$0.4^{bc}\pm 0$	$84.13^{b}\pm 0.06$	$405.06^{e}\pm0.00$
M50	$2.88^{c}\pm0.02$	1.83^{b}c±0.01	$10.17^{a}\pm0.16$	$8.45^{c}\pm0.13$	$0.35^{cd}\pm0.01$	$79.21^{c}\pm0.28$	$433.57^{c}\pm0.63$
M100	$3.67^{a}\pm 0.02$	$2.01^{a}\pm 0.05$	$10.55^{a}\pm0.16$	$2.46^{e}\pm 0.14$	$0.4^{bc}\pm0$	$84.57^{a}\pm 0.02$	$402.69^{f}\pm 0.50$
P50	$2.635^{d}\pm0.05$	$1.87^{bc}\pm 0.01$	$10.08^{a}\pm0.17$	$8.21^{c}\pm 0.13$	$0.44^{b}\pm 0.02$	$79.41^{c}\pm 0.02$	$431.85^{d}\pm 0.57$
P100	$3.21^{b}\pm 0.09$	$2.04^{a}\pm 0.02$	$10.42^{a}\pm0.16$	$2.29^{e}\pm 0.16$	$0.53^{a}\pm 0$	$84.72^{a}\pm 0.02$	$401.16^{g}\pm 0.91$

C: control cake, F: Flax seed water extrac ; M: Mustard seed water extract .; P: Psyllium seed water extract ; 50 &100: percentages of substitution in cakeData are presented as means ± SDM (n=3) & Means within a column with different letters are significantly different at $P \leq 0.05$.

The effect of replacing fat with seed water extracts on cake volume, specific volume, and density is shown in Table 5. Cakes prepared with 50 and 100% substitution had a significantly lower volume compared with the control cake (131.42 cm^3). No significant differences was observed between cakes with 100% replacement for flax seed and psyllium extracts. The specific volume of cakes decreased as replacement levels increased (Table 5), showing a reduction of 16% when the entire fat amount was substituted. The differences in specific volume were significant for all of the different extract substitutes. These results were also observed by (Zahn et al., 2010), (Felixto et al., 2015), and (Hussien 2016). (Rodríguez-García et al., 2012) explained that the decrease in specific volume could be related to an increase in bubble size in batters with low oil content, as a bigger size of bubbles provides less stability and releases the bubbles into the atmosphere. (Zahn et al., 2010) reported that the specific volume does not depend only on the air incorporated into the batter but also on the air retained after baking, which could explain why the specific volumes decreased.

Table 5. Physical Characteristics of cakes prepared by replacing fat with different levels of seed water extracts

Treatment	Volume (cm^3)	Specific Volume (cm^3/g)	Weight (g)	Density (g/cm^3)
C	131.42 ^{a} ±0.36	2.47 ^{a} ±0.03	53.28 ^{c} ±0.19	0.41 ^{d} ±0.01
F50	124.74 ^{d} ±0.20	2.34 ^{c} ±0.01	53.42^{bc} ±0.03	0.43 ^{b} ±0.03
F100	121.47 ^{f} ±0.45	2.27 ^{e} ±0.02	53.63^{ab} ±0.16	0.44 ^{a} ±0.01
M50	130.44 ^{b} ±0.35	2.45 ^{a} ±0.01	53.19 ^{c} ±0.09	0.41 ^{cd} ±0.03
M100	128.28 ^{c} ±0.31	2.40 ^{b} ±0.03	53.35 ^{c} ±0.22	0.41 ^{c} ±0.01
P50	123.79 ^{e} ±0.16	2.31 ^{d} ±0.01	53.66^{ab} ±0.05	0.43 ^{b} ±0.02
P100	121.62 ^{f} ±0.33	2.26 ^{e} ±0.04	53.80 ^{a} ±0.10	0.44 ^{a} ±0.02

C:control cake, F: Flax seed water extrac ; M: Mustard seed water extract .; P: Psyllium seed water extract ; 50 &100: percentages of substitution in cake.
Data are presented as means ± SDM (n=3) & Means within a column with different letters are significantly different at $P \leq 0.05$. ≤ 0.05.

Texture Profile of cakes

Texture profile analysis (TPA) is a very useful technique for investigating food products where tenderness and elasticity (resilience) are the main textural properties of a cake and related to quality (Vassiliki and Vassiliki, 2013). In the present study, the TPA parameters of cakes containing different levels of fat replacement extracts were measured by the texture analyzer shown in Table 6. Firmness or hardness which is defined as the maximum force of the first compression of the product at the point of 50% compression (1mm/s speed test). The firmness values of cakes with different levels and types offat replacement, showed a dramatic significant (P≤0.05) decrease with the increase in replacement levels being more pronounced in cakes replaced with P extract. It could be observed thatcakes made with psyllium extract showed lower hardness values than those made with other fat replacements. These results agree with work by Belorio*et al*, 2020. Rodríguez-García*et al.*, 2012)stated that the lower the specific cake volume, the greater the hardness. While in the study by (Belorio*et al*, 2020) they reported the greater amount of water added to the formulations could compensate the effect of the volume on the hardness. There was non-significant increase in cake cohesiveness with fat replacement. Zahn *et al.,*2010; Rodríguez-García*et al.,*2012did not find significant differences of cohesiveness when using various levels of fat replacements. Springiness of cakes with both flax seed and mustered extracts did not differ significantly from the control. Both levels of psyllium (50 and 100%), exhibited a lower value of springiness than the other-substitutes and the controls. Some studies as (Zahn *et al.,* 2010; Rodríguez-Garcíaet al.,2012 and Hussien, 2016 showed increasing springiness values with higher fat substitutions while others as Belorioet al, 2020) reported decreasing values when the fat was replaced. These results indicate that texture parameters differ depending on the fat substitute used.

