

*Full Length Research Paper*

## **Effects of methanolic tuber extract of *Cyperus esculentus* Linn (tiger nuts) on sub-acute liver damage in albino rats**

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This study evaluated the effects of *Cyperus esculentus* methanolic tuber extract (CEME) on carbon tetrachloride (CCl<sub>4</sub>)-induced sub-acute liver damage in albino rats. Dried tubers of *C. esculentus* were pulverized and extracted by cold maceration, using 80% methanol. Thirty albino rats, randomly assigned to 6 groups (A–F) of 5 each were used for the study. Sub-acute liver damage was induced in Groups A–E rats using intra-peritoneal injections of CCl<sub>4</sub>. Group A was treated with distilled water placebo, while Groups B, C and D were treated with 200, 400 and 800 mg/kg CEME, respectively. Group E was treated with 100 mg/kg Silymarin and Group F was also given distilled water placebo. Treatment was done orally for 15 days, after which hepatocellular integrity and liver function were evaluated. Results showed that treatment with CEME (at all doses) led to significantly lower ( $p < 0.05$ ) serum alanine aminotransferase and aspartate aminotransferase activities, bilirubin levels and relative liver weight of the CEME-treated groups, when compared to Group A rats. It was concluded that administration of CEME as used in the study led to significant protection of hepatocellular integrity, enhancement of hepatic excretion of bilirubin and amelioration of CCl<sub>4</sub>-induced inflammatory enlargement of the liver.

**Key words:** *Cyperus esculentus*, methanolic tuber extract, hepatoprotection, liver damage.

### **INTRODUCTION**

The liver is the largest solid organ in the body (Kuntz and Kuntz, 2006; Saukonnen et al., 2006). It is involved in the

metabolism of numerous substances (including bilirubins, porphyrins, bile acids, amino acids and proteins,

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carbohydrates, lipids and lipoproteins, hormones and vitamins), biotransformation, detoxification and acid-base balance (Kuntz and Kuntz, 2006). Its multi-various functions constantly expose it to injury that may lead to different forms of liver disorders/diseases (Ihedioha, 2005; Saukonnen et al., 2006). Liver diseases have a worldwide distribution, and are a major cause of morbidity and mortality globally (Blachier et al., 2012; Lozano et al., 2012; Nwokediuko et al., 2013; Sarin and Maiwal, 2018). Diseases of the liver have been ranked the fifth most common cause of death and the second leading cause of mortality amongst all digestive diseases (Williams, 2006; Lozano et al., 2012; Rehm et al., 2013; Sarin and Maiwal, 2018). Toxic liver diseases constitute a large proportion of liver disorders/diseases, and its occurrence has been steadily increasing over the years (Suk and Kim, 2012; Rehm et al., 2013; Nwokediuko et al., 2013).

Plants constitute a large part of traditional medicines and continue to provide mankind with therapeutic remedies and novel drug leads (Gurib-Fakim, 2006; Newman and Cragg, 2012). Despite the availability of modern medicines, medicinal plants are commonly used in developing countries to meet most primary health care needs, and many people in developed countries also patronize medicinal plants-based alternative and complementary therapies (WHO, 1999; Veeresham, 2012). However, most plant species that are traditionally used as medicines have not been scientifically evaluated for efficacy and possible orthodox medical applications (WHO, 1999; Gurib-Fakim, 2006; Atanosov et al., 2015).

*Cyperus esculentus* is a grass-like plant in the family *Cyperaceae* (De Vries, 1991; Takhatajah, 1992; Coskuner et al., 2002; Dhouha et al., 2016). It is commonly known as chufa (in Spanish) (Oderinde and Tairu, 1988; Dhouha et al., 2016), other names include tiger nut, earth nut, yellow nut sedge, ground nut and rush nut (Oderinde and Tairu, 1988; Umerie et al., 1997; Coskuner et al., 2002; Oladele and Aina, 2007; Arafat et al., 2009; Sanchez-Zapata et al., 2012; Dhouha et al., 2016). Tiger nut tubers are freely growing and are eaten uncooked in their natural form (Ejoh et al., 2006). Three varieties of tiger nut tubers are available; they are yellow, black and brown varieties (Umerie et al., 1997; Okafor et al., 2003; Belewu and Abodunrin, 2006; Oladele and Aina, 2007; Arafat et al., 2009), are widely distributed in Europe and Africa including Nigeria where they are widely consumed uncooked (Oderinde and Tairu, 1988; Omode et al., 1995; Ejoh et al., 2006; Dhouha et al., 2016). Tiger nut tubers are rich in starch, fats, sugars, proteins, oleic acid, and vitamins B, C and E (Temple et al., 1990; Omode et al., 1995; Okwu, 2005; Belewu and Belewu, 2007; Dhouha et al., 2016). It has been also reported that they are rich in minerals such as phosphorous, potassium, calcium, magnesium and iron (Temple et al., 1990; Omode et al., 1995; Belewu and Belewu, 2007; Oladele and Aina, 2007; Arafat et al.,

