ω-3 Rich *Tetracarpidum conophorum* Oil Exhibits Better Prevention Effects for Cardiovascular Risk Factors than Corn Oil in Adult of Albinos Wistar Male Rats

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### Abstract

Cardiovascular diseases are serious pathologies that affect an increasing number of people. Several preventive measures are generally used, including supplementing of oils in foods. Our objective was to compare the effects of *Tetracarpidum conophorum* oil (TC) and corn oil (CO) on serum lipid profiles of normal male rats. 42 Wistar rats were divided into 7 groups. Diets included TC oil (groups TC5, TC10 and TC20) and corn oil (groups CO5, CO10 and CO20) in proportions of 5%, 10% and 20%, with a control group (T). After 5 weeks of feeding, several parameters were measured during and after the study, including body weight, food intake and organ weights (kidney, liver and fat). Lipid profiles (total cholesterol, TG, HDL and LDL), glucose and protein levels were measured in the serum. The increase in body mass was inversely proportional to the amount of oil in the food. The decrease in body mass and adiposomatic index of group TC10 was significant (p < 0.05) compared with the other groups. The lowest glycaemia (64.17 ± 5.14 mg/dl) was noted with the diet containing 20% TC oil. A significant reduction in total cholesterol, LDL fraction and blood triglycerides was observed in the groups supplemented with TC and corn oils compared to controls. Results were also more beneficial for the TC10 group. HDL-cholesterol levels were significantly higher (p < 0.05) in the oil-supplemented groups than in the control group. Castelli’s risk indices decrease significantly (p < 0.05) with increasing oil content for TC. The oils had no impact on blood protein contents. One can...
conclude that a diet containing 10% crude oil from TC kernels could prevent or alleviate cardiovascular diseases and glycemia.

**Keywords**

*Tetracarpidium conophorum* Oil, ω-3, Corn Oil, Lipid Profiles, Glycemia

**1. Introduction**

Cardiovascular diseases are the leading cause of death worldwide [1]. They are a growing problem in most developing regions of the world. The most common cause is dyslipidemia, which contributes to atherosclerosis linked to lipid metabolism [2] [3]. Abnormalities in lipid metabolism, including elevated triglyceride levels, total cholesterol, LDL cholesterol fraction, and simultaneously decreased HDL cholesterol levels contribute to the atherosclerosis process [4] [5]. Several treatments are available to manage dyslipidemia and prevent the cardiovascular diseases in people at risk. It is accepted that the key treatment lies mainly in dietary measures aimed at improving the lipid profile by reducing saturated fats, trans fats, carbohydrates and animal proteins, and ensuring adequate consumption of monounsaturated, polyunsaturated fatty acids (ω-3 and ω-6), plant proteins and dietary fibres [6]-[8].

ω-3 fatty acids, along with ω-6, belong to the group of polyunsaturated fatty acids (PUFAs). The main representatives are linoleic acid (ω-6) and α-linolenic acid (ω-3). These two groups of acids influence the body’s metabolic functions. A number of scientific studies have shown that fatty acids in seed oils, such as sunflower and canola oils (rich in ω-6 fatty acids), protect against cardiovascular disease and cancer [9]-[12]. The investigations of Drouin-Chartier et al. [13] have demonstrated that ω-6 fatty acids have lower LDL cholesterol compared to saturated fatty acids.

Moreover, a number of studies over the years have shown that ω-3 polyunsaturated fatty acids found in seafood (sardines, trout, cod livers, eels, etc.) have beneficial effects on reducing the risk of cardiovascular disease [8] [9] [14]-[16]. Diet rich in ω-3 fats, more specifically eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), prevents cardiovascular disease [17]. Sagara et al. [18] showed that 2 g of DHA per day for five weeks improved blood pressure and lipid profiles in men with hypertension and hypercholesterolemia. The beneficial effects of ω-3 of animal origin on the management of dyslipidemia do not neglect the merit of studying the importance of ω-3 of plant origin in the same direction. For example, findings on flaxseed [16], walnut [19] and rapeseed rich in linolenic acid have proved their beneficial effect in the management of dyslipidemia. These fatty acids have the capacity to improve the lipid levels including triglycerides, cholesterol (total, LDL and HDL) [20] and inhibit atherosclerosis and thrombosis [21] [22]. These ω-3 fatty acids mainly reduce total
and LDL cholesterols and triglyceride levels, while increasing HDL cholesterol levels [8] [23].