Color Parameters

Color parameters are shown in Table 7. The crust of the cakes made with the Mustard fat replacer showed increasing L values, which means they were lighter than the others, but significant differences were only found in substitution levels above 50%. Samples, also presented significant differences in a and b values, which decreased and increased, respectively. High darkness of the crust was explained by. (Purlis 2010) who attributed the crust's extreme darkness to the Maillard reactions and caramelization that occur during baking. However, (Belorio et al., 2020) claimed that since the amounts of protein and sugar in each sample were the same, this cannot account for the variations in crust color. But (Hidalgo and Zamora 2005) mentioned that as a derivative molecule resulting from their oxidation alters the product color while present, the effect

of lipids on this type of reaction could be responsible for the color variations observed. No significant differences in *L* and *b* values were observed in the crumb color of those cakes prepared with fat replacement compared with control cakes. High a values were only observed in cakes with total fat substitution, whereas some studies on fat replacement found large differences. (Felisberto et al., 2015), when using chia mucilage gel, obtained darker crumbs, while Rodríguez-(García et al., 2012) obtained lighter cakes when incorporating inulin in the cakes. (Belorio et al., 2020) explained that the temperature inside the cakes does not exceed 100 °C, so Maillard reactions and caramelization cannot occur. In this way, the crumb color depends mainly on the color of the ingredients. Therefore, the differences found in these studies depend on the oil employed and the oil replacer.

(Belorio et al., 2020) explained that the temperature inside the cakes does not exceed 100 °C, so Maillard reactions and caramelization cannot occur. In this way, the crumb color depends mainly on the color of the ingredients. Therefore, the differences found in these studies depend on the oil employed and the oil replacer.

Table 6. Texture Profile Analysis of Cake prepared with and without Fat Replacer

Treatments	Firmness (N)	Cohesiveness	Springiness (N)
C	2.09 ^{a}±0.03	0.62 ^{a}±0.03	0.64 ^{a}±0.03
F50	2.03 ^{b}±0.01	0.65 ^{a}±0.04	0.64 ^{a}±0.01
F100	1.91 ^{c}±0.02	0.64 ^{a}±0.03	0.63 ^{a}±0.02
M50	2.01 ^{b}±0.02	0.64 ^{a}±0.00	0.64 ^{a}±0.03
M100	1.93 ^{c}±0.01	0.63 ^{a}±0.02	0.62 ^{a}±0.02
P50	1.84 ^{d}±0.03	0.62 ^{a}±0.02	0.56 ^{b}±0.05
P100	1.78 ^{e}±0.01	0.65 ^{a}±0.02	0.55 ^{b}±0.03

C: control cake, F: Flax seed water extrac ; M: Mustard seed water extract .; P: Psyllium seed water extract ; 50 &100: percentages of substitution in cake

Table 7. Color values of Cake preparedwith and without Fat Replacer

Treatments	Crust			Crumb		
	L	a	b	L	a	b
C	53.11 ^{b}±0.06	14.96 ^{a}±0.13	25.73 ^{e}±0.03	71.61 ^{a}±0.04	2.14 ^{e}±0.07	24.95 ^{a}±0.04
F50	53.35 ^{b}±0.05	14.29 ^{c}±0.04	26.08 ^{d}±0.08	71.51 ^{a}±0.03	2.60±0.07^{d}	25.08 ^{a}±0.07
F100	54.34 ^{a}±0.08	13.96 ^{d}±0.08	26.85 ^{c}±0.05	71.41 ^{a}±0.09	2.81 ^{a}±0.03	25.10 ^{a}±0.03
M50	53.48 ^{b}±0.02	14.06 ^{b}±0.06	28.38 ^{b}±0.02	71.37 ^{a}±0.03	2.56 ^{d}±0.07	25.14 ^{a}±0.06
M100	55.46 ^{a}±0.08	12.05 ^{f}±0.05	29.65 ^{a}±0.04	71.17 ^{a}±0.06	2.67 ^{c}±0.07	25.12 ^{a}±0.02
P50	53.36 ^{b}±0.08	14.09 ^{b}±0.04	26.01 ^{d}±0.09	71.41 ^{a}±0.08	2.70 ^{b}±0.06	25.09 ^{a}±0.05
P100	54.11 ^{a}±0.04	13.13 ^{e}±0.12	26.75 ^{c}±0.07	71.07 ^{a}±0.02	2.74^{ab}±0.06	25.11 ^{a}±0.03

C: control cake, F: Flax seed water extrac ; M: Mustard seed water extract .; P: Psyllium seed water extract ; 50 &100: percentages of substitution in cake

Data are presented as means ± SDM (n=3) & Means within a column with different letters are significantly different at $P \leq 0.05$.

Sensory evaluation

The results of sensory evaluation are shown in Table 6. In general, the control and cakes with 50% of the oil replaced presented higher values for all the sensory parameters, with significant differences in taste, texture, and overall acceptability compared with the rest of the samples.

However, all the cakes obtained a high evaluation (above 7) with differences that did not exceed 1 point between the control cakes and those with a fat replacer. The cakes with fat replacer showed significant differences in all sensory parameters compared with the control cakes, which reduced their global acceptability. It must be considered that these cakes had a lower specific volume and a lighter crust color, which could have negatively influenced the evaluations of the consumers. The texture of the cakes substituted with oil was given poor evaluations by the consumers. (Rodríguez-García et al. 2012) observed lower levels of acceptability of cakes with 100% fat replacement. They explained this by the crumbling and irregular crumb cell structure. (Belorio et al., 2020) stated that oil or fat substitution by fibers or other ingredients reduced consumers' acceptability of the cakes, but in the case of psyllium, this reduction was very small, and the products received a good global evaluation.

