EFFECT OF AQUEOUS LEAF EXTRACT OF HEINSIA CRINATA ON HAEMATOLOGICAL AND SOME BIOCHEMICAL INDICES OF TOXICITY IN STREPTOZOTOCIN INDUCED DIABETIC RATS

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ABSTRACT

AIM: The effect of the aqueous extract of Heinsia crinata leaves on haematological and biochemical parameters of streptozotocin induced diabetic rats were studied in vivo.

Study Design: The study which was carryout in the Biology Laboratory of the Rivers State Polytechnic, Bori; involved observing the in vivo effect of the administered pre-determined antihyperglycaemic effective dosage of the extract on Packed Cell Volume (PCV), Haemoglobin (Hb); Total White Blood Cell (TWBC), Platelets, Mean Cell Volume (MCV), Mean Cell Haemoglobin (MCH), Mean Cell Haemoglobin Concentration (MCHC), Red Blood Cell Count (RBC Count) and Differential Lymphocyte count and markers of erythrocyte toxicity (Aminotransferases and Alkaline Phosphatase) after a period of 21 days.

METHODOLOGY: The effective dosage of the extract determined from a pilot study and found to be 60mg/kg body, was administered to the streptozotocin induced (70mg/kg body weight) diabetic rats intraperitoneally. The rats weighing between 150 – 180g were divided in to four groups of five rats each representing normal control, diabetic control, glibenclamide (reference drug) and the extract treated group. They were fed with standard pellet ad libitum and housed in plastic cages with stainless steel wire gauge covering according to their groups and maintained at 12/24hours light and 27±1°C temperature. Their blood glucose levels were monitored by the one touch glucometer on a weekly basis and at the end of the 21day, the rats were painlessly sacrificed and blood obtained by cardiac puncture and used for haematological and biochemical analysis.
RESULT: There was significant ($p<0.05$) increase in the PCV (56± 9.43%), RBC (37.96± 2.32 $\times 10^9$cells/L) and Hb (13.00 ± 0.42g/dl) over the diabetic (42.67 ± 8.32% for PCV, 33.00± 6.44 $\times 10^9$cells/L for RBC and 12.77±3.62g/dl) and normal (45.33±1.65 for PCV, 26.02±4.65 for RBC and 12.93±3.43for Hb) controls respectively. Significant ($p<0.05$) reductions were also observed in levels of Total White Blood Cells, Platelet counts and Lymphocytes differential counts, AST and ALP activities in the extract treated rats as against the diabetic control.

CONCLUSION: The results showed that the use of the extract was capable of stimulating blood forming cells, reduce intracellular blood clot and maintain red cell integrity and are therefore recommended as a good nutraceutical for haemopoietic conditions.

KEYWORDS: HAEMATOLOGICAL PARAMETERS, AST, ALT, ALP, STREPTOZOTOCIN, HEINSIA CRINATA, AQUEOUS EXTRACT, HAEMOPOIETIC, NUTRACEUTICAL, INDICES OF TOXICITY.

INTRODUCTION

The use of plant extracted materials for ingestion as a means of treatment is a long standing tradition with most locals in the African continent (Okokon et al., 2009). The effectiveness of such medication is dependent on the drug interaction with the blood which is the common and major means of transportation of ingested or injected substances to their target sites. Such interaction between the potential drug and the blood is bound to evoke a reaction which will in turn add to affirm if the drug was suitable for its intended use or not.

Heinsia crinata is an edible vegetable noted as a delicacy amongst the people of Niger Delta in Nigeria (Abo et al., 2011; Miikue-Yobe et al., 2013) and so had been used most often in the processed form. However, with its emerging research finding that it contains an antidiabetic, antihypertensive and hypoglycemic active ingredient (Ajibesin et al., 2008; Okokon et al., 2009), its use has grown beyond been that of food source to that of medicinal substance. Indeed, its use by tradomedical personels in the treatment of various ailments is well known. Such use involve direct ingestion or injection the substance into the body and this may provoke some haematological as well as biochemical reactions on unsuspecting user. Consequently, this study was undertaking to assess these effects on biological system especially in a diabetic condition using the streptozotocin induced rat model.

METHODOLOGY

Plant extract.