In Cameroon, there are several species of unconventional oilseeds, including *Tetracarpidium conophorum* of the Euphorbiaceae family. Generally, eaten boiled or roasted [24]. Aqueous extract of *T. conophorum* kernels could have hypocholesterolemic and steriodogenic potential [25]. These almonds are a source of invisible lipids (50.6 g/100 almonds) [26]-[28]. Oil of these almonds contains mainly α-linolenic acid (70% of total fatty acids), one of the ω-3 family. From a nutritional point of view, research works of Tchankou Leudeu et al. [29] on young male rats show that *T. conophorum* oil incorporated at 5% in the diet is of interest in reducing the risk of vascular disease. The aim of this study was to examine the effect of *T. conophorum* oil incorporated at different concentrations (5%, 10% and 20%) on the lipid profile of adult male rats, and compare it to one of the main sources of ω-6, which is corn oil commonly used in the diet.

2. Materials and Methods

2.1. Oils

Oil from *T. conophorum* kernels was obtained by cold pressing using a KOMET screw oil expeller DD85G, number 200666 manufactured in 1991 (Germany) [30]. Refined corn oil (Lesieur, France) was purchased at a local market. To avoid oxidation, the oils were packaged in dark bottles and kept in the refrigerator until use.

**Fatty Acid Profile of Extracted Oils**

Fatty acids were determined by the analytical methods described by Focant et al. [31]. Fatty acids were converted to fatty acid methyl esters (FAMEs) before being analyzed. The FAMEs were prepared using 10 mL of 1M sodium hydroxide in methanol. Then, 4 mL of 1.2M hydrochloric acid in methanol was added to the mixture. After extraction of methyl ester, the chromatograph Thermo Finnigan, type TRACE GC was used to identify fatty acids (Milan, Italy). The capillary column used was RESTEK Rt-2560 (100 m length, 0.25 mm internal diameter, 0.20 µm film thickness) (Supelco, Bellefonte, PA, USA). Gas chromatography conditions were: a flow rate of 1 mL/min with an initial temperature of 140˚C held for 5 min. The column temperature was then increased to 250˚C at a rate of 2˚C/min, and then held at 250˚C for 15 min. Fatty acid peaks were identified using pure methyl ester standards (Larodan, Belgium).

2.2. Animal Experiments

Healthy male Wistar rats (20 - 21 weeks old, 300 - 310 g weight) were provided by the animal house of the Bioprocess Laboratory (IUT, University of Ngaoundere, Cameroon). The rats were acclimatized for a 1 week in a standard environmental atmosphere (at a 12-hours light/dark cycle, 25 ± 2˚C temperature, 50 ± 5% humidity) with a normal diet (Table 1) and water *ad libitum* in a simple cages.
Table 1. Composition of diets (g/g diet).