Table 8. Sensory evaluation of cake prepared with and without fat Replacer

Treatments	Appearance (9)	Texture (9)	Odor (9)	Taste (9)	Overall acceptability (9)
C	8.58 ^{a}±0.28	8.52 ^{a}±0.19	8.64 ^{a}±0.14	8.43 ^{a}±0.18	8.64 ^{a}±0.24
F50	8.43 ^{b}±0.19	8.54 ^{a}±0.13	8.00 ^{d}±0.11	8.14 ^{c}±0.12	8.43 ^{b}±0.14
M50	8.15 ^{e}±0.11	8.29 ^{d}±0.11	8.07 ^{cd}±0.15	8.07 ^{d}±0.17	8.29 ^{c}±0.18
P50	8.36 ^{c}±0.13	8.57 ^{a}±0.15	8.29 ^{b}±0.19	8.21 ^{b}±0.11	8.50 ^{b}±0.13
F100	8.07 ^{e}±0.17	8.33 ^{c}±0.12	7.79 ^{f}±0.12	7.86 ^{f}±0.14	7.93 ^{e}±0.11
M100	7.90 ^{f}±0.12	8.07 ^{e}±0.17	7.93 ^{e}±0.11	7.79 ^{f}±0.15	7.86 ^{f}±0.12
P100	8.21 ^{d}±0.15	8.47 ^{b}±0.14	8.10 ^{c}±0.12	7.93 ^{e}±0.13	8.14 ^{d}±0.12

C: control cake, F: Flax seed water extrac ; M: Mustard seed water extract .; P: Psyllium seed water extract ; 50 &100: percentages of substitution in cake

Data are presented as means ± SDM (n = 10, a 9-point hedonic scale: 1 (9= like extremely, 8= like very much, 7= like moderately, 6= like slightly, 5= neither like nor dislike, 4= dislike slightly, 3= dislike moderately, 2= dislike very much, 1= dislike extremely) & Means within a column with different letters are significantly different at $P \leq 0.05$.

The Microbial Quality of Cake

Certain baked goods may spoil due to post-baking contamination if they are inadequately stored for a prolonged period of time. Molds are one of the most common forms of spoilage for baked goods. Foods are considered spoilt if there are more than 10^7 or 10^5 CFU of mold or bacteria per ml or g, respectively (Mossel et al., 1995)

Tables 9. display the total bacterial and fungal counts (cfu/g) of produced cake samples during storage at room temperature for 3 weeks to be a suitable monitor for the shelf life of the product. The results showed an increase in bacterial and fungal counts during storage time. All the cake samples were not showed either bacterial or fungal count at zero time. Furthermore, results indicated that cake prepared by fat replacers (Psyllium, mustard, and flax seeds extracts) had the lowest value of bacterial and fungal counts for all storage period than control cake. Bacterial and fungal counts of cakes prepared with fat replacer was decreased with increasing replacment levels. Moreover, 100% of mustard replacment level showed the lowest bacterial and fungal counts for all storage period .The results are in agreement with (Ibrahim et al., 2016), who found that the control sample of cake contained log 5.87 and log 7.32 cfu/g for bacterial and fungal counts after three weeks of storage time. The replacement with 50% and 100% water extracts of mustard seed decreased the bacterial and fungal counts of the produced cake samples compared with the control sample since (Min-suk et al., 2003) reported that mustard seed flour contained antibacterial activity against foodborne pathogenic bacteria**.** Furthermore, Flax seed meal had antibacterial efficacy against foodborne pathogens, particularly against *S. aureus* and *E. coli* 0157.H7, as reported by (Son and Song 2017).

(Sharif et al., 2011) investigated the effect of a hydroalcoholic extract of plantago psyllium seeds against pathogenic bacteria. The results revealed that the extract inhibited the growth of *S. aureus* and *S. epidemids.*

Table 9. Total bacterial (TBC) and fungal counts (TFC) as cfu/g of produced cake samples during storage time of three weeks at room temperature

Treatments	Total count				Yeast, Mould			
	Zero time	1week	2weeks	3 weeks	Zero time	1 week	2 weeks	3 weeks
C	ND	7×10^{1}	5.2×10^{3}	7×10^{5}	ND	ND	6×10^{1}	8×10^{2}
M50	ND	ND	4×10^{2}	4×10^{4}	ND	ND	1×10^{1}	1.4×10^{2}
F50	ND	2×10^{1}	8×10^{2}	6×10^{4}	ND	ND	2.2×10^{1}	1.7×10^{2}
P50	ND	5.5×10^{1}	11×10^{2}	8×10^{4}	ND	ND	3.5×10^{1}	2.8×10^{2}
M100	ND	ND	5×10^{1}	3×10^{2}	ND	ND	ND	4.7×10^{1}
F100	ND	ND	8×10^{1}	5×10^{2}	ND	ND	1×10^{1}	5.3×10^{1}
P100	ND	$2.5\text{x}10^{1}$	11×10^{1}	7×10^{2}	ND	ND	2×10^{1}	1.1×10^{2}

C: control cake, F: Flax seed water extrac ; M: Mustard seed water extract .; P: Psyllium seed water extract ; 50 &100: percentages of substitution in cake

Data are presented as means ± SDM (n=3) & Means within a column with different letters are significantly different at $P \leq 0.05$. ND: not detected

Biological Evaluation

Table 10. shows that there were significant variations in wight gain among all treatment groups.On the other hand, oral administration of mustard, psyllium, and flaxseed extracts significantly decreased weight gain when compared to the G+ group. This may be due to the presence of soluble gel-forming mucilage from the Psyllium , flax and mustard. The development of viscous materials can help with satiety and weight loss. This result agrees with that of (Al-Askalany 2012). Table 10. displays organ kidney, liver, and heart weight were significantly increased at (P<0.05) inn the positive control group when compared to the treated group.

Table 10. Body weight (%) gain and organs (%) of rats orally administrated psyllium, mustard and flaxseed extracts .

