Spirulina and Biochemical Blood Parameters of Wistar Rats (Rattus Norvegicus) in Growth

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INTRODUCTION

Malnutrition is a phenomenon plaguing many developing countries. According to the report on state of the food security and nutrition in the world 2018, the number of hungry people in the world rose to 821 million in 2017, or one in nine (WHO, 2018). The proportions in Sub-Saharan Africa are increasing with an estimated rate of 22.7 % in 2016 against 20.8 % in 2015 (FAO, 2016a). To remedy this, world organizations such as WHO (World Health Organization) and FAO (Food and Agriculture Organization), respectively in charge of health and food policies, have recommended to researchers around the world to re-examine the potentials food of humanity (sources). In this case, many research teams have been interested in Spirulina which is a micro-alga with nutritional and therapeutic properties (Qureshi et al., 1995; Belay, 2002; Mao et al., 2005). In view of its role in the fight against malnutrition, Spirulina is one of the unconventional nutritional and therapeutic sources which is full of nutritional information. However, consumption of Spirulina could help correct or improve certain nutritional dysfunctions that have not yet been elucidated. The aim of this work is to evaluate the effect of Spirulina (Spirulina platensis) supplementation on certain biochemical blood parameters in growing wistar rat (Rattus norvegicus) for three months. Four homogeneous groups of six rats each, contained three males and three females, were used. Group 1 (control) received conventional food (FACI® granules) ad libithum and distilled water at a rate of 10 ml/kg of body weight while groups 2, 3 and 4, received FACI® granules, Spirulina at 10 (Group 2), 50 (Group 3) and 100 mg/kg (Group 4) of body weight. Blood samples collected from the orbital sinus permitted to take 5 ml of blood that were centrifuged at 3000 rpm for 10 min. The serum collected was used to determine biochemical parameters. Analysis of results revealed that transaminases (AST and ALT) decreased significantly (P < 0.05) at 50 and 100 mg/kg b.w. Lipids measured out as well as glycemia knew also a significant (P < 0.05) decrease at all doses. As for creatinine and urea, they have all known a significant (P < 0.05) decrease as well as total and conjugated bilirubins. Total protein increased significantly (P < 0.05) at 50 and 100 mg/kg b.w. In conclusion, Spirulina supplementation acts favorably on blood biochemical parameters and could therefore contribute to improve certain vital organs functioning, in particular liver and kidneys.

Keywords: Spirulina, Conventional food supplement, Biochemical blood parameters, Rat (Rattus norvegicus).

MATERIAL AND METHODS

Spirulina It consists of Spirulina platensis. This cyanobacterium was supplied by the Spirulina Laboratory of Mé (South of Côte d'Ivoire) in powder...
form. The obtained quantity was 400 grams. The nutritional composition of the alga is shown in table 1 according to Kambou et al. (2016).

Table 1: Chemical composition of administered foods

<table>
<thead>
<tr>
<th>Constituents</th>
<th>FACT%</th>
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<tbody>
<tr>
<td>crude protein material (%)</td>
<td>15</td>
</tr>
<tr>
<td>crude fat (%)</td>
<td>3.5</td>
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<tr>
<td>Cellulosic material (%)</td>
<td>12</td>
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<tr>
<td>Mineral material (%)</td>
<td>9</td>
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<tr>
<td>Calcium (%)</td>
<td>1</td>
</tr>
<tr>
<td>Phosphorus (%)</td>
<td>0.9</td>
</tr>
<tr>
<td>Sodium (%)</td>
<td>0.3</td>
</tr>
<tr>
<td>Vitamin A (IU/Kg)</td>
<td>15000</td>
</tr>
<tr>
<td>Vitamin D3 (IU/Kg)</td>
<td>3000</td>
</tr>
<tr>
<td>Vitamin E (mg/Kg)</td>
<td>10</td>
</tr>
</tbody>
</table>

Animal

Albino Wistar rats of Rattus norvegicus species, male and female weighing on mean 26.2 ± 2 g and four weeks’ old were used. They were bred in the Animal House of Physiology, Pharmacology and Pharmacopeia Laboratory of the University of Nangui Abrogoua (Abidjan, Côte d’Ivoire) according to the principles for the care and use of laboratory animals of the Ethical Committee of the University (Nanguí Abrogoua, Abidjan, Côte d’Ivoire). They were exposed to 12 h dark/ light cycle and given FACI® brand granules for rats and water ad libitum. Research was conducted in accordance with the internationally accepted principles for laboratory use and care as found in the European Council on Legislation 87/609/EEC (EU, 2010).