2009; Dhouha et al., 2016). Their antioxidant capacity is known to be relatively high because reports from previous studies have shown that they contain considerable amounts of water-soluble flavonoids and glycosides which are known natural antioxidants (Temple et al., 1990; Eteshola and Oraedu, 1996; Pietta, 2000; Oloyede et al., 2014).

For many years, the tubers of *Cyperus* species have been used traditionally as remedy for several diseases including hepatotoxicity (Mehta et al., 1999; Hassanein et al., 2011) and as antioxidative agent (Satoh et al., 2004). Although many researchers have worked on *C. esculentus* tubers, the tubers are not well utilized due to limited information on their medicinal potential and nutritional benefits (Rita, 2009; Adejuyitan, 2011; Ukwuru and Ogbodo, 2011; Oyedepo and Odoje 2014). Ameen et al. (1999) reported that oral administration of oily extracts of *C. esculentus* tuber significantly protected against carbon tetrachloride (CCl<sub>4</sub>)-induced hepatic damage in male albino rats. Oyedepo and Odoje (2014) reported that administration of tiger nut flour in varying percentages in rat pellets for 21 days preceding CCl<sub>4</sub> administration exhibited a potential hepatoprotective activity against CCl<sub>4</sub>-induced hepatotoxicity in male Wister albino rats. Also, recent report by Onuoha et al. (2017) showed that oral administration of tiger nut based nutri-milk to rats preventively ameliorated acetaminophen-induced hepatotoxicity. There is no information in available literature on the effects of methanolic tuber extract of *C. esculentus* on sub-acute liver damage, hence the present study which evaluated the effects of methanolic tuber extract of *C. esculentus* Linn on CCl<sub>4</sub>-induced sub-acute liver damage in albino rats.

## MATERIALS AND METHODS

### Drugs, chemicals, clinical chemistry assay kits and equipment

Methanol, carbon tetrachloride (CCl<sub>4</sub>) and silymarin were obtained from Sigma-Aldrich, St. Louis, Missouri, USA. Thiopentone sodium was obtained from Chandra Bhagat Pharma Pvt., Ltd., Mumbai, India. The assay kits for clinical biochemistry evaluation of alanine aminotransferase (ALT), aspartate aminotransferase (AST), and alkaline phosphatase (ALP) activities, and levels of total proteins, total cholesterol and albumin in serum were procured from Quimica Clinica Aplicada (QCA), Spain. The test kit for the assay of total bilirubin in serum was procured from Randox Laboratories Ltd, County Antrim, United Kingdom. All the clinical chemistry determinations were done using the CHEM5V3<sup>®</sup> Semi-automated Clinical Chemistry Analyzer (Erba Diagnostics, Mannheim, Germany). Other routine reagents and chemicals used for the study were of analytical grade.

### Collection of tiger nut tubers, identification and preparation of extract

Fresh yellow tubers of *C. esculentus* (Figure 1) were collected from



**Figure 1.** Picture showing yellow and fleshy tubers of *C. esculentus* Linn (tiger nuts).

Nsukka in Enugu State, Nigeria, in March 2016. The tubers were identified by a Botanist (Mr. A.O. Ozioko) at the Department of Botany, University of Nigeria, Nsukka. They were washed, allowed to dry under shade, and ground into powder. Eighty percent (80%) methanol was added to 1000 g of the powdered tubers and the resulting mixture was shaken intermittently at 2 h interval for 48 h. After that period, the mixture was filtered and concentrated to dryness using a rotary evaporator (Buchi, Switzerland), and referred to as *C. esculentus* methanol tuber extract (CEME).