Fresh leaves of Heinsia crinata was wash in distilled water and allowed to drain dry before it was ground to coarse form, 100g of it weighed and dissolved in 1000ml of distilled water. This was allowed to stand for twenty four hours with intermittent shaking to ensure proper extraction. The extract was filtered by the use of a muslin cloth and the filtrate freeze dried to yield a greenish brown solid substance that weighed 0.60g. The weighed substance was re-dissolved in 100ml 0f distilled water and used in the treatment of the diabetic rats.
Treatment of Diabetic Experimental Rats.

Five confirmed diabetic albino rats (with blood glucose levels $\geq 150$mg/dl {9.7mmol/l}) and weighing between 150 – 180g were treated with 1.0ml dosage of the aqueous extract of *Heinsia crinata* by intraperitoneal injection for a period of three days and then observed for 21 days thereafter. At the end of the period, at which point the extract’s hypoglycaemic activity was observed as reported in a previous research (Okokon *et al.*, 2009; Miikue-Yobe *et al.*, 2013), the rats were painfully sacrificed by exposure to chloroform vapour for a period of 5mins and blood obtained by blood by cardiac puncture. The blood samples were used for haematological and biochemical analysis. Result is here presented as means ± SEM. (Tables 1 and 2).

Haematological Analyses.

**Estimation Of Haemoglobin Concentration**
The Haemoglobin concentration was measured by cyanmethaemoglobin colorimetric method (Cheesbrough, 2004). A quantity of blood (20µl or 0.02ml) sample was added to 5 ml of the Drabkins solution in a test tube to achieve a 1: 250 dilutions. This was properly mixed and allowed to stand for 10mins, and thereafter absorbance ready of the test and standard samples were read at 540nm, against the reagent (Drabkin’s solution) blank.

**Determination of Packed Cell Volume**
Packed cell volume (PCV) or haematocrit was measured with micro-haematocrit, with 75x 16 mm capillary tubes filled with blood and centrifuged for 5 min (Cheesbrough, 2004). The EDTA anti-coagulated blood was collected into the capillary tubes which were then sealed at one end with plasticine, and centrifuged at 3000g for 5 min, after which red cell levels in the capillary tubes were read using the microhaematocrit reader.

**Estimation of White Blood Cell (WBC) Count**
The total white blood cell count was estimated by visual count method (Cheesbrough, 2004). The blood samples (0.02ml) were added to 0.38ml of Turks solution in a plain cuvette and thoroughly mixed. 0.01ml of the resultant mixture was loaded into the counting chamber, covered with the cover slide and allowed to settle (about 2min) before placing it under the microscope and viewed using the x10 objective. The white cells (N) present in the 4 corners and the central 1mm$^2$ areas were counted and from the result, the total WBC count was deduced by calculation.

**Differential WBC Count/ Cell Morphology**
The Leishman staining technique as reported by Cheesbrough (2004) was adopted in the differential count and cell morphology analysis. A drop of the EDTA anti-coagulated blood was placed on dry grease-free glass slide. A thin film of blood was made on the slide, which was allowed to air dry. The Leishman stain was then used to cover the film (drop wise). This was followed 2min later by the addition of the same number of drops of clean water, before mixing and allowed standing for 8 min on a flat surface. The stain was gently washed off tap water and the slide was allowed to dry in air, the dried slide was then viewed under oil immersion with a x100 objective, the various cells were counted and the requisite morphological notes taken.
Red Blood Cell (RBC) Count
The red cell count was estimated by the visual method of Cheesbourgh (2004).
EDTA anti-coagulated blood was mixed with Turks solution and viewed under the microscope.
To 4mL of the Turks solution in the plain cuvettes, was added 0.02mL of EDTA anti-coagulated blood sample, and thoroughly mixed, some of the resultant mixture (0.01 ml) was loaded into the counting chamber, covered and allowed to settle (2 min) before, placing under the microscope and viewing under x 10 objective, the white cells (N) present in the 4 corner and central 1 mm\(^2\) areas were counted and from the result, the red blood cells count was deduced by calculation.

Estimation of Mean Cell Haemoglobin (MCH)
This was calculated as reported by Cheesbrough (2004), using the following formula. It involves dividing haemoglobin concentration by the value of red blood cell count.

Estimation of Mean Cell Haemoglobin Concentration (MCHC)
The mean cell haemoglobin concentration was calculated as reported by Cheesbrough (2004), using the following formula. It requires the division of the haemoglobin concentration value by that of the packed cell volume.

Estimation of Mean Cell Volume (MCV)
The mean cell volume was calculated as reported by Cheesbrough (2004) using the following formula. This is the ratio of the packed red cell volume to that of the red blood cell multiplied by the factor of ten.