METHODS

Preparation and Administration of Spirulina in Rats

The powder of Spirulina (Spirulina platensis) was obtained from the Spirulina Laboratory of Mé (South of Côte d’Ivoire). Three doses of Spirulina solution (10, 50 and 100 mg/kg) were administered to groups 2, 3 and 4 respectively by oral route using a cannula, a volume of 1 ml of Spirulina solution/100 g of body weight as a food supplement for three months.

Measurement of Serum Biochemical Parameters in Rats

Four homogeneous groups of six rats each, contained three males and three females, were constituted. Group 1 (control) received conventional food (FACI® granules) ad libitum, and distilled water at a rate of 10 ml / kg of body weight. Groups 2, 3 and 4, received FACI® granules and Spirulina at a rate of 10 (Group 2), 50 (Group 3) and 100 mg / kg (Group 4) of body weight (b.w.) respectively. Food intake was assessed daily by determining the difference between the amount of food distributed and refusals using a precision digital balance (SF-400 and S-234 Neo Tech SA, Belgium). Blood samples were performed on the 28th and 70th days from fasted rats the previous evening and anesthetized with ether early in the morning from the orbital sinus according to Jones and Mohr (1990) method. About 5 mL of blood was collected in dry tubes, centrifuged at 3000 rpm for 10 minutes then transported to the laboratory to measure a few serum biochemical parameters including lipids, proteins, ions, renal and hepatic biomarkers using a semi- automatic analyzer (RAYTO RT 9200).

STATISTICAL ANALYSIS

Data are presented as means ± SEM. The evolution of the serum parameters according to each growth period chosen for the animals for all doses of Spirulina on the one hand, and on the other hand, the changes of these blood parameters according to each dose of Spirulina during the growth of the animals, were made using ANOVA (one and two ways) and values of p < 0.05 were considered statistically significant using Graph Pad Prism 5.01 (San Diego, California, USA) software. The analysis was followed by multiple comparisons of the mean values of the different parameters using the Bonferroni post hoc test, if significant differences were revealed between the tested means.

RESULTS

Evolution of biochemical blood parameters in rats At 28th day

These results on 28th day showed a decrease in glycemia, albumin levels and A/G ratio while total protein and globulin levels were rather in an increase from controls to supplemented group, at a rate of 100 mg/kg bw (Table I). In addition, all lipid markers decrease in rats treated with Spirulina at doses ranging from 10 to 100 mg/kg bw. However, a decrease was recorded with the levels of total and conjugated bilirubin, urea and creatinine at doses of Spirulina ranging from 10 to 100 mg/kg bw compared to controls (Table II).

Likewise, transaminases, namely AST and ALT, decreased progressively according to the doses of received Spirulina. The evaluation of levels revealed that they are between: 10.94 ± 0.31 and 11.63 ± 0.18 mg/dL for calcium, 5.18 ± 0.27 and 5.80 ± 0.09 mmol/L for potassium, 131.00 ± 7.61 and 136.92 ± 1.84 mmol/L for sodium, 99.90 ± 1.21 and 103.43 ± 1.85 mmol/L for chloride. As for Na/’K’ ratio, values are between 23.61 ± 2.04 and 25.29 ± 2.81. No significant difference was reported between Spirulina quantity at 28th day (Table II).
The obtained lipid markers values were shown a decrease in rats treated with Spirulina at doses ranging from 10 to 100 mg/kg bw compared to controls (Table II). Likewise, transaminases, namely AST and ALT, decreased according to the doses of Spirulina received. The ions different values were between 11.05 ± 0.61 and 11.48 ± 0.55 mg/dL for calcium, 5.39 ± 0.16 and 5.97 ± 0.25 mmol/L for potassium, 11.31 ± 0.74 and 11.51 ± 0.08 mg/dL for sodium, and 5.39 ± 0.16 and 11.48 ± 0.55 mg/dL for calcium, 5.39 ± 0.16 and 11.48 ± 0.55 mg/dL for calcium. With regard to Na+/K+ and Ca2+/Mg2+ ratio, ratios obtained were between 23.62 ± 0.44 and 25.08 ± 3.94. No significant difference was reported between Spiruline quantity and 25.08 ± 3.94. No significant difference was reported between Spiruline quantity and 25.08 ± 3.94.

