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EFFECT OF FAT QUALITY AND FRYING ON GROWTH AND SOME BIOCHEMICAL ASPECTS IN RATS

(With 7 Tables and 4 Figures)

By

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**تأثير نوعية الزيوت واستعمالها في التحمير على النمو
وبعض القياسات البيوكيميائية في الفئران**

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استخدمت في هذه الدراسة عدد ٢٤٠ فأر أبيض نامي عمر ٣ أسابيع حيث تم تقسيمهم إلى ١٢ مجموعة متساوية العدد تحتوى الواحدة منها على ٢٠ فأرو لقد خصصت المجموعات لإجراء تجربتين منفصلتين حيث استمرت كل منهما لمدة ٦ أسابيع وذلك بهدف اختبار تأثير نوعية الزيوت وكذلك تأثير استخدامها في التحمير. أجريت التجربة الأولى على ٦ مجموعات من الفئران حيث اعتبرت المجموعة الأولى مجموعة ضابطة تم تغذيتها على عايقه محتوية على ١٠% زيت نرة - أما المجموعات الخمسة الأخرى فتم تغذيتها على التوالي بزيت عباد الشمس- زيت بذرة القطن- زيت النخيل- زيت النخيل المهدرج- دهن الزبد وفي التجربة الثانية استخدمت نفس أنواع الزيوت السابقة ولكن بعد استعمالها في التحمير حيث أجريت علي مجموعات الفئران الستة المتبقية و التي قسمت كل منها إلى مجموعتين لاختبار فترتين من التحمير لكل زيت إحداهما ساعتين والأخرى أربعة ساعات. تم تقدير المقاييس الخاصة بتقييم كفاءة الأداء والقياسات الكيموحيوية وكذلك الباثولوجية للمقارنة بين مصادر الزيوت المختلفة وكذلك تأثير التحمير علي جودة الزيوت وقيمتها الغذائية وكذلك علي صحة الحيوان. أوضحت النتائج أن الفئران المغذاه علي زيت عباد الشمس وكذلك زيت بذرة القطن الطازج سجلت أحسن نمو مع أعلى معدلات إستفادة من الغذاء بينما أدي زيت النخيل المهدرج وزيت النرة إلي تأخر النمو وتقليل كفاءة التحويل

الغذائي كما إنه لا يوجد إختلاف معنوي في وزن الأعضاء بين لمجموعات المختلفة ماعدا في حالة زيت عباد الشمس حيث أدى إلى تسجيل أقل وزن للكبد. أما صورة الدهون في سيرم الدم فقد أوضحت أن إستهلاك زيت الذرة و النخيل أدى إلى أعلى تركيز للدهون الثلاثية والكوليسترول الكلي والليبوبروتين منخفض الكثافة و كذلك أدى إلى وجود مستوى عالي من الليبوبروتين عالي الكثافة. إستعمال الزيوت في التخمير أدى إلى إحداث كثير من التغيرات الكيمائية وخفض درجة عدم التشبع وزيادة تكون مركب البنزوأبيرين والمالونالدهيد وخاصة في زيت الذرة وزيت بذرة القطن. إستخدام زيت الذرة وزيت النخيل المهدرج وكذلك دهن الزبد في التخمير لمدة ساعتان أدى إلى تقليل النمو ومعدل الإستفادة من الغذاء وقد كانت هذه الدهون أكثر تأثرا بزيادة مدة التخمير إلى أربع ساعات. تلاحظ أن أعلى إرتفاع في مستوى الكوليسترول والليبوبروتين المنخفض الكثافة وكذلك نسبة الكوليسترول الكلي إلى الليبوبروتين عالي الكثافة قد ظهرت في مجموعات زيت النخيل المستعمل لمدة إثنان وأربع ساعات وكذلك زيت النخيل المهدرج المستعمل لمدة أربع ساعات. أظهر الفحص الباثولوجي عدم تأثر أنسجة الكبد والكلى والقلب وخاصة مع زيت عباد الشمس وزيت القطن الطازج والمستعمل في التخمير وقد أظهر زيت النخيل المهدرج تغيرات باثولوجية واضحة تتراوح بين علامات التهاب بسيطة في حالة إستعمال الزيت الطازج إلى تدمير وتليف بعض الخلايا في حالة الزيت المستعمل. إن النتائج السابقة قد أوضحت أن زيت عباد الشمس يعتبر من أحسن الزيوت تأثيرا على صحة الإنسان سواء إستخدم طازجا أو للتخمير.

SUMMARY

Two hundred and forty growing albino rats, of three weeks old were allotted into 12 equal groups each of 20 rats and specified for 2 separate experiments performed for testing oil quality and frying and each extended for 6 weeks. Six rat groups were assigned for experiment I of which the first group was taken as the control and fed on the diet supplemented with 10% corn oil. The other five groups were set for fresh sunflower oil-, cottonseed oil -, palm oil-, hydrogenated palm oil- and butter fat- treatments. In experiment II the same kinds of fat used in Exp. I were tested after being fried using the remained six rat groups. Each of the six groups in experiment II was divided equally into two subgroups in order to test two frying periods 2&4 h. The performance efficiency, biochemical and histopathological parameters were traced in order to compare among the different fat sources and test the effect of frying on fat quality, nutritional value and animal health. The results showed that rats fed on fresh sunflower oil- and cottonseed oil-diets showed the best gain and used the food at the best rate of conversion (3.03 & 3.16) while hydrogenated palm and corn oils resulted in retarded growth and inefficient food conversion (3.79 & 3.56). Statistically the organ weight did not differ among the oils used except in case of sunflower oil which showed the smallest liver weight. Serum lipid

profile of rats showed that the consumption of corn and palm oils resulted in highest levels of triglycerides, total cholesterol and LDL-cholesterol and high HDL- cholesterol concentrations while sunflower and cottonseed oil groups had the most optimal lipid profile. Frying caused several chemical alterations, decreases in the degree of the unsaturation and increases in the formation of malonaldehyde and benzo(a)pyrene contents especially in corn and cottonseed oils. Rats fed on 2 h- used corn, hydrogenated palm and butter fats gained poorly and inefficiently utilized food and also they were greatly affected by frying for 4 hours. Highly elevated levels of serum total cholesterol, LDL-cholesterol and total cholesterol/HDL-cholesterol ratio was observed with 2&4 h- fried palm oil and 4h-fried hydrogenated palm oil. Liver, kidneys and heart showed normal histological structures especially in fresh and used frying sunflower and cottonseed oil-groups. In contrast, hydrogenated palm oil was the only oil caused clear histopathological changes ranged from slight inflammatory reactions (with fresh oil) into degenerative and fibroid changes (fried one). Conclusively, sunflower oil can be considered the best oil for human health used as a salad oil or cooking oil.

Key words: *Fat quality, frying, biochemical parameters, Rats, nutritional value.*

INTRODUCTION

The importance of diet in both health and disease has long been recognized. Recently, diets are being designed in an attempt to promote health and modify different diseases. Nutrients are being considered for their ability to improve health and not just avoid deficiencies.

Oils and fats play an important role in human nutrition because they are sources of concentrated energy and of the essential fatty acids. In addition lipids improve the palatability of foods and serve as a vehicle for the delivery and absorption of fat-soluble nutrients. Body fat or adipose tissue helps to protect vital organs and provides a vital reserve to meet demands when other energy sources are unavailable.

Edible fats, both of animal and vegetable origin, consist primarily of triglycerides of fatty acids which are usually of either 16 or 18 carbon atoms and may be saturated, unsaturated, or polyunsaturated. Animal fats also contain cholesterol, either free or combined with a fatty acid as a cholesterol ester. Vegetable oils have no cholesterol but contain plant sterols which are poorly absorbed by the animal body and, in fact, interfere with the absorption of cholesterol.

The richest source of fat in the diet is the vegetable oils (such as that of corn, sunflower seed, and cottonseed, olive and vegetable shortenings) or the animal fats (such as lard and butter). Dietary fat is consumed in a fresh state or used in cooking or frying of foods.

Young children need dietary fat and cholesterol to sustain growth and to maintain a healthy body (Vergroesen and Crawford, 1989). Adults, on the other hand, are advised to consume a diet containing lower total fat, especially saturated fat and cholesterol, which are linked to an increased risk of cardiovascular diseases. There was considerable evidence that the type of dietary fat could affect plasma cholesterol concentration and lipoprotein metabolism (Terpstra *et al.*, 2000). Saturated fat is often found in animal products, such as meat, dairy products, and in certain vegetable products, such as coconut, palm kernel oil and hydrogenated shortenings, and margarines. Saturated fatty acids vary in their degree of cholesterol influences. The saturated fatty acids (12, 14 & 16 C) elevated the circulatory levels of plasma LDL-cholesterol (Spady *et al.*, 1993 and Lambert *et al.*, 1996), a major risk factor for atherosclerosis, with the exception of stearic acid which exerts its hypocholesterolemic effect by its rapid conversion to oleic acid in the body (Bonanome and Grundy, 1988 and Khachadurian, 1990)

Hydrogenated oils contain considerable amounts of trans fatty acids which were recently shown to have adverse effects on the blood cholesterol profile (Mensink and Katan, 1990; Nestel *et al.*, 1992).

Conversely, consumption of n-6 and n-3 polyunsaturated fatty acids were associated with decreased blood pressure and serum cholesterol (Connor and Connor, 1990; Iacono and Dougherty, 1993). Plant fats are predominantly polyunsaturated fatty acids. Two are essential for human, these are linoleic acid (18: 2n₆) and α -linolenic acid (18: 3n₃) (Simopoulos *et al.*, 1991 and Connor *et al.*, 1992). Linoleic acid is the most prevalent PUFA and is the predominant FA in safflower, sunflower, corn, cottonseed, soybean and sesame oils.