Biochemical Analyses

Assay of Plasma Alanine Aminotransferase (ALT) Activity
Alanine Transaminase activity was determined by monitoring the concentration of oxaloacetate hydrazone formed with 2, 4-dinitrophenylhydrazine according to Reitman and Frankel, (1957).
Two test tubes were set up labeled T\(_1\) (reagent blank) and T\(_2\) (test sample). T\(_1\) contained 0.01 mL distilled water and 0.50 ml Randox buffer solution, while T\(_2\) contained 0.01 ml plasma and 0.50 ml Randox buffer solution, the contents were mixed and incubated for 30 minutes at 37\(^{\circ}\)C to each tube of was added 0.05 ml of Randox 4-dinitrophenylhydrazine solution and the contents were mixed and allowed to stand for 20 min at 25\(^{\circ}\)C in a water bath. Then 5 ml of sodium hydroxide solution was added to each of the tubes. The contents were mixed, and after 5 min their absorbance values were read at 546 nm against the reagent blank in the spectrophotometer and the activity of the enzyme read off from the table.

Assay of Plasma Aspartate Aminotransferase (AST) Activity
The Plasma Aspartate Transferase activity was determined using Randox test kits (Randox laboratories Ltd.Crumlin. England, UK) according to Reitman and Frankel, (1957).
Aspartate transferase was determined by monitoring the concentration of oxaloacetate hydrazone formed with 2, 4-dinitrophenylhydrazine.
Two test tubes were set up labeled T\(_1\) (reagent blank) and T\(_2\) (test sample). T\(_1\) contained 0.01 ml distilled water and 0.05 ml Randox buffer solution, while T\(_2\) contained 0.01 ml plasma and 0.05 ml Randox buffer solution, the contents were mixed and incubated for 30 min at 37 \(^{\circ}\)C to each tube was added 0.05 ml of Randox 4-dinitrophenylhydrazine solution and the contents are mixed and allowed to stand for 20 min at 25\(^{\circ}\)C then 5 ml of sodium hydroxide solution as added to each of the tubes. The contents were mixed, and after 5 min their absorbances were red at 546 nm against the reagent blank in the spectrophotometer.
Assay of Plasma Alkaline Phosphatase (ALP) Activity
The Plasma Alkaline Phosphatase activity was determined using Randox Test Kits (Randox Laboratories Ltd, Crumlin England, UK) according to King and King, (1954).
The activity of alkaline phosphatase is determined by monitoring the rate of its dephosphorylation of p-nitrophenylphosphante to p-nitrophenol, a yellow colored compound whose concentration can be monitored colorimetrically at 405nm.
A 1.0 ml of the Randox ALP Reagent was added to 0.02 ml of the plasma in a cuvette at 30°C. The contents were then mixed and the initial absorbance was taken. Thereafter, readings were taken after 1, 2 and 3 minutes at 405 nm in a spectrophotometer. ALP activity was determined by using the formular.

Estimation of Plasma Bilirubin Concentration
The plasma bilirubin concentration was determined using Randox Test Kits (Randox Laboratories Ltd, Crumlin, England, UK) Malloy and Evelyn, (1937).

Total Bilirubin
Two tubes were set up labeled T₁ (sample blank) and T₂ (test sample). T₁ contained 0.20 mL Randox sulphanilic acid Reagent, 1.0 ml Randox caffeine solution and 0.2 ml of plasma, while Randox sulphanilic acid Reagent 0.05 ml Randox sodium nitrite solution, 1.0 ml of Randox titrate solution their content were mixed and allowed to stand for 30 min at 25°C in a water bath, before reading the absorbance (Aₜₐ₇) at 578 nm against the sample blank in a spectrophotometer.

Direct Bilirubin
Two test tubes were set up labeled T₁ (sample blank) and T₂ (test sample). T₁ contained 0.20 mL Randox sulphanilic acid Reagent, 2.00 ml of 0.9% sodium chloride solution and 0.20 ml plasma. The contents were mixed and allowed to stand for 5 min at 25°C, before reading the absorbance (Aₜₐ₇) at 546 nm against the sample blank in a Spectrophotometer.