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Standard (T)</th>
<th>TC5</th>
<th>TC10</th>
<th>TC20</th>
<th>CO5</th>
<th>CO10</th>
<th>CO20</th>
</tr>
</thead>
<tbody>
<tr>
<td>Delipidated fish meal</td>
<td>24</td>
<td>24</td>
<td>24</td>
<td>24</td>
<td>24</td>
<td>24</td>
<td>24</td>
</tr>
<tr>
<td>Corn starch</td>
<td>50</td>
<td>48</td>
<td>43</td>
<td>33</td>
<td>48</td>
<td>43</td>
<td>33</td>
</tr>
<tr>
<td>DL-methionine</td>
<td>0.3</td>
<td>0.3</td>
<td>0.3</td>
<td>0.3</td>
<td>0.3</td>
<td>0.3</td>
<td>0.3</td>
</tr>
<tr>
<td>Sucrose</td>
<td>12</td>
<td>12</td>
<td>12</td>
<td>12</td>
<td>12</td>
<td>12</td>
<td>12</td>
</tr>
<tr>
<td>Cellulose</td>
<td>5</td>
<td>5</td>
<td>5</td>
<td>5</td>
<td>5</td>
<td>5</td>
<td>5</td>
</tr>
<tr>
<td>Choline chloride</td>
<td>0.2</td>
<td>0.2</td>
<td>0.2</td>
<td>0.2</td>
<td>0.2</td>
<td>0.2</td>
<td>0.2</td>
</tr>
<tr>
<td>T.conophorum Oil</td>
<td>0</td>
<td>5</td>
<td>10</td>
<td>20</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Corn oil</td>
<td>0</td>
<td>0</td>
<td>10</td>
<td>5</td>
<td>10</td>
<td>20</td>
<td></td>
</tr>
<tr>
<td>Vitamin mix</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>5</td>
<td>3.5</td>
<td>3.5</td>
</tr>
<tr>
<td>Mineral mix</td>
<td>3.5</td>
<td>3.5</td>
<td>3.5</td>
<td>3.5</td>
<td>3.5</td>
<td>3.5</td>
<td>3.5</td>
</tr>
</tbody>
</table>

After this period, the rats were randomly assigned into seven groups, 6 rats per group. The TC5, TC10 and TC20 groups consumed respectively 5%, 10% and 20% of *T. conophorum* oil while the CO5, CO10 and CO20 groups consumed 5%, 10% and 20% of corn oil respectively. One control group (T) received a standard rat diet. Animals were housed individually in the cages. Body weight was performed once a week, water and food consumption was estimated daily. Diets were prepared in the Biopress laboratory and details of their compositions are presented in Table 1. Formulations were based on the modified AIN-93 protocol [32]. This study was approved by the institutional animal research committee of the Faculty of Sciences of the University of Ngaoundere.

**Growth Performances**

The following parameters were calculated:

- **Food consumption** was recorded throughout the experimental period and the average daily food consumption was determined.

- **Body weight** of each rat was recorded weekly throughout the experimental period.

- **% weight gain** = [(final weight - initial weight) × 100/initial weight].

- **Consumption Index (CI)** is the ratio that measures food consumption/body weight gain.

**2.3. Blood and Sample Collections**

At the end of the experiment, animals were fasted for 12 hours with free access to drinking water. The rats were then anesthetized using Diazepam (10 mg/Kg) according to the recommendations for the anaesthesia of experimental animals. Blood samples were collected from the caudal vein and stored in dry tubes and stabilized for 15 to 30 minutes to ensure that it could clotted at room tempera-
ture (25°C). Serum was prepared from blood by centrifugation at 3000 rpm for 10 min. This serum was used to carry out the dosages. The liver, heart, kidneys, and epididymal and abdominal fats were removed and weighed to calculate the hepatosomatic index, viscerosomatic indices, and adiposity index, respectively.

2.3.1. Biochemical Analyses of Serum Lipid Profiles
The total cholesterol (TC), high-density lipoprotein cholesterol (HDL-C) and triglyceride levels in the serum were performed using kit assay methods (FORTRESS, Northern Ireland) as per the manufacturer’s instruction. Serum low density lipoprotein cholesterol (LDL-C) concentration and very low density lipoprotein cholesterol (VLDL-C) were determined using Friedwald formula, where LDL-C = TC − (HDL-C + VLDL-C) and VLDL-C = TG/5 [33]. Castelli risk indices I (CRI-I) and Castelli risk indices II (CRI-II) were calculated using the Castelli formula, where CRI-I = LDL-C ÷ HDL-C and CRII = TC ÷ HDL-C [34].

2.3.2. Estimation of Fasting Blood Glucose
Blood glucose level was evaluated directly after the sacrifice of the rats on an automatic biochemical analyzer using enzymatic colorimetric tests (FORTRESS, Northern Ireland).

2.3.3. Estimation of Serum Total Protein
The colorimetric method described by Gornall et al. [35] was used to estimate the protein content in the blood. Peptide bonds in proteins react with Cu²⁺ in alkaline solution to form a colored complex which absorbance, proportional to the protein concentration in the specimen at 550 nm. Biuret reagent contains sodium potassium tartrate, which complexes cupric ions and maintains their solubility in alkaline solution [36].