Table III: Distribution of biochemical parameters between the different doses of Spirulina on the 70th day of sampling

<table>
<thead>
<tr>
<th>Biochemical parameters</th>
<th>Spirulina (mg/kg body weight)</th>
<th>P values</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0</td>
<td>10</td>
</tr>
<tr>
<td>Glycemia (mg/L)</td>
<td>1.05±0.05</td>
<td>1.04±0.02</td>
</tr>
<tr>
<td>Total Proteins (mg/L)</td>
<td>57.8±4.83</td>
<td>62.8±3.39</td>
</tr>
<tr>
<td>Albumin (mg/L)</td>
<td>26.7±1.46</td>
<td>27.1±2.11</td>
</tr>
<tr>
<td>Globulins (mg/L)</td>
<td>31.09±3.35</td>
<td>35.6±1.24</td>
</tr>
<tr>
<td>A/G</td>
<td>0.86±0.09</td>
<td>0.76±0.03</td>
</tr>
<tr>
<td>Triglycerids (mg/L)</td>
<td>0.76±0.03</td>
<td>0.74±0.02</td>
</tr>
<tr>
<td>T Bilirubin (g/L)</td>
<td>0.72±0.05</td>
<td>0.58±0.03</td>
</tr>
<tr>
<td>C Bilirubin (g/L)</td>
<td>0.20±0.05</td>
<td>0.21±0.06</td>
</tr>
<tr>
<td>Total Cholesterol (g/L)</td>
<td>0.36±0.01</td>
<td>0.36±0.02</td>
</tr>
<tr>
<td>Atherogenicity indices</td>
<td>T-Chol/HDL</td>
<td>3.6±0.08</td>
</tr>
<tr>
<td></td>
<td>LDL/HDL</td>
<td>1.8±0.01</td>
</tr>
<tr>
<td>T Bilirubin (g/L)</td>
<td>2.9±0.08</td>
<td>2.7±0.16</td>
</tr>
<tr>
<td>C Bilirubine (g/L)</td>
<td>0.38±0.07</td>
<td>0.36±0.01</td>
</tr>
<tr>
<td>Creatinin (mg/L)</td>
<td>7.3±0.56</td>
<td>6.1±0.48</td>
</tr>
<tr>
<td>Urea (g/L)</td>
<td>0.14±0.01</td>
<td>0.13±0.03</td>
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<tr>
<td>Ca2+ (mg/dL)</td>
<td>11.14±0.33</td>
<td>11.48±0.55</td>
</tr>
<tr>
<td>K+ (mmol/L)</td>
<td>5.39±0.16</td>
<td>5.65±0.19</td>
</tr>
<tr>
<td>Na+ (mmol/L)</td>
<td>135.16±6.31</td>
<td>136.5±9.7</td>
</tr>
<tr>
<td>Cl− (mmol/L)</td>
<td>100.2±2.41</td>
<td>103.1±0.93</td>
</tr>
<tr>
<td>Na/K</td>
<td>25.08±3.94</td>
<td>24.17±5.25</td>
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<tr>
<td>ASAT (UI/L)</td>
<td>102.0±2.97</td>
<td>102.0±4.93</td>
</tr>
<tr>
<td>ALT (UI/L)</td>
<td>51.6±4.01</td>
<td>62.8±8.12</td>
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Evolution of Biochemical Blood Parameters during Rats Growth for Each Dose of Spiruline

Glycemia and protein in rats

The administration of conventional food (FACI® granules) and Spirulina to rats did not produce any significant ($P > 0.05$) variation in glycemia on the 28th and 70th day of sampling (Figure 1-A). In absence of Spirulina, administration of the conventional food did not modify total proteins level in control rats on the 70th day of sampling compared to the 28th day (Figure 1-B). When rats were treated with Spirulina at 10 and 100 mg/kg b.w., significant ($P < 0.05$) and highly significant ($P < 0.01$) increases respectively at 50 mg/kg and at 100 mg/kg bw from the 28th to 70th day (Figure 1-B) were recorded. The administration of the different doses of Spirulina to rats has induced a non-significant ($P > 0.05$) increase in albumin level at 10 and 100 mg/kg and a highly significant increase ($P < 0.01$) at 50 mg/kg bw from day 28 to day 70 were reported. The administration of Spirulina at doses ranging from 10 to 100 mg/kg has caused a significant increase ($P < 0.05$) at 10 mg/kg and highly significant ($P < 0.01$) at the doses of 50 and 100 mg/kg of globulin on day 28. On the 70th day. The observed increase was not significant ($P > 0.05$) at 10mg/kg, highly significant ($P < 0.01$) at 50 mg/kg and very highly significant ($P < 0.001$) at 100 mg/kg compared to the level of rats in the control group (Figure 1-D). However, from the 28th to the 70th day there was no significant variation ($P > 0.05$) in the different globulin levels within doses ranging from 10 to 100 mg/kg of Spirulina administered (Figure 1-D).