Estimation of Plasma Total Protein Concentration
This was carried out by the Biuret Method, using Randox Test Kits (Randox Laboratories Ltd., Crumlin, England, UK) (Lyne, 1957).Three test tubes were set up labeled T₁ (blank), T₂ (standard) and T₃ (test sample). T₁ contained 0.04ml distilled water, 2.00ml Biuret reagent, T₂ contained 0.04mL standard protein solution and 2.00 ml burette reagent and T₃ containing 2.00ml of biuret reagent and 0.04ml of the test solution. The contents were thoroughly mixed, incubated at 25°C in a water bath for 30 minutes, before cooling and reading of absorbance at 560 nm against the blank, in a Spectrophotometer.

STATISTICAL ANALYSIS
Data were analyze statistically by the Duncan’s Multiple Range Test. Differences between groups were considered significant at p<0.05. (Sokal and Rohlf, 1969).
RESULT AND DISCUSSION

Result

The result of the study is presented in tables 1 and 2 below showing the effect of extract on haematological parameters (PCV, Hb, RBC, TWBC, WBC Differentials, MCV, MCH and MCHC) and biochemical parameters (Total Protein, AST, ALT, ALP and Bilirubin levels).

Table 1 revealed that the aqueous extract of *Heinsia crinata* caused a significant increase (p < 0.05) in the Packed Cell Volume which also reflected in the slight increase in the Haemoglobin concentration (13.00 ± 0.42 g/dl) over the other treatment groups. The extract also caused a significant rise in the Total White Blood Cell count (13.13 ± 2.50 x 10^9 cells/L) as against 6.60 ± 2.05, 11.52 ± 4.32 and 4.43 ± 1.11 x 10^9 cells/L for normal, diabetic controls and reference drug respectively. The Platelet count was observed to be significantly (p<0.05) increased (469.33 ± 4.76 x 10^4 cells/mm) than the values for normal control (373.00 ± 4.73 x 10^4 cells/mm) and reference drug (98.37 ± 2.45 x 10^4 cells/mm) but not above the value for the diabetic control (514.67 ± 2.32 x 10^4 cells/mm). Total Red Blood count was significantly reduced (p < 0.05) (7.96 ± 2.32 x 10^9 cell/L) by the extract which is increased in the normal and diabetic controls and the reference drug respectively. Lymphocytes were shown to be increased significantly (p < 0.05) by the extract (67.67 ± 0.44%) over that of the reference drug but lower than the values obtained for the normal (79.03 ± 2.51%) and the reference drug (52.67 ± 0.76) treated rats.

Total White Blood Cell count was significantly increased (p < 0.05) by the extract (8.47 ± 2.43%) than the normal control (6.60 ± 2.05%) and the reference drug (4.43 ± 1.11%) but is reduced (p<0.05) when compared to the value obtained in the diabetic controls.