#### Experimental animals

Thirty adult male albino rats (*Rattus norvegicus*) weighing between 200-250 g, were used for the study. They were kept in clean cages in the laboratory Animal House of the Department of Veterinary Physiology and Pharmacology, University of Nigeria, Nsukka, at room temperature of 23- 29°C, and acclimatized for 2 weeks before the commencement of the study. The albino rats were fed commercial rat pellets; product of Grand Cereals Nig. Ltd, Jos, Nigeria, and clean drinking water *ad libitum*. The ethical guidelines governing the use of animals for laboratory experiment were strictly adhered to (Zimmermann, 1983; Ward and Elsea, 1997). The protocol for the laboratory animal study was approved by the Faculty of Veterinary Medicine Experimental Animal Ethics Committee, University of Nigeria, Nsukka.

#### Evaluation of the effects of CEME on CCl<sub>4</sub>-induced sub-acute liver damage

The albino rats were randomly assigned to six groups (A – F) of five rats per group. Sub-acute liver damage was induced in rats

in Groups A – E by intra-peritoneal injection of 1 ml/kg body weight CCl<sub>4</sub> in equal volume of olive oil (50 % volume/volume) on day 0, and after every 72 h (3 days) for 12 days (Singh et al., 2012). Group A rats were given 10 ml/kg distilled water as placebo (negative control), Groups B, C and D rats were treated with 200, 400 and 800 mg/kg CEME respectively, Group E rats were treated with 100 mg/kg Silymarin (a known hepatoprotective drug) as positive control, Group F rats were given 10 ml/kg distilled water as placebo (normal control). The treatment with CEME and Silymarin started a day after the initial CCl<sub>4</sub> injection and was done orally twice daily for 15 days. On day 15, blood samples for clinical chemistry assay were collected from the albino rats using the orbital technique (Bolliger and Everds, 2010).

The blood samples were allowed to stand at room temperature for 45 min to clot, they were then centrifuged at 3000 revolutions per minute for ten minutes using a table centrifuge (Jenalab Medical, England). The serum was harvested and used immediately for serum biochemistry assay following standard procedures.

The serum ALT and AST activities were determined using the QCA ALT and AST test kits, based on the Reitman and Frankel colorimetric method (Reitman and Frankel, 1957; Colville, 2002). The ALT in the serum sample and standard catalyzed the reaction of L-alanine and alpha-ketoglutaric acid to form pyruvic acid and L-glutamic acid, while the AST catalyzed the reaction of L-aspartic acid with alpha- ketoglutaric acid to form oxaloacetic acid and L-glutamic acid. These ketonic acids produced were reacted with 2-4, dinitrophenyl hydrazine to form a corresponding coloured hydrazone. The optical density of the coloured hydrazone was then measured at 505 nm wavelength using the semi-automated analyzer, and ALT/AST activity quantified. Serum ALP activity was assayed using the QCA alkaline phosphatase test kit, which is based on the phenolphthalein monophosphate method (Klein et al, 1960; Colville, 2002). In the method, alkaline phosphatase in the

**Table 1.** The ALT, AST and ALP activities of rat groups\* given sub-acute toxic doses of CCl<sub>4</sub> and treated with varied doses of CEME.

Group	Means ± standard error		
	ALT (IU/L)	AST (IU/L)	ALP (IU/L)
Group A	130.22±30.87 <sup>a</sup>	177.23±34.39 <sup>a</sup>	262.64±16.22 <sup>a</sup>
Group B	71.33±26.55 <sup>b</sup>	89.38±9.95 <sup>b</sup>	225.59±10.64 <sup>ab</sup>
Group C	73.57±18.71 <sup>b</sup>	111.31±7.59 <sup>b</sup>	228.82±15.95 <sup>ab</sup>
Group D	51.25±7.18 <sup>b</sup>	96.44±8.47 <sup>b</sup>	192.02±32.51 <sup>ab</sup>
Group E	58.49 ±10.41 <sup>b</sup>	110.17±11.59 <sup>b</sup>	182.30±20.79 <sup>b</sup>
Group F	30.13±2.00 <sup>b</sup>	54.09±1.10 <sup>c</sup>	187.70±23.68 <sup>b</sup>

<sup>a, b, c</sup> Different alphabetical superscripts in a column indicate significant differences between the groups,  $P < 0.05$ . \* Group treatments: Group A – CCl<sub>4</sub> + distilled water placebo; Group B - CCl<sub>4</sub> + 200 mg/kg CEME; Group C - CCl<sub>4</sub> + 400 mg/kg CEME; Group D - CCl<sub>4</sub> + 800 mg/kg CEME; Group E - CCl<sub>4</sub> + 100 mg/kg Silymarin; Group F - Distilled water placebo only.

serum and a standard (containing 30 IU/L alkaline phosphatase) hydrolyzed a colourless substrate of phenolphthalein monophosphate giving rise to phosphoric acid and phenolphthalein which at alkaline pH turned into a pink colour. The optical density of the pink coloured solution was measured at 546 nm wavelength using the semi-automated analyzer, and the alkaline phosphatase activity quantified.