There was a growing concern that habitual intake of large quantities of PUFA, when insufficiently protected by antioxidant (Vit E), might induce atherosclerosis and carcinogenesis and this probably because they were very susceptible to peroxidation and production of free radicals (Kok *et al.*, 1994 and Fang *et al.*, 1996). Fortunately, vegetable oils containing unsaturated FAs also have large amount of Vit E.

The vegetable oils are now used widely for deep frying. Because of the association between heart diseases and saturated fats (FAO, 1978),

there is an increased tendency for the more unsaturated oils, or blends of them, to be marketed. The frying of fats, and especially the highly unsaturated ones, results in thermoxidative and hydrolytic alterations. The deterioration of lipid is correlated with its content of the unsaturated FAs (Gere, 1982; Dobarganes *et al.*, 1988; Cuesta *et al.*, 1991 and Arroyo *et al.*, 1992). Furthermore, the rate of oxidation depends on factors such as temperature, frying time, antioxidant content and kind of fried food (Goburdhun and Jhurree, 1995; Tyagi and Vasishtha, 1996).

Heating causes several chemical alterations, e.g., elevation in the level of free FAs, changes in peroxide value, decrease in the degree of unsaturation and antioxidant content, and the formation of polymers resulting in a decreased nutritive value of the oils (Kishida, *et al.*, 1990; Warner and Mounts, 1993). As a consequence of the production of these decomposition products certain oils could present toxicity, possibly affecting development and growth, as has been reported in rats (Billek, 1985; Lopez-Varela *et al.*, 1995 a; Sanchez-Muniz *et al.*, 1997).

Cuesta *et al.* (1993) reported that disappearance of polyunsaturated FAs during the frying process has been considered the primary cause of the hypercholesterolaemia observed in animals that consume oils used repeatedly for frying potatoes and which undergo a considerable degree of alteration. Frying was considered to be a source of all trans-FAs which reported to raise cholesterol in blood (Mensink and Katan, 1992; Lopez-Varela and Sanchez-Muniz, 1997).

In spite of this most studies suggested that, in moderately heated oils, the suspected toxic components were not present in significant quantities, and combined with an, otherwise, adequate diet, these oils had only limited or no detrimental effects when fed to animals (Hemans *et al.*, 1973 and Izaki *et al.*, 1984).

The present study was designed to test the feeding of four commonly used oils (corn, sunflower, cottonseed and palm oils), hydrogenated palm oil and butterfat as diet ingredients for rats. The testing was performed on the fat-sources as it is marketed or after being fried in the laboratory. The fats were added to the diet at the rate of 10% and continued to be fed for 6 weeks. The performance efficiency, haematological and histopathological parameters were traced in order to compare among the different sources and test the effect of frying on fat quality, nutritional value, and animal health.

MATERIALS and METHODS

1. Animal and management:

Two hundred and forty male albino rats (*Rattus norvegicus*) of 3 weeks-old, weighing approximately 75 g, were procured from Helwan breeding farm, Cairo, Egypt and housed in 12 separate clean cages. The animal house was kept under the ambient temperature and humidity and the animals received a photoperiod of 12 h light cycle. As the rats were brought to the animal house, they were kept for a preliminary period of one week and fed on a corn oil-supplemented diet to familiarize the animals with the feeding devices and environment. Thereafter, they were allotted into 12 equal groups each of 20 rats and specified for two separate experiments performed for testing oil quality and each extended for 6 weeks. The rats were fed on ad-libitum basis and clean water was continuously available. Body weight gain and feed intake rate were recorded weekly throughout the experiment, in addition to the health condition, biochemical parameters, and pathological lesions if any.

2. Diet preparation:

A corn oil-diet was formulated to satisfy the needs recommended and stated by NRC (1978) for growing rats. The optimal level of energy density in the diet was achieved by adding 10% of corn oil to the basal ingredients and considered as the control one. In the other experimental diets the energy density was achieved by using other fat sources nominated to be tested. The composition of the control diet is shown in Table (1).

The fats and oils used in the experimental diets are the sources commercially available for human consumption. To avoid the possibility of rancidity and spoilage the diet ingredients were mixed daily and with the fresh oils.

3. Experimental design:

The biological testing of oils and fats quality was designed to be performed in two separate experiments. The first was planned to trace the effect of oil addition and kind on health and biochemical profiles of rats, while the second experiment was to study the effect of frying on the quality of the oil, its nutritional value, and effect on rat health. Samples of the fresh and fried oils were stored for analysis.

3.1. Experiment 1:

Of the twelve rat groups, six were assigned for this experiment of which the first group was taken as the control and fed on the diet supplemented with a local highly reputed corn oil. The other five groups were set for fresh sunflower oil-, cottonseed oil-, palm oil-, hydrogenated palm oil (margarine)-, and butter fat (ghee)-treatments. In estimating the energy value of the different oil-diets the nominated oils were considered to contain the same energy (8 Kcal ME/ g fat).

Table 1: The composition of the control (corn oil -) diet

Ingredient	Percent
<u>Physical composition</u>	
Ground yellow corn	50.55
Soybean meal	19.15
Meat and bone meal	6.00
Wheat bran	13.50
Corn oil	10.00
Sodium phosphate, monobasic	0.30
Potassium chloride	0.15
DL-methionine	0.25
Mineral and vitamin premix	0.10
<u>Calculated chemical composition & energy value</u>	
Crude protein	18.01
Methionine & cystine	0.54
Lysine	0.89
Calcium	0.68
Phosphorus	0.45
Metabolizable energy (Kcal/kg)	3238

* MUVCO poultry mineral and vitamin premix.

3.2. Experiment 11:

In this experiment the same kinds of fat, used in experiment I, were tested after being fried experimentally. Laboratory frying was performed so as to simulate home use, and the pan-frying period varied from 2 to 4 h at approximately 205°C using potato sticks as the frying food. The 4h - treatment was effected in two 2 h-periods separated by 24 h of unuse.

The six rat groups were assigned respectively for the corn, sunflower, cottonseed, palm, hydrogenated palm oils and butter fat. Each

of the six rat groups was divided equally into two subgroups in order to test the two frying periods 2 & 4 h.

In formulating the six fat-diets the fried oils were incorporated, at the level of 10%, in a diet having the same physical and chemical composition as that in experiment I.

4. Sampling:

At the end of the experiment and after an overnight fast, five rats from each group were individually weighed and slaughtered for blood and tissue sampling.

4.1. Blood:

From each slaughtered animal a sample of about 5 ml blood was collected into a clean and dry centrifuge tube, left to clot at room temperature, and then centrifuged at 3000 r.p.m. for 30 minutes. The clear supernatant serum was harvested and kept frozen at -20°C for further measurements.

4.2. Tissues:

Liver, kidneys, and heart were excised, blotted and weighed then sampled. The tissue specimens were fixed in a buffered formaline for 24h, trimmed and then transferred into 70% alcohol for histopathological examination. In addition, samples of liver were kept frozen for the determination of hepatic lipids and enzyme activities.

5. Estimations:

5.1. Oil quality tests:

The identity and edibility of the tested oils and fats were evaluated by determining the iodine, saponification, acid and peroxide values using A.O.A.C. (1985) methods. Individual fatty acids of fresh and fried oils were determined by gas liquid chromatography according to Farag *et al.* (1992). Concentration of the decomposition products, benzo(a)pyrene and malonaldehyde, formed in the fried oils was measured using methods described by Dunn and Fee (1979) and Woyewoda *et al.* (1986) respectively.

5.2. Biochemical parameters:

Enzymatic colorimetric assays were employed with serum to determine triglycerides (Eggstein and Kuhlmann, 1974), total cholesterol (Trinder, 1969), HDL-cholesterol (Burnstein *et al.*, 1970), LDL-cholesterol (Bergmenyer, 1985) and phospholipids (Zilversnit and Davis, 1956). Liver functions were traced by determination of the serum enzymes; alkaline phosphatase (Kind and King, 1954) and aspartate and alanine aminotransferases (Reitman and Frankel, 1957); and estimating

the serum total protein following Peters (1968). The serum albumin was determined according to Doumas *et al.* (1971), while globulin was estimated using the equation: Globulin = total protein – albumin.

5.3. Histopathological studies:

Tissue specimens were embedded in paraffin wax, sectioned at 5 μ m, and stained with haematoxylin and eosin (H & E) for general histopathological examination using light microscope.

6. Statistical analysis:

The effect of diet (oil type), duration of oil consumption and their possible interaction on body weight, feed efficiency ratio, serum lipid profile and liver function were analyzed by using MANOVA according to Gomez and Gomez (1983).

RESULTS and DISCUSSION

The effect of fats and oils, fresh or used, on animal performance, liver function, and blood chemistry was exploited. The oils tested were five of plant origin (corn, sunflower, cottonseed and palm and margarine) and the sixth was of animal origin (butter – ghee). The results for each of the tested parameters were discussed for both the fresh (Exp. I) and fried (Exp. II) oils under the same title in order to ease the realization of frying effect .