The extract of *Heinsia crinata* caused a significant (p<0.05) reduction in the activities of the Amino Transferases (AST and ALT) but a significant activation of Alkaline Phosphatase (ALP) when compared with the reference drug, diabetic and normal controls respectively. The Total Protein level increased (p<0.05) by the extract over the other three treatment groups. The result obtained for the Bilirubin ratio for the extract and reference drug and diabetic and normal controls do not seem to differ significantly (p<0.05). This is also reflected in the trend in the results for the Total, Conjugated and Unconjugated Bilirubin.
<table>
<thead>
<tr>
<th>Parameters</th>
<th>Normal control</th>
<th>Diabetic control</th>
<th>Glibenclamide Treated</th>
<th>Extract Treated</th>
</tr>
</thead>
<tbody>
<tr>
<td>PVC (%)</td>
<td>45.33 ± 1.65</td>
<td>42.67 ± 8.32</td>
<td>36.00 ± 6.54</td>
<td>56.67 ± 9.43</td>
</tr>
<tr>
<td>TWBC Count (x 10^9 cell/L)</td>
<td>6.60 ± 2.05</td>
<td>11.52 ± 4.32</td>
<td>4.43 ± 1.11</td>
<td>9.97 ± 2.44</td>
</tr>
<tr>
<td>Platelets counts (x10^4 cells/mm)</td>
<td>373.00 ± 4.73</td>
<td>514.67 ± 2.32</td>
<td>198.37 ± 2.45</td>
<td>467.33 ± 4.76</td>
</tr>
<tr>
<td>RBC count (x10^9 cells/L)</td>
<td>26.02 ± 4.65</td>
<td>33.00 ± 6.44</td>
<td>33.79 ± 1.65</td>
<td>37.96 ± 2.32</td>
</tr>
<tr>
<td>Hb (g/dl)</td>
<td>12.93 ± 3.43</td>
<td>12.77 ± 3.62</td>
<td>11.80 ± 1.98</td>
<td>13.00 ± 0.42</td>
</tr>
<tr>
<td>Lymphocyte count (%)</td>
<td>79.03 ± 2.51</td>
<td>84.80 ± 1.43</td>
<td>52.67 ± 0.76</td>
<td>67.67 ± 0.47</td>
</tr>
<tr>
<td>Neutrophil count (%)</td>
<td>15.00 ± 0.44</td>
<td>9.00 ± 0.43</td>
<td>40.00 ± 0.87</td>
<td>28.00 ± 0.65</td>
</tr>
<tr>
<td>Monocyte count (%)</td>
<td>5.00 ± 2.11</td>
<td>6.00 ± 1.87</td>
<td>6.00 ± 1.57</td>
<td>6.00 ± 1.76</td>
</tr>
<tr>
<td>Basophil count (%)</td>
<td>0.00 ± 0.00</td>
<td>0.00 ± 0.00</td>
<td>0.00 ± 0.00</td>
<td>0.00 ± 0.00</td>
</tr>
<tr>
<td>Eosinophil count (%)</td>
<td>1.00 ± 0.11</td>
<td>1.00 ± 0.08</td>
<td>2.00 ± 0.54</td>
<td>1.00 ± 0.07</td>
</tr>
<tr>
<td>MCV (fl)</td>
<td>17.42 ± 0.35</td>
<td>12.93 ± 0.58</td>
<td>94.99 ± 0.96</td>
<td>71.19 ± 0.06</td>
</tr>
<tr>
<td>MCHC (g/dl)</td>
<td>0.27 ± 0.08</td>
<td>0.30 ± 0.44</td>
<td>0.33 ± 0.30</td>
<td>0.23 ± 0.04</td>
</tr>
<tr>
<td>MCH (pg/cell)</td>
<td>0.50 ± 0.74</td>
<td>0.39 ± 0.56</td>
<td>3.11 ± 1.20</td>
<td>1.63 ± 0.18</td>
</tr>
</tbody>
</table>

Results are Means ± SEM for n = 5 rats in each group. Experimental groups are compared with normal control and reference drug, glibenclamide. Values with different superscripts in the same row are significantly different at the P < 0.05 level. PCV = Packed cell volume, TWBC = Total White Blood Cell, RBC = Red Blood Cell, Hb = Haemoglobin, MCV = Mean Cell Volume, MCH = Mean Cell Haemoglobin and MCHC = Mean Cell Haemoglobin Concentration
### Table 2.0 INDEX OF TOXICITY OF AQUEOUS LEAF EXTRACT OF *Heinsia crinata* ON STREPTOZOTOCIN-INDUCED DIABETIC RATS

<table>
<thead>
<tr>
<th>Treatment Groups</th>
<th>Total Protein (g/l)</th>
<th>Aspartate Transaminase (AST) IU/L</th>
<th>Alanine Transaminase (ALT) IU/L</th>
<th>Alkaline Phosphatase (ALP) IU/L</th>
<th>Total Bilirubin (µmol/L)</th>
<th>Conjugated Bilirubin (µmol/L)</th>
<th>Unconjugated Bilirubin (µmol/L)</th>
<th>Uncojugated/Conjugated Bilirubin</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Normal Control</strong></td>
<td>56.00±0.03</td>
<td>826.33±1.11</td>
<td>71.33±5.4</td>
<td>70.33±0.34</td>
<td>3.67±1.43</td>
<td>3.33±1.54</td>
<td>0.34±0.60</td>
<td>0.12±0.10</td>
</tr>
<tr>
<td><strong>Diabetic Control</strong></td>
<td>59.00±0.32</td>
<td>66.67±0.4</td>
<td>22.67±0.6</td>
<td>192.67±6.5</td>
<td>2.67±0.77</td>
<td>2.00±0.87</td>
<td>0.67±0.01</td>
<td>0.34±0.11</td>
</tr>
<tr>
<td><strong>Glibenclamide</strong></td>
<td>74.33±0.43</td>
<td>63.33±2.8</td>
<td>26.67±0.2</td>
<td>201.67±5.5</td>
<td>7.33±2.43</td>
<td>2.00±0.76</td>
<td>5.33±0.80</td>
<td>2.67±1.21</td>
</tr>
<tr>
<td><strong>Extract Treated</strong></td>
<td>77.67±0.54</td>
<td>26.00±0.2</td>
<td>25.33±0.6</td>
<td>180.67±5.3</td>
<td>7.00±2.76</td>
<td>2.33±0.09</td>
<td>5.33±0.06</td>
<td>2.29±0.75</td>
</tr>
</tbody>
</table>

Results are Means ± SEM for n = 5 rats in a group. Experimental groups are compared with normal control and reference drug, glibenclamide. Values with different superscript in a column are significantly different at the P < 0.05 level.