Groups	Body weight gain %	Liver %	Kindy %	Heart %
G1	$15.3^{c}\pm0.12$	2.0	0.5	0.31
G2	$46.1^{a}\pm0.52$	2.8	1.0	0.36
G3	$20.7^{b}\pm0.55$	2.3	0.6	0.38
G4	$23.0^{b}\pm0.61$	2.4	0.6	0.42
G5	$22.3^{b}\pm0.23$	2.1	0.6	0.38

G1: Negative control .,G2: positive control group (Hyperlipidemic rats).,G3:Hyperlipidemic rats that orally administrated water extracts flaxseed.G4 :Hyperlipidemic rats that orally administrated water extracts Mustard, G5 : Hyperlipidemic rats that orally administrated water extracts psyllium Data are presented as means ± SDM (n=5) & Means within a column with different letters are significantly different at $P \leq 0.05$.

Table 10. shows that shows that Psyllium, Mustard, and Flaxseed extracts were able to protect organs against the negative physiologic effect of high lipid. On the contrary, there were no significant differences found in the liver, kidney, and heart relative weights of rats except in group G+. that a high-fat diet causes an elevation in body weight and reduces lipid metabolism, as clearly seen by the marked elevation of liver enzymes and lipid levels. On the other hand, Table 11. displays a markedly significant increase in Glucose in total cholesterol and While decrease in LDL and HDL in groups compared with those of the control group G+. However, there has been an increase in HDL-C concentration in the treatment group. Psyllium extract and flax extract have shown a significant decrease in total cholesterol and LDL cholesterol. due to being rich in monounsaturated fatty acids. High-fat diets show a remarkable increase in blood glucose (Haley et al., 2013).Treatment with an extract of 200 mg/kg decreased the blood glucose level (Vessby, 2000). psyllium extract soluble fiber attaches, forming a complex that prevents reabsorbing bile from the small intestine,thus increasing the production and secretion of the bile acids by drawing cholesterol to be used as a substitution for the missed acids, thereby lowering blood cholesterol levels. and increased serum triglyceride concentration (Klop et al., 2013).

Table 11. Lipid profile and glucose level of rats orally administrated on psyllium, flax and mustard seed water extracts

Groups	TC (mg/dl)	HDL (mg/dl)	LDL (mg/dl)	VLDL (mg/dl)	TG (mg/dl)	Glucose (mg/dl)
G1	97.5^{e}±0.12	85.5^{a}±2.90	20.4^{e}±2.80	32.4^{c}±1.70	162.0^{d}±2.90	78.0^{d}±2.6
G2	210^{a}±2.55	58.5^{b}±2.13	102^{a}±2.45	48.6^{a}±2.50	243.0^{a}±2.84	100.0^{a}±1.55
G3	105.0^{c}±0.12	42.0^{c}±1.00	28.0^{d}±1.55	35.0^{b}±0.22	175.0^{c}±1.32	75.0^{e}±1.76
G4	103.5^{d}±0.02	34.5^{e}±1.95	33.0^{b}±0.50	36.0^{b}±1.12	180.0^{b}±2.50	80.0^{c}±1.78
G5	106.5^{b}±0.05	38.5^{d}±2.15	32.8^{c}±1.53	35.2^{b}±1.00	176.0^{c}±0.90	83.0^{b}±1.92

G1: Negative control .,G2: positive control group (Hyperlipidemic rats).,G3:Hyperlipidemic rats that orally administrated water extracts flaxseed.G4 :Hyperlipidemic rats that orally administrated water extracts Mustard, G5 : Hyperlipidemic rats that orally administrated water extracts psyllium

It is clear from Table 12. that there was a significant increase in the level of serum ALT, AST, and ALP in group control G2 compared with the normal group. On the other hand, psyllium, fxed seed, and mustard extract caused a reduction in the activities of serum enzymes AST and ALT, which decreased significantly in all treated groups compared with the G+ control group. These results may be due to bioactive compounds in water extract, such as phenolic compounds, and may improve kidney functions (Takasaki 2005).

Table 12. Serum enzyme of rat livers orally administrated flax, mustard and psyllium seed water extract

Groups	ALT(U/L)	AST(U/L)	ALP(U/L)
G1	17.5^{c}±2.60	26.69^{d}±0.05	128^{e}±3.90
G2	45.5^{a}±2.55	38.97^{a}±2.90	232^{a}±3.78
G3	27.0^{b}±1.00	27.1^{c}±0.23	157^{d}±2.10
G4	28.0^{b}±1.02	28.28^{b}±0.04	160^{c}±0.32
G5	26.0^{b}±1.02	24.1^{e}±0.12	162^{b}±1.00

G1: Negative control .,G2: positive control group (Hyperlipidemic rats).,G3:Hyperlipidemic rats that orally administrated water extracts flaxseed.G4 :Hyperlipidemic rats that orally administrated water extracts Mustard, G5 : Hyperlipidemic rats that orally administrated water extracts psyllium

Data are presented as means ± SDM (n=5) & Means within a column with different letters are significantly different at $P \leq 0.05$.