I- Performance

1- Feed intake

In experiment I maintaining the experimental animals on diets containing 10% oil or fat showed a rate of food intake from 414.8 g in butter fat – diet to 531.8 in palm oil–diet throughout the 6 weeks of feeding (Table 2). Comparing the intake of the groups (2–6) with that of group 1 (Corn oil–group), it was found that while the sunflower and cottonseed groups nearly equated that of the control group , palm oil addition increased the intake by 15% and the hydrogenated palm oil or butter fat reduced it by 12 & 14% respectively. The results of other researchers are more conflicting in spite of using many oil and fat sources. Some found an improving effect of margarine on the feed intake of rats and the olive oil had a reducing effect (Maccoll *et al.*, 1996) while others found no significant differences between sunflower oil and animal fat (Ziemiński *et al.*, 1985; Biermat and Grajeta, 1997), linseed, sunflower and fish oils (Rustan *et al.*, 1992), coconut; olive; palm and sunflower oils (Calleja *et al.*, 1999) and between olive oil, high oleic sunflower oil and fish oil (Ruiz – Gutierrez *et al.*, 1999).

Frying the oils in experiment II did reduce the feed intake but the effect varied from one source to another. Frying for two hour had a clear negative effect in the palm oil – diet group where the intake was reduced by about 10% while the effect in the other groups did not exceed 4.5%. The decreasing effect was more clear as the frying time increased with a reduction reaching a highest degree (17.7%) with palm oil and followed by hydrogenated palm oil (15.3 %) and corn (10.4%). The registered effects of frying on feed intake in other studies differed from a significant lowering effect with oxidized palm oil (Owu *et al.*, 1998) and repeatedly used sunflower oil (Sanchez – Muniz *et al.*, 1992); to a non significant one with discontinuously (15 times) used oils (Lopez-Varela and Sanchez-Muniz, 1997).

2- Weight gain and feed conversion:

Feeding the animals on diets satisfying NRC (1978) requirements and supplemented with 10% fat substance of different sources and for 6 weeks induced a body weight gain ranging from 112 g in the hydrogenated palm oil–diet to a maximum of 165.9 g in the sunflower oil–diet. Expressing the figures in rounded figures and considering the gain of corn oil as 100 the other oils scored an extra gain of 22.9 % in sunflower oil, 17.5 % in cottonseed oil, 15.4% in palm oil while hydrogenated palm oil and butter fat reduced the gain by 17.3 and 9.9% respectively. The higher weight gain was also obtained with the consumption of sunflower oil when compared with lard (Biernat and Grajeta, 1997) and fish oil (Ruiz – Gutierrez *et al.*, 1999). On the other hand it cased the animal to gain less weight than those on the other oil-diets, butter; palm stearin; coconut; rapeseed; and olive oil (Trautwein *et al.*, 1997 and Calleja *et al.*, 1999). No differences in the rat weight gains connected with the type of fat were noted by Rustan *et al.* (1992) and Romijn *et al.* (1998).

Feed conversion indices calculate the amount of food in grams consumed for each gram of gain and eventually point to the efficiency of utilization and conversion in body tissues. The two oils, sunflower and cottonseed, showing the best gain used the food at the best rate of conversion (3.03 and 3.16 respectively) while the oils showing retarded growth (hydrogenated palm oil and butter fat) inefficiently converted the food (3.79 and 3.40). It is interesting to note that palm oil increased the growth rate due to an increase in feed intake and showed a feed conversion rate as low as butter fat. Corn oil in spite it was taken as the control it did not get high score in feed conversion (3.56) in addition to the modest growth.

Using the oils after frying for 2 hours had a slight decreasing effect on body weight gain starting from 4.7 % in butter fat to 8.0 % in corn, the only more significant effect was in the palm oil which reached to 13.1%. When the frying time was increased to 4 h, the decreasing effect on body gain was also nearly duplicated in corn, sunflower and cottonseed oils, triplicated in butter and quadruplicated in hydrogenated palm oil, while it did not add any reducing effect in palm oil.

Johnson *et al.* (1956), Rice *et al.* (1960) and Nwanguma *et al.* (1999) reported that thermal oxidation of corn oil led to a definite growth-depressing action while margarine base stock gave only slight growth depression, and was noted with butter fat (Johnson *et al.*, 1956; Rice *et al.*, 1960). Johnson *et al.* (1956) also found that this effect was not a permanent one as animals that were changed to a normal diet quickly recovered and grew to maturity. The growth-depressing effect appeared to be multiple in nature and may be due to an irritation of the intestinal tract, causing diarrhea, disturbance of normal metabolism, and possibly due to an enzyme – inhibiting or vitamin – destroying effect .

Also Sanchez-Muniz *et al.* (1997) reported that rats fed diets containing a repeatedly used frying sunflower seed – oil had significant lower weight gain and might be related to its lower feed intake or lower feed efficiency (Lopez Varela *et al.*, 1995a). Nielsan *et al.* (1985) pointed that almost all amino acids react with primary and secondary products of oxidized lipids, thereby decreasing the digestive utilization of AAs which may affect weight gains.

This was in contrast to the findings by Izaki *et al.* (1984), Kok *et al.* (1988) and Hageman (1991) in which no effect on body weight gain was observed in fried palm oil.

It could be concluded that corn, hydrogenated palm oil and butter fat are the oils which have the disadvantage of reducing growth and all the oils are greatly affected by frying for 4 hours except that of sunflower.

Frying in addition to decreasing the rate of body gain increased the feed cost of gain. The increase in cost in 2h – frying varied from an insignificant value 0.03g in sunflower oil to 0.2g food/g gain in hydrogenated palm oil. The effect was more clear in 4 h – frying in all diets except that of sunflower oil (0.1g) where it differed from 0.30 to 0.50 g with the highest figure also in the hydrogenated palm oil.

3- Relative organ weights:

The relative weight of the organs in case of feeding fresh oils differed from 2.97% in sunflower oil to 4.20% in hydrogenated palm oil

for liver, 0.60% in sunflower to 0.76% in corn for kidney and 0.29% in sunflower to 0.35% in corn for heart.

Statistically the organ weight did not differ among the oils used except in case of sunflower oil which showed the smallest liver weight. Trautwein *et al.* (1997) reported that livers of hamsters fed on the butter and palm stearin diets were significantly heavier after 5 w compared with that of coconut and sunflower oil-fed hamsters but after 7 w no differences in liver weight existed between the diets. Also, higher liver weights and liver weights as a percentage of body weight was observed by Terpstra *et al.* (2000) in the olive oil – fed hamsters than in those fed with palm oil or maize oil, although this effect was not statistically significant for the maize – oil group.

Frying did increase the weight of organs but by decimals of one unit percent (except sunflower oil 1.02) in liver and hundredths of one unit percent in kidney (except hydrogenated palm oil 0.1, butter fat 0.13) and heart. Increasing the time of frying did not make any great differences except in hydrogenated palm oil in liver, cottonseed and hydrogenated palm in kidney and palm oil in heart. Rodriguez *et al.* (1984) did not find any significant difference in the liver weight and hepatosomatic index of rats fed olive oil or palm oil used in frying. While Sanchez–Muniz *et al.* (1992, 1997) noted that rats fed sunflower oil and olive oil used several times for frying displayed a higher hepatosomatic index than those fed 1st or 2nd used frying sunflower and olive oils, respectively.

Thermally oxidized corn oil (Nwanguma *et al.*, 1999) and heated cottonseed oil (Rice *et al.*, 1960) caused significantly higher relative liver weights than the fresh oils. long–term feeding of heated oils led to increases in organ weights, especially in the liver (Izaki *et al.*, 1984 and Kok *et al.*, 1988) while shorter periods of feeding (4w) showed no changes in liver weights (Hageman *et al.*, 1991 and Lop'ez – Varela *et al.*, 1995).

II- Oil quality:

Quality characteristics of the fresh oils and fats (Table 6) showed them to be in accordance with the Egyptian standard (1993). Also from this Table it is indicated that there were analytically measurable changes due to oxidation, polymerization and hydrolysis (Cuesta, 1993; Lop'ez – Varela *et al.*, 1995; Sanchez –Muniz *et al.*, 1997). These changes were translated by increases in the acid, peroxide and saponification values and decrease in the iodine value. The oils did not rank the same orders for the all different chemical values, however, the used frying corn and

cottonseed oils recorded the highest values and the butter fat had the lowest ones. Steiner *et al.* (1993) reported that heating impaired quality in all 32 studied samples of vegetable oils; non-refined olive and sunflower seed oils were especially adversely affected. In Egypt, Farag *et al.* (1992) found that change in the chemical values indicating deterioration, due to frying, were much higher for cottonseed oil than for hydrogenated palm oil.

Quality of the used frying oils was more affected with extending the time of frying to 4h. Chang *et al.* (1978) reported that acidity % of corn oil and hydrogenated cottonseed oil was increased gradually during frying to 1.4% within 90 h and less than 60 h respectively. While Nolen *et al.* (1967) found small but irregular increases in the free fatty acid levels and peroxide value, and small in iodine values with the use of partially hydrogenated soybean oils, cottonseed oil, and lard for frying under practical restaurant – type frying conditions.

The concentration of benzo(a) pyrene and malonaldehyde compounds in fresh oils increased with heat treatment. There was a positive relationship between the frying time and the level of these compounds in the used oils. After 4 hours of frying, the concentrations increased by about 11.8, 10.5, 8.2, 6.7, 3.8 and 2.8% for corn, cotton, palm, hydrogenated palm, sunflower and butter fat each than its respective fresh oil. Howard and Fazio (1980) found that the highest concentration (43.7 g/kg) of benzo(a)pyrene was in oil from smoked dried coconut, while sunflower seed and palm kernel oils contained 10.6 and 40.1 mg/kg, respectively. Serag El-Din (2001) reported that the benzo(a)pyrene content of used-frying oils increased with the increase of frying time and corn oil recorded higher values when compared with the sunflower oil. Benzo(a)pyrene and malonaldehyde compounds have been shown to be mutagens and carcinogens as reported by Plakunov *et al.* (1987) and Hawkins *et al.* (1990).