As for the albumin-globulin ration, the administration of the different doses of Spirulina was revealed a significant reduction ($P < 0.05$) at 10 mg/kg b.w and very highly significant ($P < 0.001$) at 50 and 100 mg/kg relative to control group. Likewise, this ratio decreased in doses ranging from 10 to 100 mg/kg bw compared to control group. This decrease was significant ($P < 0.05$) at 10 mg/kg, highly significant ($P < 0.01$) at 50 mg/kg and very highly significant ($P < 0.001$) at 100 mg/kg b.w. However, from 28th to the 70th days of sampling, A/G ratio increased in all doses. This increase was highly significant ($P < 0.01$) at 10 mg/kg and significant ($P < 0.05$) at 50 and 100 mg/kg (Figure 1-E).
Blood Lipids Profile
Administration of FACI® granules *ad libitum* to control rats, did not induce any modification of the triglyceride level at the 28th and 70th days of sampling (Figure 2-A). In contrast, in the group of rats which has received Spirulina orally at 10 mg/kg, a non-significant decrease ($P > 0.05$) from $0.80 \pm 0.02$ (28th day) to $0.74 \pm 0.02$ g/L (70th day) was recorded compared to the 28th day of treatment (Figure 2-A). In addition, in rats treated with Spirulina at 50 and 100 mg/kg, respectively, a non-significant decrease ($P > 0.05$) in triglyceride level ranging from $0.69 \pm 0.04$ to $0.65 \pm 0.05$ g/L (50 mg/kg) and $0.63 \pm 0.03$ to $0.61 \pm 0.04$ g/L (100 mg/kg) was observed compared to the 28th day of collection (Figure 2-A). With regard to total cholesterol level, no variation was recorded in absence of Spirulina in control rats on the 70th day compared to the 28th day (Figure 2-B).

However, Spirulina administration at 10 and 100 mg/kg to rats in presence of conventional food, induced a reduction in total cholesterol level on the 70th day compared to the 28th day of sampling. This decrease was very highly significant ($P < 0.001$) at 50 mg/kg, highly significant ($P < 0.01$) at 10 mg/kg and not significant ($P > 0.05$) at 100 mg/kg of Spirulina on the 70th day compared to the 28th day of sampling (Figure 2-B). HDL cholesterol level did not increase significantly ($P > 0.05$) at 10 mg/kg dose and remained almost identical to 50 mg/kg. At 100 mg/kg, a
significant ($P < 0.05$) decrease was obtained from the 28th to 70th day of sampling. As for LDL cholesterol level, it underwent a highly significant decrease ($P < 0.01$) in control rats from 28th to 70th sample and highly significant ($P < 0.001$) at doses between 10 and 100 mg/kg (Figure 2-C and 2-D). Indeed, administration of Spirulina at 100 mg/kg did not cause any significant change ($P > 0.05$) in atherogenicity index from the 28th to the 70th days evaluated on the ratio total/HDL cholesterol. However, this same ratio induced a very highly significant decrease ($P < 0.001$) in the same period at 10 and 50 mg/kg b.w (Figure 2-F). Regarding atherogenicity index calculated on LDL/HDL ratio, a very highly significant decrease ($P < 0.001$) was observed at all doses of Spirulina ranging from 10 to 100 mg/kg from 28th to 70th days of sampling (Figure 2-E).

**Figure 2:** Lipids profile and atherogenicity indices during the investigation for each dose of Spirulina in rats
A: Triglycerids, B: Total Cholesterol, C: HDL-Cholesterol, D: LDL-Cholesterol, E: LDL/HDL, F: Total Cholesterol/HDL.