2.4. Statistical Analyses
Results were expressed as means ± standard deviation. For each group, the result obtained was the mean for 6 rats. All results were analysed using a one-way analysis of variance. Duncan’s Multiple Range test was performed to evaluate differences between groups. Differences between means were considered to be significant at p < 0.05.

3. Results
3.1. Fatty Acids Composition of Oils
In Table 2, fatty acids composition of the two oils is presented. Corn oil contains nearly 56.13 ± 0.30% linoleic acid and only 1.07 ± 0.02% of α-linolenic acid, compared to T. conophorum oil with nearly 70% α-linolenic acid and 10.56 ± 0.18 of linoleic acid. The ω-3/ω-6 ratio of T. conophorum oil is 6.52 while this ratio is 0.01 for corn oil. Both oils are rich in essential fatty acids; which is a great advantage in diet formulations the risks of cardiovascular diseases.
Table 2. Fatty acid composition of *T. conophorum* almond oils and corn oil (%/100 g of oil).

<table>
<thead>
<tr>
<th>Fatty acids</th>
<th><em>T. conophorum</em></th>
<th>Corn oil</th>
</tr>
</thead>
<tbody>
<tr>
<td>C14:0</td>
<td>Myristic acid</td>
<td>0.02 ± 0.00</td>
</tr>
<tr>
<td>C16:0</td>
<td>Palmitic acid</td>
<td>1.91 ± 0.02</td>
</tr>
<tr>
<td>C16:1C9</td>
<td>Palmitoleic acid</td>
<td>0.01 ± 0.00</td>
</tr>
<tr>
<td>C18:0</td>
<td>Stearic acid</td>
<td>3.67 ± 0.10</td>
</tr>
<tr>
<td>C18:1C9</td>
<td>Oleic acid</td>
<td>14.35 ± 0.15</td>
</tr>
<tr>
<td>C18:1C11</td>
<td></td>
<td>0.42 ± 0.01</td>
</tr>
<tr>
<td>C18:2C9C12</td>
<td>Linoleic acid</td>
<td>10.56 ± 0.18</td>
</tr>
<tr>
<td>C20:0</td>
<td>Arachidic acid</td>
<td>0.16 ± 0.00</td>
</tr>
<tr>
<td>C18:3C9C12C15</td>
<td>α-linolenic acid</td>
<td>68.90 ± 0.17</td>
</tr>
<tr>
<td>∑ Saturated (%)</td>
<td>5.76</td>
<td>13.7</td>
</tr>
<tr>
<td>∑ Monounsaturated (%)</td>
<td>14.78</td>
<td>29.1</td>
</tr>
<tr>
<td>∑ Polyunsaturated (%)</td>
<td>79.46</td>
<td>57.2</td>
</tr>
<tr>
<td>C18:2/C16:0</td>
<td></td>
<td>5.77</td>
</tr>
<tr>
<td>ω-3/ω-6</td>
<td></td>
<td>6.52</td>
</tr>
</tbody>
</table>

3.2. Evolution of Body Mass

Figure 1 shows the evolution of body mass (in g) of rats fed with the different diets over a 5-week period. In the first two weeks, there was no change in body masses compared to starting day. From week 3 to week 5, the increase of body mass was inversely proportional to the amount of oil in the feed. Thus, groups T, CO5 and TC5 evolved together, as do groups CO10, TC10 and CO20. The body mass of the TC20 group decreased from the first week till the end of the experiment compared to the other groups, and the difference between this group and the other groups was significant (p < 0.05).

Figure 1. Evolution of rats body masses. T: Control group; CO5: Group of 5% of corn oil; CO10: Group of 10% of corn oil; CO20: Group of 20% of corn oil; TC5: Group of 5% of *T. conophorum* oil; TC10: Group of 10% of *T. conophorum* oil; TC20: Group of 20% of *T. conophorum* oil. Means ± SD (n = 6).
3.3. Effect of the Two Oils on Food Intake, Consumption Index and Hydration Index with Respect to the Percentage

Daily feed intakes after 5 weeks of feeding are summarized in Table 3. Irrespective of the type of oil, feed intake decreased with increasing oil content (p < 0.05). One can agree that fats slow down the digestion process and leave a prolonged feeling of satiety in the stomach. Thus, the quantity of oil consumed per rat per day is 0.75 for the 5% diets compared to 1.35 for the 10% diets and 2.5 for the 20% diets.