Table 13. shows that an increase in urea and uric acid in group G+ while a decrease in the treatment group revealed that the rats in the positive control had a significant increase in kidney functions compared to rats in the negative control. However, the rats administrate on HFD with Mustard and Psyllum (extract) resulted in a significant decrease in kidney functions compared to the positive control. due to the higher mucilage content of psyllium seed. (Kumar and Shsrma, 2013)

Table 13. Serum enzyme of rat kidney orally administrated flax, mustard and psyllium seed water extract

Treatment	Urea, (mg/dl)	Uric acid, (mg/dl)	Creatinine, (mg/dl)
G1	40.0^{e}±1.90	3.1^{c}±0.01	0.5^{b}±0.01
G2	59.0^{a}±2.45	3.4^{a}±0.12	1.5^{a}±0.02
G3	43.0^{c}±1.90	2.9^{c}±0.03	0.45^{c}±0.03
G4	46.0^{b}±2.00	3.2^{b}±0.02	0.42^{c}±0.02
G5	41.0^{d}±1.53	3.0^{c}±0.01	0.44^{c}±0.02

G1: Negative control .,G2: positive control group (Hyperlipidemic rats).,G3:Hyperlipidemic rats that orally administrated water extracts flaxseed.G4 :Hyperlipidemic rats that orally administrated water extracts Mustard, G5 : Hyperlipidemic rats that orally administrated water extracts psyllium
Data are presented as means ± SDM (n=5) & Means within a column with different letters are significantly different at $P \leq 0.05$.

Histopathological results of the liver

Concerning HFD (high-fat diet) and treated groups, various histopathological alterations have been observed in the liver and kidney compared to the control group (Figures 1–5). Figure 2. showed a normal structure for the negative control (no histopathological changes). While positive control G2 which fed on HFD, revealed vacuolation of tunica media Figure 2. showed the histopathologic alterations in the rat's liver. While rats' livers from groups treated with 2 ml/day of psyllium or flaxseed extracts showed little fatty degeneration of hepatocytes (Figs. The slides in Figure 3. showed the histopathological alterations for liver from groups that fed on HFD with either F or M and P. Our results are in line with (Xing et al., 2017) who recorded that in a rat study assessing the effect of Psyllium and Flaxseed extract on liver, rats drinking P and F extract showed no undesirable side effects in liver function.

Histopathological results of kidneys

Microscopically, the kidneys of rats from group 1 revealed the normal histological structure of renal tissue (Figure 1). On the other hand, the kidneys of rats from group 2 showed vacuolation of the endothelial lining, glomerular tuft, and renal parenchyma. Some examined sections from Group 3 showed no histopathological changes (Fig. 3), whereas other sections revealed slight congestion of renal blood vessels and vacuolation of the epithelial lining of some renal tubules. Examined sections from Group 4 showed no changes except slight congestion of the glomerular tuft and vacuolation of the epithelial lining of some renal tubules. Meanwhile, the kidneys of group 5 rats showed necrobiosis of the epithelial lining of some renal tubules and slight congestion of the glomerular tuft.

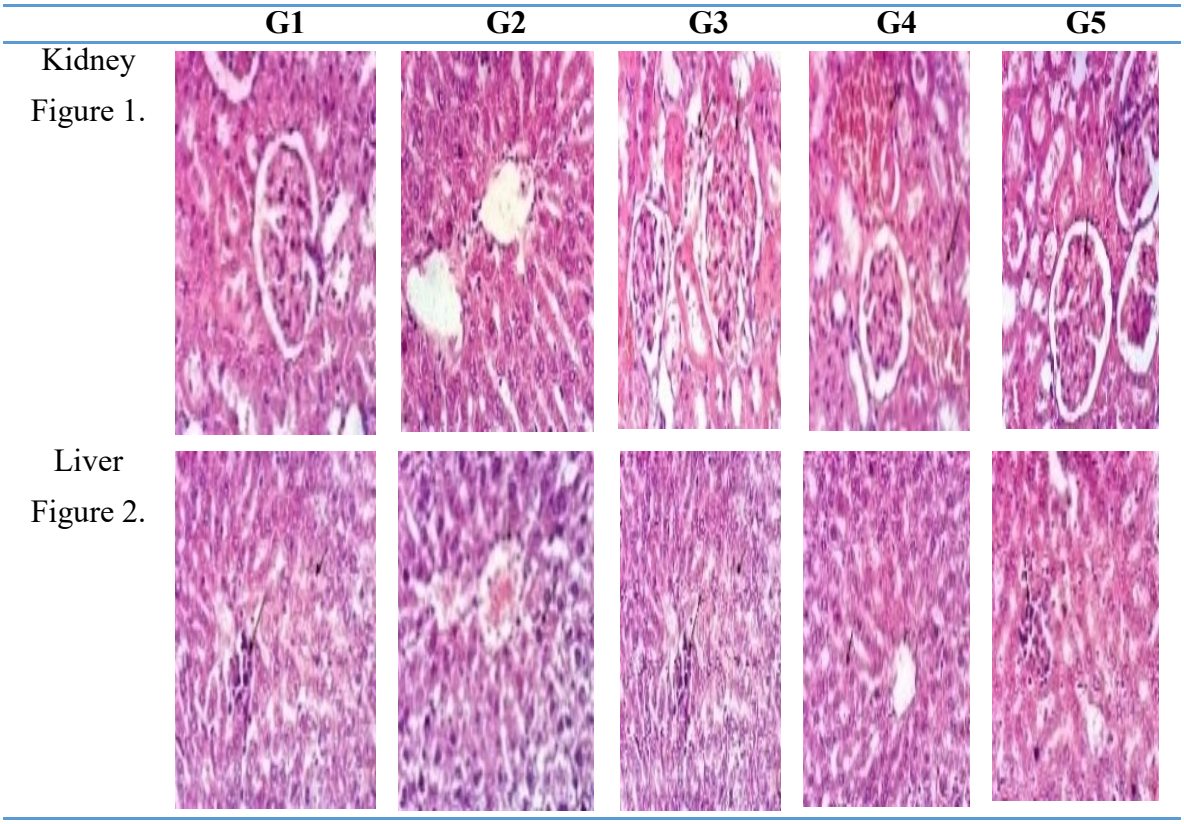

Figure 1. Rat's kidney of different treated groups: G1) Rat' kidney of rats from negative control showing normal histological structure of Rat's kidney of rats from positive G2. G3 Rat's kidney of rats from group fed on HFD with 2ml/day/rat water extract of flax seed. G4 Rat's kidney of rats from group fed on HFD with 2ml/day/rat water extract of mustard. G5 Rat's kidny of rats from group fed on HFD with 2ml/day/rat water extract of psyllium . .