Serum total protein levels were determined using the QCA total protein test kit based on the direct Biuret method (Lubran, 1978; Johnson, 2008). This procedure involved a reaction of the proteins in the serum samples and a standard (containing 5 g/dl of proteins) with copper ions in the Biuret reagent in an alkaline medium, which resulted in the formation of a stable coloured complex. The optical density of the coloured complex was measured at 546 nm wavelength using the semi-automated analyzer, and the serum total protein quantified. The serum albumin was assayed using the QCA albumin test kit, which is based on the bromocresol green method (Doumas and Peters, 1997; Johnson, 2008). This procedure involved the reaction of the albumin in the serum samples and standard (containing 5 g/dl of albumin) with bromocresol reagent at acid pH to form a coloured complex. The optical density of the coloured complex was measured at 630 nm wavelength using the semi-automated analyzer, and serum albumin level quantified. The globulin levels were calculated by subtracting the serum albumin levels from the total protein levels (Johnson, 2008). The serum total cholesterol levels were determined using the QCA total cholesterol test kit, which is based on the enzymatic colorimetric method (Allain et al., 1974; Rifai et al., 2008). In this procedure, total cholesterol in the serum samples and a standard (containing 200 mg/dl of cholesterol) was enzymatically hydrolyzed by cholesterol esterase and further oxidized by cholesterol oxidase contained in the QCA total cholesterol working reagent. The reactions led to formation of a coloured quinonic derivative. The optical density of the coloured quinonic solution was measured at 505 nm wavelength using the semi-automated analyzer, and total serum cholesterol quantified. The serum total bilirubin levels in the serum samples were assayed using the Randox<sup>®</sup> bilirubin test kit (Randox Laboratories Ltd, County Antrim, United Kingdom), which is based on the Jendrassik and Grof method (Doumas et al., 1973; Higgins et al., 2008). In this determination, the serum samples were reacted with diazotized sulfanilic acid in the presence of caffeine to produce an azopigment, and their optical densities were measured at 578 nm using the semi-automated analyzer, and the total serum bilirubin quantified. After blood sample collection, the rats were sacrificed by euthanizing them with intra-peritoneal injection of 250 mg/kg thiopentone sodium and confirmatory exsanguinations (AVMA,

2013). The liver of each rat was carefully dissected and weighed, and the relative liver weight was calculated.

#### Data analysis

The clinical biochemistry data were subjected to one way analysis of variance (ANOVA) using the SPSS software (version 16). The least significant difference procedure was used to separate variant means, post-hoc. Probability less than 0.05 was accepted as significant, and a summary of the results were presented as means ± standard error, in tables.

## RESULTS

### Plant extraction

Plant extraction yielded golden brown coloured CEME, which was soluble in water with percentage yield of 25.43% weight/weight.

### Effects of CEME on CCl<sub>4</sub>-induced sub-acute liver damage

The mean serum ALT activity of the Group A rats was more than four times that of Group F rats, while that of Groups B, C, D and E rats were about 1.5 times to 2 times that of Group F rats (Table 1). The serum ALT activity of the Groups B, C, D and E rats were significantly lower ( $P < 0.05$ ) than that of Group A, and were not significantly different ( $P > 0.05$ ) from that of Group F rats (Table 1). The mean serum AST activity of the Group A rats was more than 3 times that of Group F rats, while that of Groups B, C, D and E were about double that of Group F (Table 1). The serum AST of Groups B, C, D and E were significantly lower ( $P < 0.05$ ) than that of Group A, and were also significantly higher ( $P < 0.05$ ) than that of Group F rats (Table 1). The mean serum ALP activity of the Group A rats was significantly

**Table 2.** Levels of serum proteins in rat groups\* given sub-acute toxic doses of CCl<sub>4</sub> and treated with varied doses of CEME.