The consumption index makes it possible to determine feed consumption in relation to growth rate. This index is low for TC diets compared to CO diets.

Overall, water consumption is proportional to the amount of oil, so the more the oil content of a feed, the more the water consumption. Thus, the water consumption is 6.87, 5.30, 8.08 and 8.55 respectively for regimes T, TC5, TC10 and TC20. Water consumption therefore depends on the palatability stimulated by the type of oil.

3.4. Hepatosomatic (HSI), Adiposomatic (ASI) and Viscerosomatic Indices of Rats on Different Diets

Hepatosomatic, adiposomatic and viscerosomatic indices are determined to better appreciate the effect of the quantity of different oils on organ development.

Ratios of liver weight to body weight in rats (hepatosomatic index) (HSI) of the seven study groups are shown in Table 4. Data analysis shows that there was no significant difference (p < 0.05) between the control group and the other groups, irrespective of oil type and quantity.

It was found that the adiposomatic index (ASI) decreased with increasing oil quantity for the T. conophorum diet groups, but the difference was not significant (p < 0.05). Furthermore, the lowest values were observed for the T. conophorum diet (p < 0.05) compared to the corn oil diet, whatever the oil percentage.

Table 3. Effect of variation of T. conophorum and corn oil on food intake consumption index, quantity of oil consumed and hydration index of rats fed with different diets.

<table>
<thead>
<tr>
<th>Group</th>
<th>Food intake/g/day/rat</th>
<th>Quantity of oil consumed/g/day/rat</th>
<th>Consumption index</th>
<th>Hydration index</th>
</tr>
</thead>
<tbody>
<tr>
<td>T</td>
<td>17.00 ± 0.70a</td>
<td>0.00 ± 0.00a</td>
<td>2.58 ± 0.20b</td>
<td>6.87 ± 0.47c</td>
</tr>
<tr>
<td>CO5</td>
<td>15.49 ± 1.60a</td>
<td>0.75 ± 0.08c</td>
<td>3.10 ± 0.31a</td>
<td>5.40 ± 0.55c</td>
</tr>
<tr>
<td>CO10</td>
<td>13.42 ± 0.86ab</td>
<td>1.35 ± 0.18b</td>
<td>2.21 ± 0.31bc</td>
<td>7.23 ± 0.80bc</td>
</tr>
<tr>
<td>CO20</td>
<td>12.85 ± 0.89b</td>
<td>2.50 ± 0.23a</td>
<td>2.56 ± 0.18b</td>
<td>10.10 ± 1.60c</td>
</tr>
<tr>
<td>TC5</td>
<td>15.00 ± 1.42a</td>
<td>0.75 ± 0.07c</td>
<td>2.88 ± 0.20a</td>
<td>5.30 ± 0.66c</td>
</tr>
<tr>
<td>TC10</td>
<td>13.20 ± 0.42b</td>
<td>1.35 ± 0.12b</td>
<td>1.74 ± 0.31c</td>
<td>8.05 ± 0.80bc</td>
</tr>
<tr>
<td>TC20</td>
<td>12.62 ± 0.82b</td>
<td>2.50 ± 0.21a</td>
<td>-3.54 ± 0.36d</td>
<td>8.55 ± 1.00bc</td>
</tr>
</tbody>
</table>