Table 7 shows that the fatty acid composition of fresh corn, sunflower, cotton and palm oil is characterized by its high content of total unsaturated FAs (89.05, 86.93, 87.39 & 80.71 %) while hydrogenated palm oil and butter fat by its low content (61.71 and 71.62 % respectively). In all of these oils, C_{18:1} and C_{18:2} constitute more than 90% of the unsaturated fatty acids. The use of oils in frying resulted in a change in the degree of unsaturation (decrease in unsaturated FAs and increase in saturated FAs) reflected in a decrease of P/S ratio by about 60% in corn & sunflower oils after 4h.

III- Blood biochemical parameters:

The recorded results in Table 4 are in agreement with that obtained by Terpstra *et al.* (2000) who reported that the type of dietary fat could affect total cholesterol concentrations and lipoprotein metabolism. Rats fed on corn and palm oil-diets recorded the highest levels in serum triglycerides, total cholesterol and LDL-cholesterol concentrations and high level of HDL-cholesterol. Groot *et al.* (1988) found that hypertriglyceridemia found in palm oil-fed animals was due to less efficient catabolism and not to increased synthesis of plasma triacylglycerol. In contrast, Purushothama *et al.* (1994) reported that the palm oil had a favourable effect on the cholesterol metabolism similar to unsaturated sunflower oil. Also the obtained data showed that sunflower and cotton seed oil-fed rats had optimal lipid profile translated in a lowered total serum cholesterol and LDL-cholesterol levels by about 30% and total cholesterol / HDL-cholesterol ratio (risk factor) by 60 % when compared with the highest values recorded by corn and palm oils and hydrogenated palm oil respectively. Butter fat was intermediate. This coincides with that found by Trautwein *et al.* (1997), Romijn *et al.* (1998) and Sarkkinen *et al.* (1998), while Sarkkinen *et al.* (1998) added that the significant decrease in LDL-cholesterol with sunflower oil was probably through cholesterol malabsorption with a compensatory increase in cholesterol synthesis in addition to changes induced by dietary fatty acid composition.

Abnormal alteration in levels of protein fractions (decrease in albumin and increase in globulin) leading to significant decrease in albumin/globulin ratio (0.21), which reflect liver injury, was observed with hydrogenated palm oil and lesser by about 57% than that of sunflower oil (0.49). The figures for asparatate and alanine aminotransferase in the serum of rats fed palm and hydrogenated palm oils and hydrogenated palm and corn were nearly double that found in sunflower, corn and sunflower and butter fats respectively (Table 5).

Generally, the values of biochemical parameters recorded by fried oils were less than that of fresh oils and this may be due to the contribution of both hydrolysis by pancreatic lipase of the complex molecules formed during heating and the poor absorption of the released complex FA_s to the reduced absorbability of fried fats (Yoshida and Alexander, 1983 and Marquez-Ruiz *et al.*, 1993). Also the decrease in feed intake occurred with fried oils may also contribute to the differences recorded between fried and fresh fats.

The consumption of diets containing palm oil used in frying for 2 h induced highest serum total and LDL-cholesterol values and total cholesterol/HDL- cholesterol ratio and alanine and aspartate aminotransferases activities and exhibited a similar trend when fried for 4 h. Increasing the time of frying also had bad effect on the biochemical parameters of the hydrogenated palm oil group while that of sunflower and cottonseed oil fed – rat group showed the least effect at 2 or 4 hours of frying.

IV Histopathological studies:

There was no detectable histopathological changes in livers, kidneys and hearts of rats fed on diets containing fresh oils except slight inflammatory reactions were observed with corn oil, hydrogenated palm oil and butter fats. Richter *et al.* (1996) reported that livers of rats fed on sunflower oil -olive oil- or rapeseed oil- supplemented diets showed moderate non – degenerative fat infiltration of the hepatocytes while heart and aorta appeared normal.

The use of sunflower and cottonseed oils in frying for 2h had no detrimental effect on organ tissues and even with increasing the frying hours, healthy tissues were observed. On the contrary, livers of rats fed 2-&4- h used frying corn oil and palm oil showed histopathological changes e.g. hyperemia, inflammatory cell infiltration and kupffer cell proliferation. The severity of injury increased with 4-h frying hydrogenated palm oil where there were degeneration of the hepatocytes and fibrosis. Shibayama (1992) reported that ingestion of heated and oxygenated corn induced hepatic injury and might relate to liver cell membrane damage due to active O₂ radicals contained in heated and oxygenated corn oil. Kidney and heart of 4 h – used frying hydrogenated palm oil showed hyperemia and inflammatory cell infiltration.

From the results we can conclude that any oil had no symmetrical effect on the different tested parameters and sunflower oil can be considered the best for use as a salad or cooking oil.

REFERENCES

- Arroyo, R., C. Cuesta , C. Garrido- Polonio, S. Lopez – Varela and F.J. Sanchez – Muniz (1992): High performance size exclusion chromatographic studies on polar components formed in Sunflower oil used for frying. *J. Am. Oil Chem. Soc.* 69: 557
- Association of Official Analytical Chemists (A.O.A.C.) (1985): Official methods of analysis of the association of official analytical chemists. 14th ed., Washington, D.C. USA.

- Biernat, J. and H. Grajeta (1997):* Effect of high – fat cholesterol enriched diets on hypolipemic action of *Oenothera paradoxa* oil in rats. Part 1. Blood serum and liver lipids. *Nahrung* 41 Nr. 1: 46.
- Billek, G. (1985):* Heated fats in the diet in: *The Role of Fats in Human Nutrition*. Padley, FB. And J. Podmore (eds.) In collaboration with Bran, JP., R. Burt and B.W. Nicols, Ellis Horwood Ltd, Chichester, UK, PP 163.
- Bonanome, A. and S.M. Grundy (1988):* Effect of dietary stearic acid on plasma cholesterol and lipoprotein levels. *N Engl. J. Med.* 318 : 1244.
- Burnstein, M., H.R. Sholmick and R. Morfin (1970):* Rapid method for the isolation of lipoproteins from human serum by precipitation with polyanions. *J. Lipid Res.* 11: 583.
- Calleja, L., M.A. Paris A. Paul, E. Vilella, J. Joven, A. Jimenez G. Beltran, M. Uceda, N. Maeda, and J. Osada (1999):* Low – cholesterol and high fat diets reduce atherosclerotic lesion development in Apo E-Knockout mice. *Arterioscler Thromb. Vasc. Biol.* 19: 2368.
- Chang, S.S., R.J. Peterson and H. Chi-Tang (1978):* Chemical reactions involved in the deep – fat frying of foods. *J. Am. Oil Chem. Soc.* 55: 718.
- Connor, W.E. and S.D. Connor (1990):* *Adv. Intern. Med.* 35: 139.
- Connor, W.E., M. Neuringer and S. Reisbick (1992):* *Nutr. Rev.* 50: 21.
- Cuesta, C., F.J. Sanchez–Muniz, C. Garrido–Polonio S. Lopez–Varela and R. Arroyo (1993):* Thermoxidative and hydrolytic changes in sunflower oil used in frying with a fast turnover of fresh oil. *J. Am. Oil Chem. Soc.* 70: 1069.
- Cuesta, C., F.J. Sanchez – Muniz and I. Hernandez (1991):* Evaluation of non polar methyl esters by column and gas chromatography for the assessment of used frying olive oils. *J. Am. Oil Chem. Soc.* 68: 443.
- Dobarganes, M.C., M.C. Perez–Camino, G. Marquez–Ruiz (1988):* High performance size exclusion chromatography of polar compounds in heated and non heated fats. *Fat Sci. Technol.* 90: 308.
- Doumas, B.T., W.A. Watson and H.G. Biggs (1971):* Determination of serum albumin. *J. Clin. Chem. Acta.* 31: 87.

- Dunn, B.P. and J. Fee (1979):* Polycyclic aromatic hydrocarbon carcinogens in commercial foods. J. fish Res. Board. An. 36: 1469.
- Eggstein, M.L. and E. Kuhlmann (1974):* Enzymatic colorimetric method for determination of triglycerides. In Methods of Enzymatic Analysis. HU. Bergmyeyer ed., Academic Press, P. 1830.
- Egyptian Standard (1993):* Vegetable edible oils. Effect of sunflower seed oil used for frying on histological structure of rat organs. Bull. Nutr. Inst. Cairo, Egypt. 12: 48.
- Fang, J.L., C.E. Vaca, L.M. Valsta and M. Mutanen (1996):* Determination of DNA adducts of malonaldehyde in humans: effects of dietary fatty acid composition. Carcinogenesis. 17: 1035.
- FAO. (1978):* Specifications for Identity and Purity of Thickening Agents. Rome.
- Farag, R.S., F.M. Hewedi, S.H. Abu-Raiia and G.S. El-batoty (1992):* Comparative study on the deterioration of oils by microwave and conventional heating. J. Food Protec. 55: 707.
- Gere, A. (1982):* Studies of the changes in edible fats during heating and frying. Die Nahrung 26: 923.
- Goburdhun, D. and B. Jhuree (1995):* Effect of deep. Fat-frying on fat oxidation in soybean oil. Int. J. Food Sci. Nutr. 46: 363.
- Gomez, K.A. and A.A. Gomez (1983):* Statistical procedures for agricultural research. 2nd Ed. John Wiley and Sons. New York.680.
- Groot, P.H., B.C. de Boer, E. Haddeman, U.M. Houtsmuller and W.C. Hulsman (1988):* Effect of dietary fat composition on the metabolism of triacylglycerol-rich plasma lipoproteins in the postprandial phase in meal- fed rats. J. Lipid research. 29: 541.
- Hageman, G., H. Verhagen, B. Schutte and J. Kleinjans (1991):* Biological effects of short – term feeding to rats of repeatedly used deep-frying fats in relation to fat mutagen content. Food Chem. Toxicol. 29: 689.
- Hawkins, E.W., W.W. Walker, R.M. Overstreet, T.F. Lytle and J.S. Lytle. (1990):* Carcinogenic effects of some polycyclic aromatic hydrocarbons on the Japanese Medaka and Guppy in water borne exposure. The Science of the Total Environment. 94: 155.