Group	Means ± standard error		
	Total proteins (g/dl)	Albumins (g /dl)	Globulins (g/dl)
Group A	5.87±0.20 <sup>a</sup>	2.92±0.30 <sup>a</sup>	2.95±0.33 <sup>a</sup>
Group B	5.95±0.31 <sup>a</sup>	3.31±0.29 <sup>ab</sup>	2.64±0.23 <sup>a</sup>
Group C	6.37±0.30 <sup>ab</sup>	3.60±0.20 <sup>ab</sup>	2.77±0.20 <sup>a</sup>
Group D	6.47±0.06 <sup>ab</sup>	3.45±0.16 <sup>ab</sup>	3.01±0.17 <sup>a</sup>
Group E	6.79±0.36 <sup>b</sup>	3.53±0.27 <sup>ab</sup>	3.26±0.22 <sup>a</sup>
Group F	6.73±0.24 <sup>b</sup>	3.72±0.15 <sup>b</sup>	3.02±0.12 <sup>a</sup>

<sup>a, b, c</sup> Different alphabetical superscripts in a column indicate significant differences between the groups,  $P < 0.05$ . \* Group treatments: Group A – CCl<sub>4</sub> + distilled water placebo; Group B - CCl<sub>4</sub> + 200 mg/kg CEME; Group C - CCl<sub>4</sub> + 400 mg/kg CEME; Group D - CCl<sub>4</sub> + 800 mg/kg CEME; Group E - CCl<sub>4</sub> + 100mg/kg Silymarin; Group F - Distilled water placebo only.

**Table 3.** Serum levels of total cholesterol and bilirubin, and the relative liver weight of rat groups\* given sub-acute toxic doses of CCl<sub>4</sub> and treated with varied doses of CEME.

Group	Means ± standard error		
	Total cholesterol (mg/dl)	Total Bilirubin (mg/dl)	Relative liver weight
Group A	89.54±10.42 <sup>a</sup>	2.58±0.10 <sup>a</sup>	4.23 ±0.14 <sup>a</sup>
Group B	86.42±7.51 <sup>a</sup>	2.24±0.03 <sup>b</sup>	4.05 ± 0.14 <sup>ab</sup>
Group C	81.95±5.18 <sup>a</sup>	2.36±0.12 <sup>ab</sup>	3.70 ± 0.15 <sup>b</sup>
Group D	84.54±6.96 <sup>a</sup>	2.17±0.10 <sup>b</sup>	3.78 ± 0.05 <sup>b</sup>
Group E	87.38±5.35 <sup>a</sup>	2.29±0.12 <sup>ab</sup>	3.74 ± 0.13 <sup>b</sup>
Group F	73.60±5.07 <sup>a</sup>	1.46±0.06 <sup>c</sup>	3.31 ± 0.09 <sup>c</sup>

<sup>a, b, c</sup> Different alphabetical superscripts in a column indicate significant differences between the groups,  $P < 0.05$ . \* Group treatments: Group A – CCl<sub>4</sub> + distilled water placebo; Group B - CCl<sub>4</sub> + 200 mg/kg CEME; Group C - CCl<sub>4</sub> + 400 mg/kg CEME; Group D - CCl<sub>4</sub> + 800 mg/kg CEME; Group E - CCl<sub>4</sub> + 100mg/kg Silymarin; Group F - Distilled water placebo only.

higher ( $P < 0.05$ ) than that of Groups E and F, but there were no significant differences ( $P > 0.05$ ) between the serum ALP activity of the treated groups (B, C and D) and all other groups (Table 1).

The mean serum total protein levels of the rats in Groups A and B were significantly lower ( $P < 0.05$ ) than that of rats in Groups E and F, but there were no significant differences ( $P > 0.05$ ) between the serum total proteins of rats in Groups C, D and other rat groups (Table 2). The mean serum albumin levels of rats in group A were significantly lower ( $P < 0.05$ ) than that of rats in Group F, but there were no significant differences ( $P > 0.05$ ) between the serum albumin levels of rats in Groups B, C, D and E and all other groups (Table 2). There were no significant variations ( $P > 0.05$ ) between the serum globulin (Table 2) and total cholesterol (Table 3) levels of rats in all the groups. The mean serum total bilirubin levels of the Group A rats was significantly ( $P < 0.05$ ) higher than that of Groups B, D and F, while that of rats in Group F was significantly lower ( $P < 0.05$ ) than that of all other groups (Table 3). The relative liver weight of the Group A rats was significantly higher ( $P < 0.05$ )

than that of rats in Groups C, D, E and F, while that of rats in Group F was significantly lower ( $P < 0.05$ ) than that of rats in all other groups (Table 3).