**IONOGRAM**
Administration of spirulina at doses ranging from 10 to 100 mg/kg b.w. did not produce a noticeable change in calcium different levels from 28th to 70th days at all doses (Figure 3-A).

Calcium and potassium administration at different doses of Spirulina did not induce a significant ($P > 0.05$) change from 28th to 70th days in treated rats (Figure 3-B). A little variation of sodium level was observed after Spirulina administration at different doses ranging from 10 to 100 mg/kg. The obtained values were also identical at all doses and independent of sampling period. Chlorine level also did not change following Spirulina administration at various doses (10 mg/kg b.w, 50 mg/kg b.w and 100 mg/kg b.w) from the 25th to 70th days of sampling (Figure 3-C and 3-D). Furthermore, Na'/K' ratio from 28th to 70th day of
treatment did not produce any significant ($P > 0.05$) variation at all doses of Spirulina (Figure 3-E).

![Figure 3](image)

**Figure 3**: Ionogram during the investigation for each dose of Spirulina in rats
A: Calcium, B: Sodium, C: Potassium, D: Chloride, E: Na/K, n=6.

**Kidney and Liver Blood Parameters**

In Spirulina absence, administration of FACI® granules *ad libitum* caused in control rats, a non-significant decrease ($P > 0.05$) in urea level from 0.15 ± 0.02 (28th day) to 0.26 ± 0.05 g/L on 70th day compared to the 28th day of collection (Figure 4-B). Furthermore, excepted the dose of 50 mg/kg b.w of Spirulina where the decrease was significant ($P < 0.05$), when the rats received in addition granules and Spirulina at 10 and 100 mg/kg b.w, a non-significant decrease ($P > 0.05$) in urea level was observed on 70th day compared to 28th day of sampling (Figure 4-B). Also, creatinine level showed a non-significant decrease ($P > 0.05$) in the absence and presence of Spirulina (10 to 100 mg/kg b.w) on the 70th day of sampling compared to the 28th day (Figure 4-A). In absence of Spirulina, when rats received granules *ad libitum* from 28th to 70th day, AST level did not significantly ($P > 0.05$) increased from 91 ± 2.08 (28th day) to 102 ± 2.97 IU/L (70th day) at 70th day compared to the 28th day (Figure 4-C). On the other hand, when Spirulina in addition to the granules was administered orally to rats at doses ranging between 10 and 100 mg/kg b.w from 28th to 70th days, this caused a very highly significant decrease ($P < 0.001$) in AST level at 50 and 100 mg/kg on the 70th day compared to the 28th day of administration (Figure 4-C). In addition, this observed decrease in ASAT level was not significant ($P > 0.05$) at 10 mg/kg b.w on 70th day compared to the 28th day (Figure 4-C). Furthermore, when control rats did not receive Spirulina as a food supplement, ALT level significantly ($P > 0.05$) did not decrease from 52.33 ± 2.96 (day 28) to 51.6 ± 4.01 IU/L on 70th day compared to the 28th day of sampling (Figure 4-D). In addition, oral administration of Spirulina to rats at doses ranging from 10 to 100 mg/kg to conventional food, induced a decrease in different ALT levels on the 28th and 70th days of treatment. Indeed, a non-significant decrease ($P > 0.05$) was recorded in rats receiving doses of 10 and 50 mg/kg b.w and significantly ($P < 0.05$) at 100 mg/kg on 70th day.
compared to 28th day of sampling (Figure 4-D). Liver markers were evaluated during the experiment using different types of bilirubins. Regarding total bilirubin, the single administration of the conventional food on the one hand and Spirulina (10, 50 and 100 mg/kg bw) preceded by the conventional food on the other hand, produced on 70th day of sampling, a very highly significant decrease ($P < 0.001$) in this parameter in all the animals (controls and treated) compared to the sampling on the 28th day (Figure 4-E). As for conjugated bilirubin, on 28th and 70th day of sampling, a significant increase ($P < 0.05$) in its level in control rats from 0.31 ± 0.07 g/L (28th day) to 0.40 ± 0.02 g/L (70th day) was recorded (Figure 4-F). In addition, with rats that received Spirulina at doses varying from 10 to 100 mg/kg b.w, a very highly significant increase ($P < 0.001$) in conjugated bilirubin level compared to 28th day of sampling was observed at 10 and 100 mg/kg of Spirulina while a very highly significant decrease ($P < 0.001$) was recorded at 50 mg/kg on 70th day compared to 28th day of sampling (Figure 4-F).