- Hemans, C., F. Kummerow and E.G. Perkins (1973):* Influence of protein and vitamin levels on the nutritional value of heated fats for rats. *J. Nutr.* 103: 1665.
- Howard, J.W. and T. Fazio (1980):* Review of polycyclic aromatic hydrocarbons in foods. *J. Assoc. Off. Anal. Chem.* 63: 1077.
- Iacono, J.M. and R.M. Dougherty (1993):* *Annu. Rev. Nutr.* 13: 243.
- Izaki, Y. S. Yoshikawa and M. Uchiyama (1984):* Effect of ingestion of thermally oxidized frying oil on peroxidative criteria in rats. *Lipids* 19: 324.
- Johnson O.C., T. Sarkuragi, and F.A. kummerow (1956):* A comparative study of the nutritive value of thermally oxidized oils. *J. Am. Oil Chemists' Soc.* 33: 433.
- Khachadurian, A.K. (1990):* The role of cholesterol in atherosclerosis. *Advances in cholesterol research.* The telford press Inc., culdwell, New Jersey.
- Kind, P.R.N. and E. J. King (1954):* Estimation of alkaline phosphatase *J. Clin. Pathol.* 7: 322.
- Kishida, E., M. Oribe and S. Kojo (1990):* Relationship among malondaldehyde, TBA- reactive substances, and tocopherols in the oxidation of rapeseed oil. *J. Nutr. Sci. Vitaminol.* 36: 619.
- Kok, T.S., P.G. Harris and J.C. Alexander (1988):* Heated canola oil and oxidative stress in rats. *Nutr. Res.* 8: 673.
- Kok, T.M., F. Vaarwerk, I. Zwingman, J.M. Maanen and J. C. Kleinjans (1994):* Peroxidation of linoleic, arachidonic and oleic acid in relation to the induction of oxidative DNA damage and cytogenetic effects. *Carcinogenesis.* 15: 1399.
- Lambert, M.S., K. M. Botham and P.A. Mayes (1996):* Modification of the fatty acid composition of dietary oils and fats on incorporation into chylomicrons and chylomicron remnants. *British J. of Nutrition,* 76: 435.
- Lopez-Varela, S. and F.J. Sanchez-Muniz (1997):* Lipaemia and liver composition in pregnant rats consuming olive oil and olive oil used for frying.
- Lopez-Varela, S., F.J. Sanchez-Muniz and C. Cuesta (1995 a):* Decreased food efficiency ratio, growth retardation and changes in liver composition in rats consuming thermoxidized and polymerized sunflower oil used for frying. *Fd. Chem. Toxic.* 33: 181.

- Lopez-Varela S., F.J. Sanchez-Muniz, C. Garrido – Polonio, R. Arroyo and C. Cuesta (1995):* Relationship between chemical and physical indexes and column and HPS chromatography methods for evaluating frying oil. *Ernahrungswiss* 34: 308.
- Maccoll, A. J., K.A.C. James and C.L. Booth. (1996):* Erythrocyte morphology and filterability in rats fed on diets containing different fats and oils. *British Journal of Nutrition.* 76: 133.
- Marquez-Ruiz, G., M.C. Perez-Camina and M.C. Dobarganes (1993):* Evaluation of hydrolysis and absorption of thermally oxidized olive oil in non-absorbed lipids in the rat. *Ann. Nutr. Metab.* 37: 121.
- Mensink, R.P. and M.B. Katan (1990):* *N. Eng. J. Med.* 323: 439.
- Mensink, R.P. and M.B. Katan (1992):* Effect of dietary fatty acids on serum lipids and lipoproteins. A meta-analysis of 27 trials. *Arterioscler Thromb.* 12: 911.
- National Research Council (1978):* Nutrient requirements of laboratory rat In: Nutrient requirements of laboratory animals, 3rd revised Ed., National Academy of Sciences, Washington, DC. 7 – 37.
- Nestel, P., M. Noakes and B. Belling (1992):* *J. Lipid Res.* 33: 1029.
- Nielsen, H.K., P.A. Finot and R.F. Hurrell (1985):* Reactions of proteins with oxidizing Lipids 2. Influence on protein quality and on the bioavailability of lysine, methionine, cysteine and tryptophan as measured in rat assays. *Br. J. Nutr.* 109: 1653.
- Nolen, G.A., J.C. Alexander and N.R. Artman (1967):* Long-term rat feeding study with used frying fats. *J. Nutrition.* 93: 337.
- Nwanguma, B.C., A.C. Achebe, L.U. Ezeanyika and L.C. Eze (1999):* Toxicity of oxidized fats II : tissue levels of lipids peroxides in rats fed a thermally oxidized corn oil diet. *Food Chem. Toxicol.* 37: 413.
- Owu, D.U., E.E. Osim and P.E. Ebong (1998):* Serum liver enzymes profile of wistar rats following chronic consumption of fresh or oxidized palm oil diets. *Acta Tropica* 69: 65.
- Peters, T.J. (1968):* Proposals for standardization of total protein assays. *J. Clin. Chem.* 14 : 1147.
- Plakunov, I., T.A. Smolarek, L.D. Fischer, J.C. Wiley and W.M. Baird (1987):* Separation by ion- pair high performance liquid chromatography of the glucuronoid, sulfate and glutathion conjugates formed from benzo(a)pyrene in cell cultures, rodents, fish and humans *Carcinogenesis.* 8: 59.

- Purushothama, S., K. Narasimfamurthy, P.L. Raina, and K. Hariharan (1994):* A study of plasma and liver lipid profile of rats fed palm oil or safflower oil along with cholesterol. *Nutr. Res.* 14: 255.
- Reitman, S. and S.A. Frankel (1957):* A colorimetric method for the determination of serum glutamic oxaloacetic and glutamic pyruvic transaminases. *Am. J. Clin. Pathol.* 28: 56.
- Rice, E.E., C.E. Poling, P.E. Mone and W.D. Warner (1960):* A Nutritive evaluation of over heated fats. *J. Am. Oil Chemists' Soc.* 37: 607.
- Richter, K.D., K.D. Mukherjee and N. Weber (1996):* Fat infiltration in liver of rats induced by different dietary plant oils: high oleic – medium oleic – and high petroselinic acid – oils. *Zeitschrift – fur – Ernahrungswissenschaft.* 35: 241.
- Romijn, D., S.A. Wiseman, L.M. Scheek, N.I. de Fouw and A. van Tol. (1998):* A linoleic acid enriched diet increases serum cholesterol esterification by lecithin: cholesterol acyltransferase in meal – fed rats. *Amm – Nutr. Metab.* 42: 244.
- Ruiz–Gutiérrez, V., A. Perez–Espinosa, C.M. Vazquez and C. Santa–Maria (1999):* Effects of dietary fats (fish, olive and high–oleic–acid sunflower oils) on lipid composition and antioxidant enzymes in rat liver. *British Journal of Nutrition.* 82: 233.
- Rustan, A.C., E.N. Christiansen, and C.A. Drevon (1992):* Serum lipids, hepatic glycerolipid metabolism and peroxisomal fatty acid oxidation in rats fed w–3 and w–6 fatty acids. *Biochem. J.* 283: 333.
- Sánchez–Muniz, F.J., F. Cava, J.M. Viejo, S. Bastida, E. Higon and A. Marcos (1997):* Olive oil–fried sardines in the prevention of dietary hypercholesterolemia in rats. Effects on some serum lipids and cell–damage marker enzymes. *Nutrition research.* 16: 111.
- Sánchez–Muniz, F.J., E. Higon, F. Cava and J. M. Vieja (1992):* Prevention of dietary hypercholesterolemia in rats using sunflower– oil fried sardines. Effects on cholesterol and serum enzymes. *J. Agric. Food Chem.* 40: 2252.
- Sánchez – Muniz, S., F.J. Sánchez – muniz, C. Cuesta. (1995):* Decreased food efficiency ratio, growth retardation and changes in liver composition in rats consuming thermoxidized and polymerized sunflower oil used for frying. *Fd Chem. Toxic.* 33: 181.