Note: Means ± SD (n = 6) followed by different letters are significantly different (p < 0.05) as determined by Duncan’s multiple range test. T: Control group; CO5: Group of 5% of corn oil; CO10: Group of 10% of corn oil; CO20: Group of 20% of corn oil; TC5: Group of 5% of T. conophorum oil; TC10: Group of 10% of T. conophorum oil; TC20: Group of 20% of T. conophorum oil.
Table 4. Effect of variation *T. conophorum* and corn oil on adiposity, hepatosomatic and viscerosomatic indices of rats fed with different diets.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Groups</th>
<th>HSI</th>
<th>ASI</th>
<th>Heart masses/body masses</th>
<th>Kidney masses/body masses</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Heart masses/body masses</td>
<td>T</td>
<td>3.14 ± 0.42&lt;sup&gt;ab&lt;/sup&gt;</td>
<td>2.24 ± 0.29&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.33 ± 0.90&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.66 ± 0.06&lt;sup&gt;ab&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>CO5</td>
<td>2.82 ± 0.12&lt;sup&gt;a&lt;/sup&gt;</td>
<td>2.32 ± 0.11&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.29 ± 0.02&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.65 ± 0.02&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>CO10</td>
<td>3.26 ± 0.37&lt;sup&gt;b&lt;/sup&gt;</td>
<td>3.40 ± 0.27&lt;sup&gt;c&lt;/sup&gt;</td>
<td>0.31 ± 0.30&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.71 ± 0.04&lt;sup&gt;ab&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>CO20</td>
<td>3.20 ± 0.36&lt;sup&gt;ab&lt;/sup&gt;</td>
<td>3.06 ± 0.29&lt;sup&gt;c&lt;/sup&gt;</td>
<td>0.34 ± 0.50&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.69 ± 0.08&lt;sup&gt;ab&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>TC5</td>
<td>3.16 ± 0.36&lt;sup&gt;ab&lt;/sup&gt;</td>
<td>1.81 ± 0.38&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.29 ± 0.40&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.74 ± 0.07&lt;sup&gt;bc&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>TC10</td>
<td>3.08 ± 0.27&lt;sup&gt;ab&lt;/sup&gt;</td>
<td>1.89 ± 0.23&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.31 ± 0.40&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.81 ± 0.06&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>TC20</td>
<td>3.25 ± 0.18&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.74 ± 0.16&lt;sup&gt;c&lt;/sup&gt;</td>
<td>0.32 ± 0.10&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.81 ± 0.09&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Note: Means ± SD (n = 6) followed by different letters in the same column are significantly different (p < 0.05) as determined by Duncan’s multiple range test. T: Control group; CO5: Group of 5% of corn oil; CO10: Group of 10% of corn oil; CO20: Group of 20% of corn oil; TC5: Group of 5% of *T. conophorum* oil; TC10: Group of 10% of *T. conophorum* oil; TC20: Group of 20% of *T. conophorum* oil.

These indices vary from 1.7 - 1.8 for the TC groups compared to 2.3 - 3.4 for the CO groups.

There was no significant difference between the different groups studied in terms of viscerosomatic indices (heart and kidney weight to body weight ratios).

3.5. Biochemical Serum Parameters

3.5.1. Levels of Serum Glucose in Different Groups

Glycemia in this study provides information on the influence of lipids consumed on glucose metabolism. Regardless of the type of oil, glucose levels in fasting rats decreased as the amount of oil in the food increased. However, the decrease is more marked with diets rich in *T. conophorum* oil, but significant for both CO10 and TC10 groups (Figure 2).

![Figure 2](image.png)

**Figure 2.** Serum glucose levels (mg/dl) in rats fed with CO and TC oils with respect to the concentration. Means ± SD (n = 6). CO: Corn oil; TC: *T. conophorum* oil.
3.5.2. Levels of Total Cholesterol in Different Groups
The total cholesterol in the blood of rats was determined and the results are presented in Figure 3. At 0% oil, total cholesterol was 80 ± 3 mg/dl. The addition of oils in the diets resulted in a decrease in serum total cholesterol (p < 0.05) depending on the oil type and the concentration. The drop was −18 mg/dL, −15 mg/dL, −46 and −45 mg/dL respectively for the CO10, CO20, TC10 and TC20 groups. There was no significant difference between TC10 and TC20.

![Figure 3. Serum total cholesterol levels (mg/dl) in rats fed with CO, TC with respect to the oil percentage. Means ± SD (n = 6). CO: Corn oil; TC: T. conophorum oil.](image)

3.5.3. Levels of LDL and HDL Cholesterols in Different Groups
The levels of LDL and HDL cholesterols were evaluated in the serum of rats fed with CO and TC oils, and the results are illustrated on Figure 4.