**Figure 4:** Evolution of renal and hepatic blood parameters for each dose of Spirulina in rats
A: Creatinin, B: Urea, C: AST, D: ALT, E: Total Bilirubin, F: Conjugated Bilirubin

$*$ : $p < 0.05$ ; $**$ : $p < 0.01$ ; $***$ : $p < 0.001$, n=6.

**DISCUSSION**

The analysis of the biochemical blood parameters of rats permitted to estimate the performances related to the consumption of Spirulina (*Spirulina platensis*) and to identify the appropriate doses. Cholesterol and triglycerides are produced in large enough proportions in liver to supply the entire body (Thibaut, 2012). Their significant increase could be linked to excess weight. Therefore, during this study, these lipid markers underwent modifications. Indeed, administration of Spirulina at 10 mg/kg induced a highly significant decrease in triglyceride level. As for cholesterol level, it produced a decrease at all doses. This situation leads to say that Spirulina is a lipid-lowering substance as confirmed by Chamorro *et al.*
rubin produced a significant decrease in total cholesterol level in rabbits subjected to a hypercholesterolemic diet. These authors showed also that in rabbits, Spirulina caused a decrease in triglycerides. Likewise, Ponce-Canchihuanmán et al. (2010) showed that Spirulina maxima prevented significant changes induced by lead acetate on the levels of plasma and hepatic lipids and on the antioxidant status of the liver and kidneys in the male rat. In addition, Spirulina maxima succeeded in improving biochemical parameters of liver and kidneys towards the normal values of the control group. These beneficial effects could be justified by certain components of Spirulina itself. Indeed, Spirulina contained omega-3 and omega-6 fatty acids, beta-carotene, alpha tocopherol, phyocyanin, phenols and certain minerals which are substances deemed lipid-lowering (Chamorro et al., 2002).

Kato et al. (1984) have shown that cholesterol level decrease when food is supplemented by 16% with Spirulina. In carried out study on rats by Iwata et al. (1987), they have revealed that a diet enriched with 5, 10 and 15% Spirulina induced a significant reduction in total HDL cholesterols, triglycerides and phospholipids. In addition, total Cholesterol/HDL and LDL/HDL reports permitted to determine atherogenicity indices which provide information on arterial and coronary risk. Indeed, when these ratios are respectively higher than 4.85 (Total Cholesterol/HDL) and 3.55 (LDL/HDL) it means that subjects are exposed to cardiovascular diseases, in particular arteriosclerosis. However, the analysis of the results of this study has shown that the atherogenicity indices are respectively lower than 4.85 (total cholesterol/HDL) and 3.55 (LDL/HDL) suggesting that administration of Spirulina would prevent development of cardiovascular disease. Spirulina is therefore a lipid-lowering substance and could have a protective effect on the cardiovascular system. These results are in agreement with those of Nayaka et al. (1988) who demonstrated the ability of Spirulina to lower serum cholesterol, triglyceride and LDL levels in adult men.

Bilirubin is a bile pigment formed from the breakdown of hemoglobin, the rupture of veins but also other hemoproteins (cytochromes, catalases ...). Then it is picked up by the liver (conjugated or direct bilirubin) and degraded. It helps diagnose the causes of certain pathologies and dysfunctions of the organism such as anemia and liver diseases. According to Sutan et al. (1991), the level of conjugated bilirubin is increased in liver and biliary disorders, in particular the different types of hepatitis (viral, toxic, drug), rare metabolic abnormalities (Rotor disease of Dubin Johnson), gallstones, cholelithiasis, pancreatitis, cancer of the pancreas or bile ducts. Its dosage makes it possible to assess the elimination of bile by the bile ducts (Borenstein et al., 2006). Regarding studied serum bilirubin levels in this work, the results showed a significant decrease in mean values of total bilirubin in rat group treated with Spirulina at doses 50 and 100 mg/kg bw. However, at 10 mg/kg bw and also in the controls fed only with granules, these mean values remained practically identical. Hemolysis of red blood cells is reported to release the heme which is transformed into biliverdin and then bilirubin (Silbernagl and Lang, 2000). It should therefore be noted that following the administration of Spirulina, there would have been no hemolysis which could be at the origin of the increase in the bilirubin level. Similarly, conjugated bilirubin produced a significant decrease at 10 and 100 mg/kg bw and highly significant at 50 mg/kg bw from the 28th to 70th day of treatment. These results could therefore testify that the daily use of Spirulina does not lead to the deterioration of the organs in relation to the ailments caused.