- Sarkkinen, E.S., M.I. Uusitupa, H. Gylling and T.A. Miettinen (1998):* Fat modified diets influence serum concentration of cholesterol precursors and plant sterols in hypercholesterolemic subjects. *Metabolism.* 47: 744.
- Serag El-Din, M.F. (2001):* Analysis, occurrence and formation of some toxic compounds in some edible oils as the result of cooking and processing. M.Sc. thesis, Fac. Of Home Economics, Minufiya University, shebin El-Kom, Egypt.
- Shibayama, Y. (1992):* Hepatotoxicity of heated and oxygenated corn oil. *Exp. Toxicol. Pathol.* 44: 255.
- Simopoulos, A.P., R.R. kifer, R.E. Martin and S.M. Barlow (1991):* Health effects of W-3 polyunsaturated fatty acids in seafoods. *World Rev. Nutr. Diet.* 66: XVI.
- Spady, D.K., L.A. Woollett and J.M. Dietschy (1993):* Regulation of plasma LDL- cholesterol levels by dietary cholesterol and fatty acids. *Annual Review of Nutrition* 13: 355.
- Steiner, I., M. Fischer and J. Washuett I. (1993):* Analysis of edible oils with special consideration of oils containing polyunsaturated fatty acids. *Fethwiss. Technolo.* 95: 461.
- Terpstra, A.H.M., P. van den Berg, H. Jansen, A.C. Beynen, and A. van Tol (2000):* Decreasing dietary fat saturation lowers HDL-cholesterol and increases hepatic HDL binding in hamsters. *British J. of Nutri.* 83: 151.
- Trautwein, E.A., A.Kunath-Rau, J. Dietrich, S. Drusch and H.F. Erbersdobler (1997):* Effect of dietary fats rich in lauric, myristic, palmitic, oleic or linoleic acid on plasma, hepatic and biliary lipids in cholesterol -fed hamsters. *British J. of Nutri.* 77: 605.
- Trinder, P. (1969):* Determination of glucose in blood using glucose oxidase with an alternative oxygen acceptor. *Annals of Clinical Biochemistry.* 6: 24.
- Tyagi, V.K. and A.K. Vasishtha (1996):* Changes in the characteristics and composition of oils during deep-fat frying. *J. Am. Oil Chem. Soc.* 73: 499.
- Vergroesen, A.J. and M. Crawford (1989):* The Role of Fats in Human Nutrition, Academic Press, New York.
- Warner, K. and T.L. Mounts (1993):* Frying stability of soybean and canola oils with modified fatty acid compositions. *J. Am. Oil Chem. Soc.* 70: 983.

Woyewoda, A.D., S.J. Shaw, P.J. Ke and B.G. Burns (1986): Recommended laboratory methods for assement of fish quality. Canadian Technical Report of Fisheries and Aquatic Sciences. 1448: 65.

Yoshida, H. and J.C. Alexander (1983): Enzymatic hydrolysis of fractionated products from oils thermally oxidized in the laboratory. *Lipids* 18: 402.

Ziemiński, S., B. Panczenko Kresowska, G. Okolska, E. Wielgus-Serafinska and K. Zelakiewicz (1985): Effect of dietary fats on experimental hypertension. *Ann. Nutr. Metab.* 29: 223.

Zilversnit, D.B. and A.K. Davis (1956): Micro determination of plasma phospholipids by chloroacetic precipitation. *J. Lab. Clin. Med.* 35: 155.

Table 2: Performance of rats fed diets containing fresh or used frying oils, for 6 weeks

Frying hours	Diets (oil added as 10 %)					
	Corn	Sunflower	Cottonseed	Palm	Hydrogenated palm	Butter fat
Feed intake (g)						
0	482.0	503.5	502.7	531.8	424.8	414.8
2	461.2	480.9	492.8	475.8	418.4	409.1
4	431.7	470.3	481.8	437.7	359.8	389.6
Weight gain (g)						
0	135.5	165.9	159.2	156.3	112.0	122.1
2	124.6	157.0	149.5	135.8	104.9	116.4
4	109.3	150.1	136.9	118.3	83.9	105.0
Feed conversion index (g diet/ g gain)						
0	3.56	3.03	3.16	3.40	3.79	3.40
2	3.70	3.06	3.30	3.50	3.99	3.51
4	3.95	3.13	3.52	3.70	4.29	3.71

The corn group (1) was considered the control for comparison.

Table 3: Liver, kidneys and heart relative weight (as a body weight) in the different rat groups

Frying Hours	Diets (oil added as 10 %)					
	Corn	Sunflower	Cottonseed	Palm	Hydrogenated palm	Butter fat
Liver						
0	3.69 ± 0.2	2.97±0.33**	3.64±0.27	3.57±0.07	4.02±0.18	3.89±0.04
2	4.48 ± 0.07	3.99±0.19	4.08±0.17	4.21±0.02	4.69±0.33	4.69±0.14
4	4.33 ± 0.12	3.99±0.18	4.22±0.21	4.26±0.24	5.13±0.96**	4.39±0.17
Kidney						
0	0.76±0.03	0.60±0.02	0.68±0.03	0.68±0.02	0.75±0.01	0.70±0.28
2	0.83±0.04	0.69±0.05	0.71±0.01	0.75±0.05	0.85±0.02	0.83±0.03
4	0.83±0.02	0.70±0.04	0.77±0.02	0.78±0.04	0.95±0.04	0.84±0.04
Heart						
0	0.35±0.02	0.29±0.03	0.30±0.02	0.30±0.02	0.32±0.03	0.30±0.01
2	0.33±0.03	0.32±0.01	0.32±0.02	0.31±0.01	0.34±0.04	0.32±0.01
4	0.32±0.01	0.30±0.01	0.31±0.03	0.35±0.03	0.35±0.03	0.33±0.03

** Significant at p < 0.01

Table 4: Effect of fat Source and frying on serum lipids in rats

Frying hours	Diets (Oil added as 10 %)					
	Corn	Sunflower	Cottonseed	Palm	Hydrogenated palm	Butter fat
Triglycerides (mg/dl)						
0	199.6±2.07 ^a	117.9±1.31 ^c	173.1±2.04 ^b	204.0±1.43 ^a	161.0±0.98 ^b	162.2±2.18 ^b
2	198.8±1.19 ^a	109.8±1.37 ^{cd}	86.6±1.20 ^c	93.2±2.08 ^c	158.2±7.45 ^d	188.7±2.73 ^a
4	107.1±1.76 ^a	87.7±1.19 ^b	117.0±1.52 ^c	96.7±0.88 ^{ab}	163.6±2.31 ^c	86.1±1.22 ^b
Phospholipids (mg/dl)						
0	109.4±1.00 ^a	166.1±1.44 ^c	103.8±1.09 ^c	111.1±1.22 ^a	91.2±1.45 ^a	195.2±1.13 ^c
2	78.6±1.09 ^a	148.8±1.04 ^b	180.3±1.30 ^c	170.8±1.38 ^c	75.3±1.31 ^a	101.5±1.15 ^d
4	73.7±1.36 ^a	194.0±0.70 ^c	122.7±1.44 ^{bd}	100.5±0.85 ^b	92.3±1.24 ^a	134.8±1.06 ^c
Total cholesterol (mg/dl)						
0	89.9±1.44 ^a	61.2±1.43 ^{cd}	58.5±0.62 ^c	84.4±1.76 ^a	72.5±1.36 ^{bd}	77.3±1.64 ^b
2	54.8±0.40 ^a	47.8±1.82 ^a	71.7±2.10 ^b	79.1±2.04 ^b	77.8±1.66 ^b	63.3±1.43 ^{bd}
4	79.0±1.19 ^a	67.2±1.08 ^{dc}	59.2±1.18 ^c	84.0±2.00 ^a	97.1±2.18 ^b	64.6±1.51 ^{dc}
HDL - cholesterol (mg/dl)						
0	51.0±0.61 ^a	52.4±0.36 ^a	40.5±0.78 ^b	47.0±1.72 ^a	20.4±0.38 ^c	38.5±0.65 ^b
2	39.5±0.63 ^a	45.8±0.50 ^c	46.3±0.50 ^b	40.5±0.72 ^a	45.8±0.49 ^b	56.6±0.72 ^a
4	29.4±0.63 ^a	51.5±0.49 ^c	34.6±0.83 ^a	27.6±0.72 ^a	36.8±0.50 ^a	36.8±0.72 ^a
LDL - cholesterol (mg/dl)						
0	87.8±0.94 ^a	61.1±1.42 ^{cb}	58.4±0.63 ^c	86.9±0.49 ^a	72.4±1.36 ^b	75.5±0.65 ^b
2	54.8±0.40 ^a	47.3±1.66 ^b	71.7±2.10 ^{cd}	79.1±2.04 ^c	77.8±1.82 ^{cd}	63.3±1.43 ^{cd}
4	79.0±1.19 ^a	57.2±1.08 ^c	59.2±1.18 ^c	84.0±2.00 ^a	97.1±2.18 ^b	64.6±1.51 ^b
Risk factor						
0	1.8±0.01 ^a	1.2±0.16 ^b	1.5±0.04 ^b	1.8±0.08 ^a	3.5±0.22 ^c	2.0±0.04 ^a
2	1.4±0.02 ^a	1.0±0.04 ^c	1.6±0.05 ^c	2.0±0.04 ^a	1.7±0.06 ^b	1.3±0.03 ^a
4	2.6±0.06 ^a	1.3±0.03 ^b	1.7±0.03 ^b	3.0±0.04 ^c	3.6±0.06 ^b	1.8±0.07 ^b

Risk factor = serum total cholesterol /HDL - cholesterol.

Mean values within a row not sharing a common superscript letter were significantly different: a, b, d at p ≤ 0.05; c at p ≤ 0.01.