There was a significant drop in serum LDL-C levels at the different treatment levels compared to group T (p < 0.05) (Figure 4(a)). These reductions were as follows: −4, −42 and −30 mg/dL respectively for 5%, 10% and 20% corn oil, compared to −18, −42 and −47 mg/dL for T. conophorum oil with the same oil percentages. T. conophorum oil induced a significant reduction in LDL levels compared to corn oil. There was no difference between the TC10 and TC20 groups.

![Figure 4. Serum LDL cholesterol (mg/dl) (a) and HDL cholesterol (mg/dl) (b) concentrations in rats with respect to the percentage of oil from CO and TC. Means ± SD (n = 6). CO: Corn oil; TC: T. conophorum oil.](image)
The results also mentioned that, irrespective of the oil percentage in the diet, HDL cholesterol levels were significantly (p < 0.05) higher with the corn and *T. conophorum* oil diet groups compared with the control diet (Figure 4(b)). This increase was proportional to the percentage of oil for CO5 and CO10. The results also indicate that TC diets induced a significantly augmentation of higher serum HDL-C compared to CO diets. This increase was 29, 46 and 45 mg/dL respectively for TC5, TC10 and TC20 diets. There was no difference between the TC10 and TC20 diets.

### 3.5.4. Levels of Triglyceride in Different Groups

Triglycerides can come from the diet or from endogenous synthesis. Their measurement is generally used to classify different types of lipoproteinemia.

Changes in triglyceride levels after 5 weeks of experimentation are shown in Figure 5. Regardless of oil type and percentage, triglyceridemia decreased.

![Figure 5. Serum triglyceride levels (mg/dl) in rats fed with CO and TC at different percentages. Means ± SD (n = 6). CO: Corn oil; TC: *T. conophorum* oil.](image)

### 3.5.5. Effect of *T. conophorum* Oil and Corn Oil on Atherogenic Index

Castelli risk indices (CT/HDL (CRI-I) and LDL/HDL (CRI-II)) are the best markers for identifying and minimizing the risks of cardiovascular disease. These indices also allow early management of cardiovascular diseases [37]. The CRI-I is also called cardiac risk indices. The higher it is, the greater the risk of developing cardiovascular diseases. The results depicted in Figure 6 show that for all the groups supplemented with oil, CRI-I is less than 3.5, which is the favorable limit [38]. In this work, the values are lower for the TC regimes than for the CO regimes. Moreover, this ratio decreases at 5% oil (p < 0.05) and remains constant between 10 and 20% for TC oil.

The LDL/HDL or CRI-II ratio is used makes to justify a predisposition to atherosclerosis. It is favorable when its value is less than 3 [38], which is the case for all groups receiving corn oil and *T. conophorum* oil (Figure 6). Regardless of the
type of oil, CRI-II decreases with increasing oil content; values remain constant between 10 and 20% oil. This reduction is more represented with TC diets than CO diets.

3.5.6. Total Serum Protein Levels
Serum assays carried out on the rats after 5 weeks of experimentation were used to determine the protein levels in each group of rats (Figure 7). Protein levels increased with 5% oil, then decrease between 10 and 20% for all types of oils; but there is no significant difference (p < 0.05) between the CO and TC groups whatever the oil content.
4. Discussion

Worldwide, cardiovascular disease is the leading cause of death, responsible for around 30% of all deaths, and this figure is obviously rising in low- and middle-income countries [39]. Dyslipidemia is a major determinant of cardiovascular disease [40].

The aim of this research work was to determine the effect of linoleic and α-linolenic acid-rich sources on the lipemia of adult rats.