Figure 2. Rat's liver of different controls (positive control G1) Rat's liver of negative control . G2) Rat's liver of negative control G3 Rat's liver of rats from group fed on HFD with 2ml/day/rat water extract of flax seed. G4 Rat's liver of rats from group fed on HFD with 2ml/day/rat water extract of mustard. G5 Rat's liver of rats from group fed on HFD with 2ml/day/rat water extract of psyllium.

4. Conclusion

It could be concluded that psyllium, flaxseed, and mustard extracts could be used as successful fat replacers in baked products, especially cakes, thus allowing the production of potentially healthier food items. Thus, it could be quite worthwhile for commercial applications of healthy food. However, 50 and 100% fat replacement with psyllium, mustard and flaxseed extracts resulted in cakes with highly acceptable sensorial properties with a lower fat content. In addition, results pointed out the promising role of psyllium, mustard, and flaxseed water extract as a natural product on liver and kidney function enzymes and the lipid profile of hyperlipidemic rats.

References

AACC. American Association of Cereal Chemists (2002). Approved Methods of the AACC. 8th Edition, American Association Cereal Chemistry. Method 54-21, St. Paul.

AL-ASkalany, S.A. (2012).The effect of Okra and Cucumber extracts in improving elderly biological parameters. J. Biol. Chem. Environ. sci., 7(4):589-606.

AOAC (2019).Official Methods of Analysis of the Association of Official Analytical Chemists. 21th Edition, Washington DC,USA.

Aziz, S.S., El-Zayat, M.M. and El-Khateeb, A. Y. (2020). Phytochemical Characterization, Antioxidant and Antimicrobial Activities of *Brassica juncea* (L.) Mustard Seeds Aqueous and Ethanolic Extracts. J. of Plant Production, Mansoura Univ., 11 (2):85 – 88.

Baixauli, R., Salvador, A. and Fiszman, S. M. (2008). Textural and colour changes during storage and sensory shelf life of muffins containing resistant starch. Eur. Food Res. Technol., 226: 523–530.

Bancroft, J.D. and Gamble, M.(2008).Theory and Practice of Histopathological Tecniques. 6th edition, Churchilllivingstone. NewYork. London and Madrid

Bavaro, A. R., Di Biase, M., Conte, A., Lonigro, S. L., Caputo, L., Cedola, A., Del Nobile, M. A., Logrieco, A. F., Lavermicocca, P., andValerio, F. (2021). Weissellacibaria short-fermented liquid sourdoughs based on quinoa or amaranth flours as fat replacer in focaccia bread formulation. Int. J. Food Sci. Technol., 56(7):3197–3208.

Belorio,M. , Marcondes, G. and Gómez, M. I. (2020) Influence of psyllium versus xanthan gum in starch properties. Food Hydrocolloids , 105:105843.

Bennion, E.B. and Bamford, G.S.T. (1997) The Technology of Cake Making. 6th Edition, Blacking Academic and Professional, Chapman & Hall, London, Pp2-328.

Bhatty, R.S. (1993). Further compositional analysis of flax: mucilage, trypsin inhibitors and hydrocyanic acid. J. Am. Oil Chem. Soc., 70:899-904.

Borneo, R., Aguirre, A. and Leon, A. E. (2010). Chia (Salvia hispanica L) gel can be used as egg or oil replacer in cake formulations. J. Am. Diet. Assoc., 110 (6): 946-949.

Brand-Williams, W., Cuvelier, M.E. and Berset, C. (1995). Use of a free radical method to evaluate antioxidant activity. LWT-Food Tech., 28: 25-30.

Brookfield Manual (1998) Brookfield Manual Operating Instruction. No.M/98-211-B0104. Brookfield Engineering Laboratories Inc., Middleborough.

Buckner, A.L., Buckner, C.A., Montaut, S. and Lafrenie, R.M. (2019)Treatment with flaxseed oil induces apoptosis in cultured malignant cells. Heliyon, 5, e02251

Colla, K., Costanzo, A., and Gamlath, S. (2018). Fat replacers in baked food products. Foods, 7 (12):192-199.

Divakaran, M.; Babu, K.N. Mustard (2016). In Encyclopedia of Food and Health; Caballero, B., Finglas, P.M., Toldrá, F., Eds.; Academic Press: Oxford, UK, pp. 9–19.

Drury, R.A. and Wallington, E.A. (1980) Carletons Histological Technique. 5thEd., Oxford University, New York. Toronto

Essam, E. and Maha, M. (2012). The protective effect of Turnip leaves against oxidative stress induced by high cholesterol diet in adult rats. World Appl. Sci. J. ,20 (1): 154 – 163.

El-Sayed, M.A., Shaltout, O.E., El-Difrawy, E.A, Osman, H.M. and Abo-El Naga, M.M. (2014).Production and Evaluation of Low Fat Cake Containing Flaxseed and Okra gums as a Fat Replacer. Alex. J. Food Sci. Tech., 11(1): 53-60

Felisberto, M.H.F., Wahanik, A.L., Gomes-Ruffi, C.R., Clerici, M.T.P.S., Chang, Y.K. and Steel, C.J., (2015). Use of chia *(Salvia hispanica L.)* mucilage gel to reduce fat in pound cakes. Alex. J. Food Sci. Tech., 63(2):1049-1055.

Gammone, M.A., Riccioni, G., Parrinello, G., D'Orazio, N. (2019). Omega-3 polyunsaturatedfatty acids: Benefits and endpoints insport. Nu trients, 11, 46.:1-16.

Grygier, A. (2015) .Mustard seeds as a bioactive component of food. Food Rev. Int. 2022, 1–14; ahead-of-print. https://oi.org/10.1080/87559129.2021.2015774.