Table 5: Proteins and enzyme concentration in the serum of rats

Frying hours	Diets (oil added as 10 %)					
	Corn	Sunflower	Cottonseed	Palm	Hydrogenated palm	Butter fat
Total protein (g/d)						
0	13.63 ± 0.10 ^a	14.07 ± 0.09 ^b	12.77 ± 0.10 ^a	15.29 ± 0.13 ^{bd}	18.47 ± 0.09 ^c	16.31 ± 0.09 ^{cd}
2	11.44 ± 0.08 ^a	15.07 ± 0.13 ^b	17.39 ± 0.13 ^{cd}	15.89 ± 0.08 ^{bd}	13.11 ± 0.10 ^a	18.24 ± 0.08 ^c
4	14.45 ± 0.09 ^a	14.93 ± 0.12 ^a	12.56 ± 0.09 ^b	12.22 ± 0.11 ^b	15.96 ± 0.09 ^a	19.10 ± 0.11 ^c
Albumin (g/dl)						
0	3.21 ± 0.12 ^a	4.61 ± 0.23 ^c	3.60 ± 0.24 ^b	3.47 ± 0.20 ^{ab}	3.23 ± 0.19 ^a	4.33 ± 0.15 ^c
2	4.20 ± 0.14 ^a	4.80 ± 0.28 ^b	4.24 ± 0.35 ^a	5.71 ± 0.09 ^{bd}	4.67 ± 0.21 ^b	5.05 ± 0.12 ^b
4	6.59 ± 0.15 ^a	5.90 ± 0.18 ^b	5.82 ± 0.15 ^b	6.36 ± 0.11 ^a	4.70 ± 0.21 ^c	5.46 ± 0.27 ^b
Globulin (g/dl)						
0	9.28 ± 0.08 ^a	9.47 ± 0.21 ^a	9.17 ± 0.29 ^a	11.82 ± 0.33 ^b	15.25 ± 0.18 ^c	11.99 ± 0.12 ^b
2	7.24 ± 0.16 ^a	10.26 ± 0.22 ^b	13.14 ± 0.39 ^c	10.18 ± 0.12 ^b	8.43 ± 0.24 ^a	13.19 ± 0.19 ^c
4	7.86 ± 0.21 ^a	9.03 ± 0.24 ^b	6.74 ± 0.15 ^c	5.86 ± 0.16 ^b	11.26 ± 0.17 ^a	13.64 ± 0.26 ^c
A/G ratio						
0	0.31 ± 0.01 ^a	0.49 ± 0.02 ^b	0.39 ± 0.03 ^a	0.30 ± 0.04 ^a	0.21 ± 0.02 ^{bd}	0.36 ± 0.01 ^{cd}
2	0.58 ± 0.03 ^a	0.53 ± 0.08 ^c	0.33 ± 0.03 ^b	0.55 ± 0.01 ^a	0.50 ± 0.04 ^d	0.37 ± 0.01 ^b
4	0.82 ± 0.04 ^a	0.53 ± 0.05 ^c	0.87 ± 0.04 ^a	0.96 ± 0.12 ^a	0.42 ± 0.03 ^c	0.40 ± 0.03 ^c
Enzymes (U/l)						
AST						
0	36.54 ± 0.71 ^a	40.02 ± 1.16 ^{ab}	51.46 ± 0.78 ^b	73.20 ± 0.81 ^c	67.8 ± 1.18 ^c	63.03 ± 0.20 ^c
2	60.12 ± 0.76 ^a	45.50 ± 1.19 ^b	52.28 ± 1.58 ^c	87.92 ± 0.30 ^c	85.42 ± 1.11 ^{bd}	75.28 ± 1.71 ^{bd}
4	58.24 ± 1.28 ^a	33.72 ± 0.88 ^d	43.56 ± 2.14 ^{bd}	76.16 ± 0.83 ^c	85.22 ± 0.65 ^c	57.30 ± 1.00 ^a
ALT						
0	14.48 ± 0.81 ^a	9.56 ± 0.44 ^b	10.88 ± 0.55 ^b	13.64 ± 0.40 ^a	19.2 ± 0.48 ^c	8.78 ± 0.72 ^c
2	8.62 ± 0.68 ^a	9.22 ± 0.69 ^{bd}	10.48 ± 0.42 ^{bd}	14.62 ± 0.76 ^c	20.84 ± 0.23 ^c	14.62 ± 0.41 ^c
4	16.20 ± 0.47 ^a	8.34 ± 0.44 ^c	8.18 ± 0.56 ^c	12.38 ± 0.61 ^c	12.38 ± 0.49 ^c	8.44 ± 0.36 ^c

A/G ratio = the ratio of albumin to globulin in blood serum .

AST = Aspartate aminotransferase ALT = Alanine aminotransferase .

Mean values within a row not sharing a common superscript letter were significantly different: a ,b ,d at p ≤ 0.05 ; c at p ≤ 0.01.

Table 6: Chemical characteristics and carcinogen concentration in fresh and used frying oils and butter fat

Analysis	Type of oil and frying hours																	
	Corn oil			Sunflower oil			Cottonseed oil			Palm oil			Hydrogenated palm oil			Butter fat		
	0	2	4	0	2	4	0	2	4	0	2	4	0	2	4	0	2	4
Acid value (mg KOH/g)	0.76	0.80	0.85	0.79	0.76	0.82	0.76	0.79	0.84	0.73	0.76	0.79	0.75	0.78	0.80	0.72	0.74	0.74
	±	±	±	±	±	±	±	±	±	±	±	±	±	±	±	±	±	±
	0.11	0.10	0.20	0.07	0.19	0.13	0.11	0.05	0.17	0.10	0.14	0.12	0.10	0.13	0.09	0.08	0.12	0.16
Peroxide value (meq/Kg)	8.01	9.11	9.93	7.81	8.31	8.79	7.97	9.03	9.91	7.63	8.30	8.52	7.72	8.52	8.57	7.52	7.82	8.04
	±	±	±	±	±	±	±	±	±	±	±	±	±	±	±	±	±	±
	1.08	0.75	0.70	0.98	1.11	8.89	1.03	1.14	0.85	1.02	0.98	1.15	0.90	0.87	1.12	0.97	0.85	0.91
Iodine value (Hanus solution)	116.50	117.20	114.63	115.83	114.25	114.87	113.68	113.14	111.28	114.87	114.61	114.98	96.06	96.06	95.34	94.58	94.36	94.18
	±	±	±	±	±	±	±	±	±	±	±	±	±	±	±	±	±	±
	3.21	2.89	3.85	2.17	3.12	3.55	2.49	2.86	2.58	2.96	3.17	2.89	2.89	2.55	3.27	2.38	2.49	3.09
Saponification value (mg KOH/g)	125.20	125.91	136.01	122.91	123.51	130.71	125.51	124.61	134.07	122.00	123.91	127.25	123.25	123.46	127.38	122.00	122.60	126.51
	±	±	±	±	±	±	±	±	±	±	±	±	±	±	±	±	±	±
	2.17	3.21	3.58	3.72	4.00	4.12	2.72	3.20	3.42	2.95	3.11	3.51	2.89	3.15	4.20	2.24	2.87	3.18
Malonaldehyde (mg/kg)	6.97	7.43	8.56	6.72	7.40	8.06	6.74	7.41	8.32	9.51	6.97	7.42	6.53	6.86	7.31	6.50	6.82	7.20
	±	±	±	±	±	±	±	±	±	±	±	±	±	±	±	±	±	±
	0.98	0.85	1.01	0.90	0.54	1.14	0.74	0.95	0.99	1.14	0.77	0.49	0.81	0.73	1.10	1.07	1.11	0.96
Benzo(a) pyrene (mg/kg)	0.98	1.03	1.19	0.91	1.01	1.19	0.90	1.07	1.17	0.83	0.88	0.91	0.76	0.89	0.94	0.72	0.33	0.88
	±	±	±	±	±	±	±	±	±	±	±	±	±	±	±	±	±	±
	0.16	0.17	0.21	0.20	0.14	0.21	0.22	0.15	0.23	0.14	0.20	0.22	0.11	0.19	0.10	0.13	0.20	0.19

Table 7: Fatty acid composition of fresh and used frying oils and fats (g/100 g total fatty acid)

Fatty acid	Type of oil and frying hours																	
	Corn oil			Sunflower oil			Cottonseed oil			Palm oil			Hydrogenated palm oil			Butter fat		
	0	2	4	0	2	4	0	2	4	0	2	4	0	2	4	0	2	4
Saturated																		
12:0	-	0.01	0.52	-	0.32	0.58	-	-	0.50	0.06	0.24	0.83	-	0.14	1.33	-	0.17	0.71
14:0	0.09	1.01	2.08	1.07	1.51	2.56	0.91	1.77	1.83	0.72	0.79	2.96	1.29	2.15	3.46	11.94	1.26	3.63
16:0	10.31	11.93	18.98	8.79	13.23	16.30	7.80	15.44	15.95	14.14	14.29	16.65	24.45	19.77	17.02	13.79	14.08	21.37
18:0	0.23	1.59	3.12	1.29	3.01	9.08	3.90	5.15	5.81	3.06	3.02	5.55	10.74	12.00	13.96	1.92	2.18	4.23
20:0	0.18	0.14	-	1.29	0.86	0.47	Traces	Traces	-	0.30	0.24	-	1.35	1.33	0.80	0.60	0.59	0.50
22:0	0.14	0.07	-	0.64	-	-	Traces	Traces	-	0.24	0.16	0.09	0.78	0.86	-	0.46	0.50	0.30
Total	10.95	14.75	24.70	13.08	18.93	28.99	12.61	22.35	24.09	18.52	18.74	26.08	38.61	36.25	36.57	28.71	18.78	30.74
Unsaturated																		
12:0	0.14	0.29	0.26	1.61	0.97	0.35	5.20	5.88	8.30	0.48	0.56	0.65	2.72	4.44	2.93	0.66	0.75	0.91
14:0	24.75	20.12	19.23	28.40	25.80	27.00	22.37	20.59	18.44	24.87	26.35	27.75	15.20	16.33	13.30	21.02	25.99	18.75
16:0	64.16	64.84	55.81	56.91	54.30	43.66	59.82	51.18	49.17	56.13	54.35	45.52	43.47	42.98	47.20	49.61	54.48	49.60
18:0	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
20:0	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
22:0	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Total	89.05	85.25	75.30	86.92	81.08	71.01	87.39	77.65	75.91	81.48	81.26	73.92	61.39	63.75	63.43	71.29	81.22	69.26
U/S	8.13	5.78	3.05	6.65	4.28	2.45	6.93	3.47	3.15	4.40	4.34	2.83	1.59	1.76	1.73	2.48	4.32	2.25
P/S	5.86	4.40	2.26	4.35	2.87	1.51	4.74	2.29	2.04	3.03	2.90	1.75	1.13	1.19	1.29	1.73	2.90	1.61