**Discussion**

The assessment of haematological parameters is a useful guide to determining the extent of deleterious effect of foreign compounds including plant extracts (Mohammed et al., 2009; Edet et al., 2013). They are also used to determine possible alterations in the levels of biomolecules such as enzymes, metabolic product, haematology, normal functioning and histomorphology of the organs (Magalhaes et al., 2008; Oyedemi et al., 2011).

Anaemia has been severally reported as a complication of diabetes mellitus (Kotharia and Bokariya, 2012). It results due to the increase in non-enzymatic glycosylation of Red blood cells (RBC) membrane proteins. The oxidation of these proteins and in the presence of hyperglycemia as obtainable in diabetes results to lyases of the blood cells and so anaemia ensues (Oyedemi et al., 2011).

In diabetes, the value of PCV, Hb, RBC, MCV, MCH and MCHC are reduced due to lyases of blood cells caused by reactive oxygen species (ROS) and the resulting oxidative stress (oyedemi et al., 2011; Mohammad et al., 2013; Uko et al., 2013). The implication is an accompanying anaemia in diabetes (Kotharia and Bokariya, 2012). This is the trend observe in our study as the diabetic control result is compared with the normal control result. However, extract treatment caused significant (p<0.05) changes in the value of these parameters such that it brought about a significant increase in the MCH value, a factor that measures the rate of erythrocyte synthesis. It therefore can be deduced that extract was able to reverse the lytic effect of ROS and so reduced or rather completely prevent oxidative stress thereby giving room for the regeneration of erythropoietin cells, a process mediated by erythropoietin.
secretion from the bone marrow (Ohission *et al.*, 2006; Oyedemi *et al.*, 2011). The overall effect is the restoration of the oxygen carrying capacity of the Red Blood Cells as a result of the inhibition of the process of lipid peroxidation in the membrane of RBC (Ashafa *et al.*, 2009).

Changes in TWBC have been associated with insulin resistance and complications of CVD (Mohammad, 2013; Uko *et al.*, 2013). The result of our study showed a significant (p<0.05) increase in the value of TWBC in the diabetic control which became reduced significantly on extract treatment. This may be interpreted to mean the extract’s ability to restore insulin sensitivity to the cells (Uko *et al.*, 2013). This position was further strengthened by the observed reduction in Leucocytes particularly those of Lymphocytes in comparison with the diabetic control. This agrees with other reports (Edet *et al.*, 2013; Tanko *et al.*, 2008) implying a reduction in the process of advance glycation and oxidative stress within the blood cells.

The liver is known to play an important role in the metabolism of carbohydrate and so its cellular integrity can be compromised in diabetes and related disease (Harris, 2005). Therefore, its test of function is performed to assess its pathological condition. Prominent among these tests are the analysis of AST, ALT, ALP activities and Bilirubin levels. Our finding as shown in table 2 shows that extract treatment significantly (p<0.05) reduced AST and ALP activities and similarly increased activity of ALT and the level of total Bilirubin. ALT is known to participate in gluconeogenesis and its transcription is suppressed by insulin. Its increased activity is therefore suggestive of impairment of insulin signaling and not hepatocyte injury. Similarly reduction of ALP activity as shown by extract treatment is suggestive of the extract ability to protect the cell from cytotoxic injury. Mild increase in bilirubin levels has been proposed to have a protective effect on cells (Vitek, 2012). The increases observed in this study in the levels of Bilirubin go to support its cell protecting potential.

**CONCLUSION**

It is therefore concluded that the effect of the aqueous extract of *Heinsia crinata* on streptozotocin induced diabetic rats as demonstrated in this study is capable of stimulating blood cell formation (erythropoiesis) and confer protection to hepatocyte against cell injury due the effect of oxidative stress and lipid peroxidation. It is therefore recommended that the extract be considered as a nutraceutical and should be used in the treatment of anaemia and Liver disorders that result from oxidative stress reactions.

**CONFLICT OF INTEREST**

The authors state that they do not have any conflict of interest regarding any part of the publication.

**ACKNOWLEDGEMENT**

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REFERENCES