Haley, A.P.M.: Gonzales, T.N. Tarumi, T. and Tanaka, H. (2013), "Dyslipidemia links obesity to early cerebral neurochemical alterations, Obesity, 21, (10):2007–2013.

Hidalgo, F. JandZamoraR. (2005) Coordinate contribution of lipid oxidation and Maillard reaction to the nonenzymatic food browning. Crit Rev Food SciNutr ,45(1):49-59.

Hussien, Hanan A. (2016).Using Vegetable puree as a fat substitute in cakes. Int. J. Food Sci. Nutr.. 5 (40), 284-292.

Ibrahim, O.S. ,Alfauomy, G.A., and Ali, M.M.A. (2016). Influence of Corn and Millet Starches as Fat Substitutes on Cake Quality. Inter. J. Nutri. and Food Sci. 5(5): 352-360.

Imran, M., Ahmad, N., Anjum, F.M., Khan, M.K., Mushtaq, Z., Nadeem, M. and Hussain, S. (2015). Potential protective properties of flax lignan secoisolariciresinoldiglucoside. Nutr. J. 14, 71-82.

Jabbar, A., Yasin, M.T., Ahmad, K., Abdullah, R., Hayat, M.Q., Qazi, N., Sultan, I.N., Amanat, K., and Bokhari, S.A.I. (2020). Exploration of the Phytochemicals, Antimicrobial and Antioxidant Activities of *Plantago Ovata* (Ispaghol). Indo Am. j. pharm., 7(4), 440– 447

Klop, J.B., Elte, W.F. and Cabezas, M.C. (2013) "Dyslipidemia in obesity: mechanisms and potential targets," Nutrients,. 5 (4):1218–1240.

Kumar, R and Shsrma, K. (2013).Biodegradable polymethacrylic acid grafted psyllium for controlled drug delivery sustems. Front chem. Sci. Eng., 7(1):116-122.

Kumar, Y. (2021). Development of low-fat/reduced -fat processed meat products using fat replacers and analoguesFood Rev. Int., 37(3): 296–312.

Ladak, Z., Khairy, M., Armstrong, E.A. and Yager, J.Y.(2021) Chapter 11-Glucosinolates: Paradoxically beneficial in fighting both brain cell death and cancer. In Nutraceuticals in Brain Health and Beyond; Ghosh, D., Ed.; Academic Press: Cambridge, MA, USA, pp. 155–167.

Li, C., Xin-Bo, M., Yu-Hong, L., Shi-Cheng, P., Yi -Ping, F. and Min, W. (2011) Effects of persimmon leaf total flavonoid on enzyme of lipoprotein metabolism and antioxidation in hyperlipidemia rats. Chin. J. Natu. Med., 9, 74.

Li, J.-M., and Nie, S.P. (2016). The functional and nutritional aspects of hydrocolloids in foods. Food Hydrocolloids, 53: 46–61

Lim, D.W., Kim, Y.T, Yang, Y.J., Kim, Y.E and Han, D. (2017). Anti-obesity Effect of Artemisiacapillaris Extract in High-Fat Diet-Induced Obese Rats. Molecules, 18: 9241 – 9252.

McGuire, R.G. (1992) Reporting of Objective Color Measurements. HortScience, 27: 1254-1255.

Melrose, J. (2019). The glucosinolates: A sulphur-glucoside family of mustard anti-tumour and antimicrobial phytochemicals of potential therapeutic application. Biomedicines, 7, 62:1-27.

Min-Suk, R., Sun-Young, L., Richard, H.D., and Dong-Hyun, K. (2003). Antimicrobial Effects of Mustard Flour and Acetic Acid against*Escherichia coli* O157:H7, *Listeria monocytogenes*, and*Salmonella enterica* Serovar Typhimurium Appl. Environ. Microbiol., 69 (5); 2959–2963

Mislivec, P.B, Beuchat, L.R. and Cousin, M.A. (1992). Yeast and molds. pp. 239-249. In: Compendium of Methods for the Microbiological Examination of Foods, 3rd ed. -Vanderzant. C. and Splittstoesser, D.F. (eds.). American Public Health Association, Washington, D.C.

Mossel, D.A.A. Corry, J.E.L., Struijk, C.B., and-Baird, R.M., (1995). Essentials of the microbiology of foods: a textbook for advanced studies. Chichester (England): John Wiley and Sons. 287-289.

Mousa, R.M.A. (2021). Development of 95% fat-free hamburgers using binary and ternary composites from polysaccharide hydrocolloids and fruit peel flours as fat replacer systems. J. Food Process. Preserv., 45(7), e15457.

Murtaza, M.S., Sameen, A., Huma, N., and Hussain, F. (2017). Influence of hydrocolloid gums on textural, functional and sensory properties of low-fat cheddar cheese from buffalo milk. Pak. J. Zool., 49(1), 27–34.

Niu Y, Xie Z, Hao J, Yao W, Yue J, Yu L. (2012). Preparation of succinylated derivatives of psyllium and their physicochemical and bile acid-binding properties. Food Chem., 132(2):1025–32.

Oostenbach, L.H., E. Slits, E. Robinson, and G. Sacks. (2019). Systematic review of the impact

of nutrition claims related to fat, sugar and energy content on food choices and energy intake. BMC Public Health 19 (1):1296–1306

Patel, A.R. and Dewettinck, K. (2016). Edible oil structuring: An overview and recent updates. Food & Function 7(1):20–29.

Patton, C.J. and Crouch, S.R. (1977). Enzymatic colorimetric method for determination of urea in serum. Anal .Chem.,49,464-469

Peng, X., and Y. Yao. (2017). Carbohydrates as fat replacers. Annu. rev. food sci. technol., 8:331–51.