As far as changes in body mass are concerned, at the start of the experiment, there was almost no change in body mass. This can be explained by the age of the animals. The animals are 20 - 21 weeks old (adults) and are no longer growing, so they eat to maintain their weight. Subsequently, animals in the TC groups showed a decrease in body weight and adiposomatic index compared to those in the CO groups. This may be explained by the high α-linolenic acid content of the groups. Bashir et al. [41] noted that flaxseed oil supplementation (containing around 68% α-linolenic acid) significantly reduced body weight. This is also in line with the study carried out by Yari et al. [42], who found that consuming 30 g of flaxseed in people suffering from hepatic steatosis significantly reduced body mass index. In fact, consumption of unsaturated fatty acids down-regulates genes linked to lipogenesis and up-regulates genes linked to fatty acid oxidation, thus inducing a reduction in serum triglycerides, fatty acid levels, cellular uptake of fatty acids and the extent of fat deposits [43].

Regarding the liver weight of the rats, the results indicate that there is no induction of fat in the liver. These oils are not stored in the liver and would be metabolized by β-oxidation for energy production or to serve other syntheses. This is even more elucidated when the diet is rich in ω-3, such as the TC20 diet. Indeed, work by Huang et al. [44]; Jiang et al. [45] have shown that polyunsaturated fatty acids, particularly ω-3, have a significant impact on reducing hyperglycemia and its associated complications. These results suggest that corn and T. conophorum oils have hypoglycemic actions. The most appropriate proportion for T. conophorum oil was 10%.

The low levels of total cholesterol and LDL-C in the corn and T. conophorum oil-fed groups, compared with the control group, can be explained by the essential fatty acids (linoleic and linolenic). Indeed, high levels of polyunsaturated fatty acids in the body lead to a reduction in total serum cholesterol levels, and this is related to the high quantities of oil in the feed [46] [47]. According to [48], polyunsaturated fatty acids lower LDL cholesterol levels compared to saturated fatty acids. In addition, unsaturated fatty acids are associated with increased uptake of plasma LDL by the liver via LDL receptors in rats [49].

The fact that total cholesterol, LDL and triglyceride levels were lower for the T. conophorum oil groups than for corn oil may be justified by the proportion of α-linolenic acid, which has the capacity to lower total cholesterol levels more significantly than linoleic acid [46]. In addition, Lecerf [50]; Aloufi [51] and Sokola-Wysoczańska et al. [52] have shown that an increase in fatty acids from...
the ω-3 family generally decreases triglyceride levels. Maybe this decrease is due to an inhibition of hepatic lipogenesis as well as an increase in β-oxidation of fatty acids in the liver, leading to a decrease in VLDL synthesis and secretion by the liver [53]. Similar observations were made by Ghabadi et al. [54] who indicated that rapeseed oil consumption was associated with reductions of nearly 7.24 mg/dL in total Cholesterol and 6.4 mg/dL in LDL-C compared to sunflower oil. Yue et al. [47] found that administration of α-linolenic acid to normal rats reduced triglyceride, total and LDL cholesterol levels, with no effect on HDL cholesterol levels. Jamilian et al. [55] demonstrated that daily supplementation for 6 weeks with flaxseed oil capsules in women with gestational diabetes reduced triglyceride (−40.5 mg/dL) and total cholesterol (−22.7 mg/dL) levels compared with a matched group given sunflower oil capsules. The same trend is true with findings of Sokola-Wysoczanska et al. [52].

Increasing consumption of T. conophorum oil to 10% could potentially prevent the risk of cardiovascular disease.

Corn and T. conophorum oils facilitate HDL cholesterol synthesis. The highest levels are observed with T. conophorum oil regardless of the percentage, probably due to the high proportion of α-linolenic acid. Similar observations were made by Aloufi [51] using linseed oil. Indeed, ω-3 leads to an increase in HDL levels this effect is attributed to a decrease in plasma-free fatty acids and a further decrease in the transfer of cholesterol-free esters from HDL particles to LDL and VLDL particles [56]. The results therefore show that T. conophorum and corn oils, whatever their proportions, minimize the risk of atherosclerosis.

Corn and T. conophorum oils, whatever their proportions, contribute to minimizing cardiovascular risk, based on the values found for Castelli’s risk indexes.

The high polyunsaturated fatty acid content of T. conophorum and corn oils does not induce protein synthesis.

5. Conclusion

T. conophorum and corn oils can be useful in reducing the risk of atherosclerosis and can help prevent cardiovascular disease. A proportion of 10% TC oil is the most suitable.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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