U/S = Unsaturated / saturated fatty acid ratio

P/S = Polyunsaturated / saturated fatty acid ratio

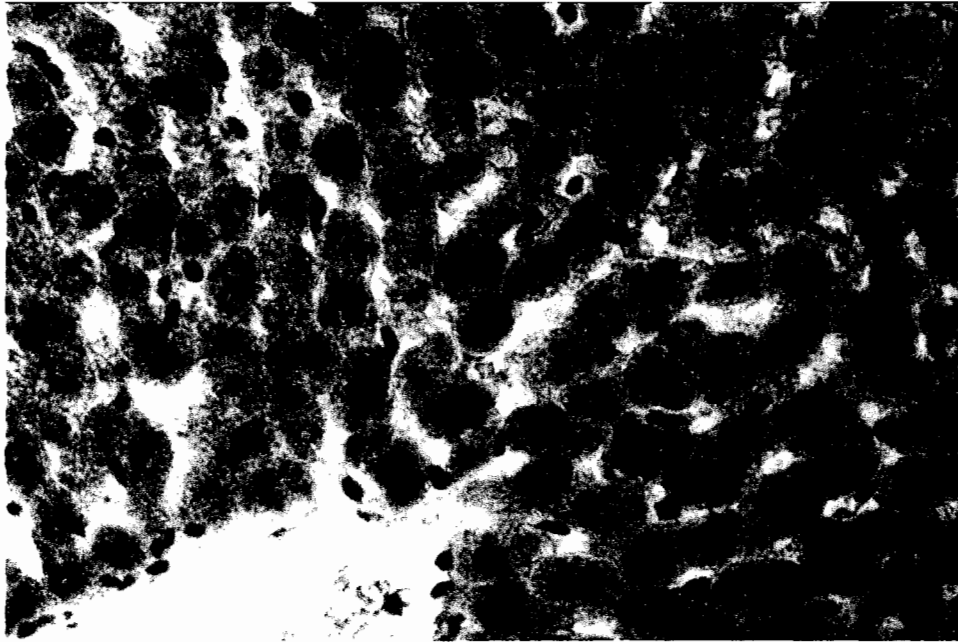


Fig. 1: Liver of rat fed on fresh corn oil showed dilated central vein and sinusoids with proliferation active of kupffer cells.

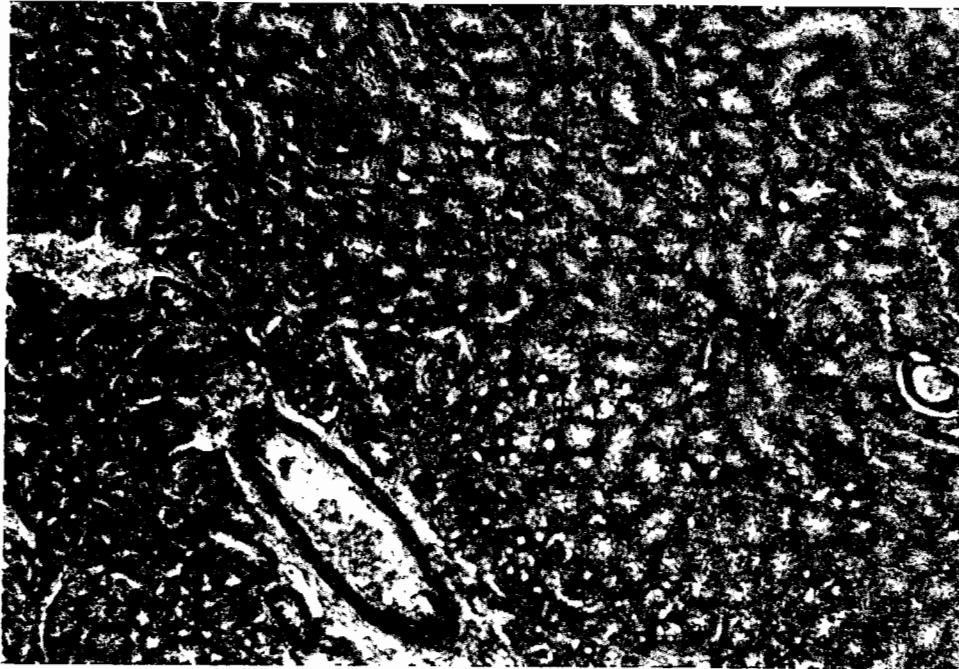


Fig. 2: Liver of rat fed on 2h fried hydrogenated palm oil showed leucocytic inflammatory cellular infiltration in the portal area associated with distended and newly formed bile ducts.

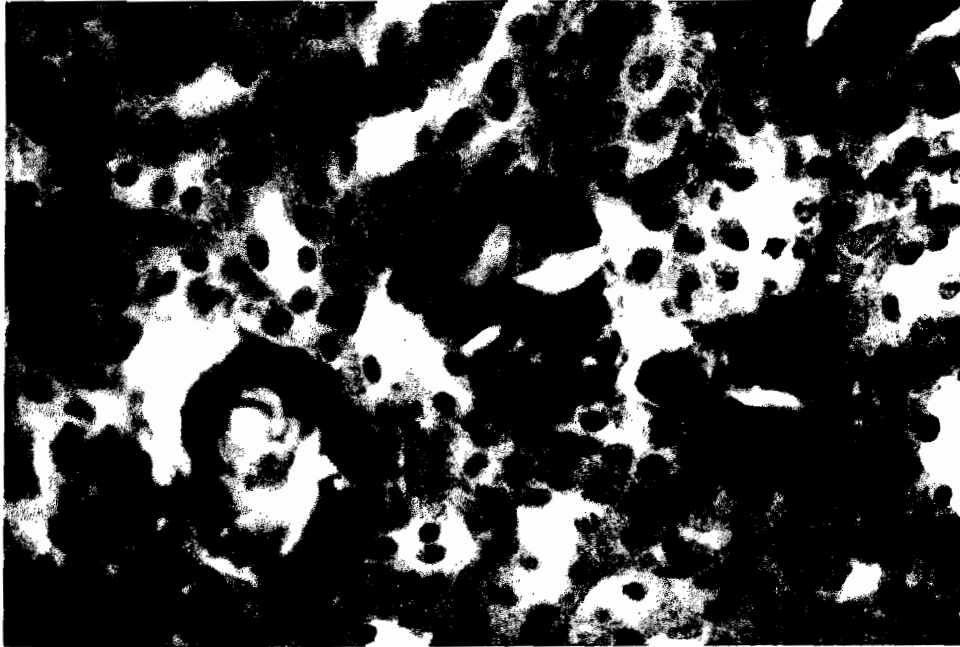


Fig. 3: Kidney of rat fed on 4h. fried hydrogenated palm oil showed basophilic casts in the lumen of the renal tubules in medullary portion with dilatation of inter tubular blood capillaries.

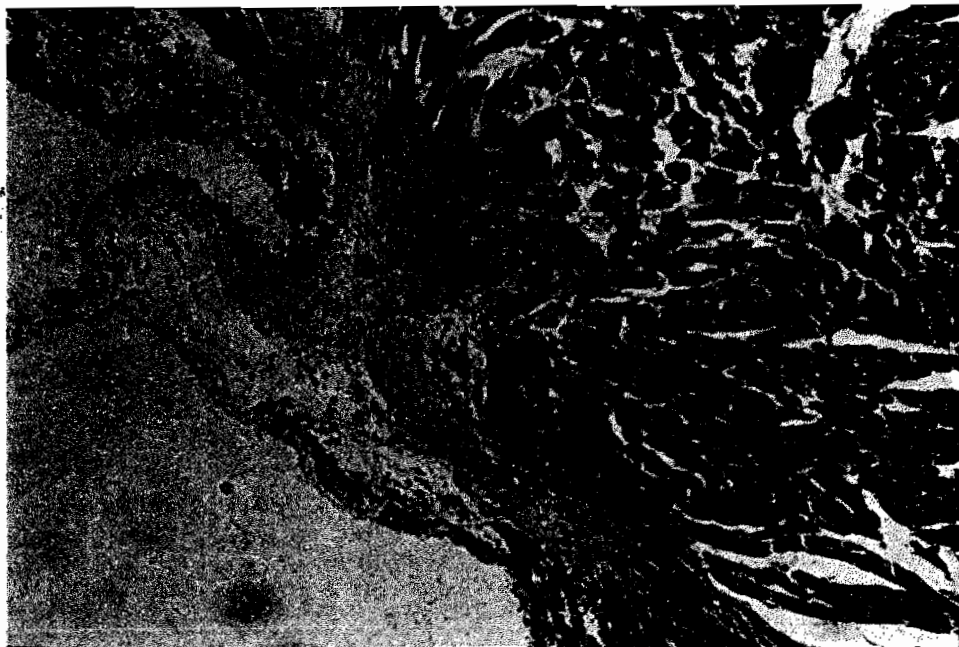


Fig. 4: Heart of rat fed on 4h. fried hydrogenated palm oil showed oedema with inflammatory cellular infiltration in the endocardium.