Creatinine and urea are used to assess kidney function. Blood creatinine levels are a better indicator of kidney function. Higher levels of creatinine indicate a decrease in the rate of glomerular filtration and therefore a decrease in the ability of the kidneys to excrete waste (Gbakon et al., 2018). Creatinine and urea are excellent kidney markers, their modification reflects kidney dysfunction (Sirwal et al., 2004). The urea level decreased on the 70th day of sampling with all doses of Spirulina, thus attesting to a good functioning of the renal system of rats. Urea comes from the destruction of proteins. Its excretion is mainly by the kidneys and its rate reflects the overall functioning of the kidneys (Maurizi and Zaoui, 2005). Thus, certain factors such as protein-energy malnutrition and liver dysfunction can also decrease uremia (Lagrange, 2010). Creatinemia is, like uremia, used as an indirect marker of kidney function (Seronie et al., 2004). A decrease in creatinine level on the 70th day compared to the 28th day sample is also observed. This decrease in creatinine production in rats could confirm normal functioning of the renal physiology of these animals. Indeed, creatinine is formed in muscle from a non-enzymatic breakdown of creatine and is eliminated only by kidneys through glomerulus (Pierre et al., 2010). Similar observations have been reported by Zannou et al. (2011) who indicate that rats fed diets containing mixtures of cassava flour with soybeans have kidneys that weigh more than rats fed the casein diet. The obtained results also agree with those attest that Spirulina would reduce blood cholesterol level, would stimulate immunological system, would warn in order to prevent cancers, would reduce the nephrotoxicity of pharmaceuticals and metals toxic and would provide protection against the harmful effects of radiation (Sami et al., 2016).

In addition, serum calcium, sodium, chloride and potassium levels revealed good renal secretion in view of the almost stable values obtained following the administration of Spirulina at doses ranging from 10 to

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100 mg/kg bw. Sodium, potassium and chlorine ions are very important in maintaining osmotic pressure and water movements in body as well as in acid-base balance (Dieusaert, 2015). These results confirm those obtained with creatinine and urea and also reinforce the idea that Spirulina is a hepatoprotective substance and reduces nephrotoxicity as suggested by Mao et al. (2005) and Sami et al. (2016). However, these results contradict those of Iwasa et al. (2002) and Mazokopakis et al. (2008) who reported adverse effects on hepatic, dermatological, digestive and haematological, renal and electrolyte disorders in humans in Japan.

The evaluation of transaminases (ALT and AST) which are enzymes with important metabolic activity inside liver cells, allowed the exploration of the liver functions during the treatment. According to Najafi et al. (2012), the increase in these enzymes is mainly due to a leakage through the hepatic cytosol and their discharge into the blood stream (Saba, 2010; Najafi et al., 2012). Also, increased rate of AST is associated with myocardial infarction and liver damage and that of ALT with only liver damage (Ndouyang et al., 2018). ALT and AST levels rise rapidly when the liver is damaged for many reasons including hepatic cell necrosis, cirrhosis, and hepatotoxicity of certain substances (Pratt and Kaplan 2000; Dufour et al., 2000). This study showed a highly significant decrease, for AST and ALT levels, at doses 50 and 100 mg/kg bw, thus reflecting the good functioning of the liver of rats subjected to Spirulina. These results are similar to those of Mazokopakis et al. (2014) when they highlighted the lipid-lowering effect of the use of Spirulina on hepatic steatosis in adult dyslipidemic patients. These results are also consistent with those given by El-Bialy et al. (2016) when they evaluated the antioxidant effects of Spirulina in male mice where it showed that Spirulina normalizes the serum concentrations of transaminases.

CONCLUSION

The effects of Spirulina intake on biochemical blood parameters in rats show a hepatoprotective effect of transaminases including ALT and AST among all the studied parameters. Lipid markers as well as those of kidney, namely triglycerides, cholesterol levels, uremia and creatininemia have all also induced good regulation with regard to results obtained.

These results suggest the possibility of developing new nutritional approaches with Spirulina to help curb the disorders and dysfunctions related to malnutrition.

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Arthrospira platensis


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