Prosky, L., Asp, N., Schweizer, T, Devries, S. and Furda, I.. (1988) Determination of insoluble, soluble and total dietary fiber in food and food products: interlaboratory study. J. Assoc. Off. Anal. Chem., 71: 1017-1023.

Purlis , E. (2010) Browning development in bakery products – A review. J Food Eng., 99: .239–249.

Rafiee, M., Mortazavi, P., and Asghari, A. (2022). Evaluation of Plant agoovataethanolic extract on histopathological, immunohistochemical and biochemical indexs in the liver of BDL-induced cholestatic rats. 22 April 2022, PREPRINT (Version 1) available at Research Square [https://doi.org/10.21203/rs.3.rs-1492690/v1].

Rahman, M., Khatun, A., Liu, L. and Barkla, B.J. (2018). Brassicaceae mustards: Traditional and agronomic uses in Australia and New Zealand. Molecules, 23: 231.

Reeves, P.G., Nielsen, F.H. and Fahey, G.C. (1993) AIN-93 Purified diets for laboratory rodents; final report of the American institute of Nutrition Ad HOC writing committee on the reformulation of the AIN-76 a rodent diet .J.Nutr.,123(12):1939-195

Reitman S. and Frankel S. (1957) Acalormetric method for the determination of glutamic oxalacetic and glutamic pyruvic transaminase. J. Clin. Path., 28:56-63.

Rios, R.V., Garzón, R., Lannes, S.C. and Rosell, C.M., (2018). Use of succinyl chitosan as fat replacer on cake formulations. LWT, 96:.260-265.

Rodríguez-García, J., Puig, A., Salvador, A. and Hernando, I. (2012). Optimization of a Sponge Cake Formulation with Inulin as Fat Replacer: Structure, Physicochemical, and Sensory Properties. J. Food Sci., 77; 189-197.

Romero-Baranzini, O.G. Rodriguez, G.A. Yanez-Farias, J. M. Barron-Hoyos, P. and Rayas-Duarte (2006). Chemical, Physicochemical, and Nutritional Evaluation of Plantago (Plantago ovata Forsk). Cereal Chem., 83(4):358–36.

Saha, D., and Bhattacharya, S. (2010). Hydrocolloids as thickening and gelling agents in food: a critical review. J. Food Sci. Technol., 47(6): 587 –597.

Schermer, S. (1967). The blood Morphology of laboratory Animals. Philadelphia; F.A.Davies Co.,P.359

Sharif, A.M., Naghmachi, J., and Khosravani S.A.M. (2011).A Study on Antimicrobial Effects of Plantago Psyllium. Armaghane Danesh, 16(2): 191-199.

Singleton, V.L. and Rossi, J.A. (1965)Colorim- etry of Total Phenolics with Phosphomolybdic Phosphotungstic Acid Reagents. Am. J. Enol. Vitic. 16, 144-158

Steel, R.G.D., Torrie, J.H. and Dicky, D.A. (1997) Principles and Procedures of Statistics, A Biometrical Approach. 3rd Edition, McGraw Hill, Inc. Book Co., New York, 352-358.

Stein E.A. (1986). In :Textbook of Clinical Clinical Chemistry. N.W. Tiez,ed. W.B. Saunders, Philadelbhia., pp.879-886, 1818, 1829.

Son,H. and Song, K.B. (2017). Antimicrobial Activity of Flaxseed Meal Extract against Escherichiacoli O157:H7 and Staphylococcus aureus Inoculated on Red Mustard. J. Microbiol. Biotechnol., 27(1), 67–71.

Swanson, K.M.J., Busta, F.F., Peterson, E.H. and Johnson, M.G. (1992). Colony count methods. PP. 75-95. In: Compendium of Methods for the Microbiological Examination of Foods, 3rd ed. Vanderzant C. and US Department of Agriculture, 2004. Agricultural Research Service, USDA National Nutrient Database for Standard Reference, Release 17, Nutrient Data Laboratory Home Page,

http:// www. nal. Usda .gov /fnic /foodcomp.

Takasaki, Y. (2005). "Serum lipid levels and factors affecting atherogenic index in Japanese children," J Physiol Anthropol Appl Hum Sci, 24, (4) : 511–515.

Variey, H., Gewenlock, A. and Bell, M. (1980). Practical Clinical Biochemistry, Vol.1. 5th Ed. Pp741-897. London; William Medical. Books. Ltd.

Vassiliki, P. and Vassiliki, O. (2013). The effect of fat replacers on batter and cake properties. J. Food Sci., 78: 1495-1502.

Vessby, B. (2000). "Dietary fat and insulin action in humans," British Journal of Nutrition, vol. 83, supplement 1, pp. S91–S96, 2000.

Wallach, J. (1992). Interpretion of diagnostic tests. A synopsis of laboratory medicine.5th Ed. A little brown hand book, Boston.

WHO, World Health Organization. (2018). Healthy diet- fact sheet No: 394. Last Modified August, 2018. Accessed June 26, 2021.

Woods, S., Seeley, R., Rushing, P., D'Alessio, D. and Tso, P. (2003) A Controlled High-fat diet induces an obese syndrome in rats. J. Nutr., 1081-1087.

Xing, L.C., Santhi, D., Shar, A.G., Saeed, M., Arain, M.A., Shar, A.H., Bhutto, Z.A., Kakar, M.U., Manzoor, R., Abd El-Hack, M.E., Alagawany, M., Dhama, K. and ling, M.C.2017. Psyllium Husk (*Plantago ovata*) as a Potent Hypocholesterolemic Agent in Animal, Human and Poultry. Int J Pharmacol. , 13: 690-697.

Zahn, S., Pepke, F. and Rohm, H. (2010). Effect of inulin as a fat replacer on texture and sensory properties of muffins. Int. J. Food Sci. Technol, 45(12): 2531–2544.