## DISCUSSION

The comparatively higher serum enzyme activity of ALT, AST and ALP in all the groups that were given CCl<sub>4</sub> shows that CCl<sub>4</sub> induced liver damage by altering the integrity of the hepatocytes and affecting liver function adversely (Mukherjee, 2003; Kim et al., 2010). The alteration of serum ALT, AST and ALP activity levels implied that CCl<sub>4</sub> caused damage to the liver hepatocytes. Elevation in serum transaminases (AST and ALT) is a biomarker of hepatocellular necrosis and hepatotoxicity (Friedman et al., 1996). The elevated levels of ALT, AST and ALP as recorded in this study validates the reports of Nishigaki et al. (1992), Ameen et al. (1999), Raja et al. (2007), Shafaq et al. (2009) and Oyedepo and Odoje (2014), who also reported elevated levels of these hepatic enzymes in serum of albino rats

given hepatotoxic doses of CCl<sub>4</sub>.

The administration of CEME (at all doses) protected liver function of the rats whose livers were damaged with CCl<sub>4</sub> by protecting their hepatocellular integrity, and in this case CEME administered at 800 mg/kg, compared favourably with Silymarin (a standard hepatoprotective drug). The ability of administered CEME to protect hepatocellular integrity as recorded in this study agrees with the reports of Ameen et al. (1999), Farok et al. (2011) and Onuoha et al. (2017) who reported significant decreases in ALT and AST in rats treated with oily extracts of *C. esculentus* tuber, aqueous extract of *C. esculentus* tuber, and tiger nut milk, respectively. Oyedepo and Odoje (2014) reported a marked decline in ALT, AST and ALP in rats pretreated with varying percentages of tiger nut flour in rat pellets for 21 days preceding CCl<sub>4</sub> administration. *In vitro* studies on the hepatoprotective activity of solvent-free microwave extract of *C. esculentus* also showed that the IC<sub>50</sub> of the essential oil content of *C. esculentus* tubers on monolayers of rat hepatocytes was > 1000 µg/ml, and exhibited hepatoprotection at 18.5 µg/ml (Hassanein et al., 2011). The ability of CEME to protect hepatocellular integrity in this present study may be due to hepatocyte membrane stabilization by active phytochemicals like flavonoids and water soluble glycosides which are reported constituents of *C. esculentus* tubers (Temple et al. 1990; Eteshola and Oraedu, 1996; Oloyede et al., 2014), and which are well known natural antioxidants (Temple et al., 1990; Pietta, 2000; Satoh et al., 2004; Oloyede et al., 2014). Since free radicals play important role in CCl<sub>4</sub>-induced liver damage, it is believed that compounds that neutralize such radicals may have hepatoprotective properties. Other natural products that possess antioxidant properties have also been reported to protect against CCl<sub>4</sub>-induced hepatotoxicity (Hsiao et al., 2003).

The depletion of serum total proteins and albumins in rats that were given CCl<sub>4</sub> is also an indication of liver dysfunction associated with CCl<sub>4</sub> administration (Navarro and Senior, 2006). Serum albumin is the major protein in the blood synthesized by the liver. It is a clinically useful marker of hepatic synthetic function (Friedman et al., 1996). The administration of CCl<sub>4</sub> in this study adversely affected hepatic synthesis of albumins, and treatment with CEME led to slight elevation of serum total protein and albumin of the treated rats. Chukwuma et al. (2010) and Hwang (2004) reported increases in the level of serum total proteins and albumins in rats treated with aqueous extract of *C. esculentus* tuber. Protein synthesis stimulation has been recognized as a hepatoprotective mechanism. It helps to accelerate the process of regeneration and the production of replacement liver cells (Rip et al, 1985; Tadeusz et al., 2001).

The lack of significant variations in the serum globulin, and total cholesterol levels among the groups shows that

CCl<sub>4</sub> administration and CEME treatment had no effects on these parameters. The significantly lower serum bilirubin of rats in groups B and D suggests that treatment with 200 and 800 mg/kg enhanced hepatic excretion of bilirubin which was adversely affected by CCl<sub>4</sub> administration. Bilirubin is a diagnostic marker of liver and blood disorders, it is the end product of the breakdown of haemoglobin (Singh et al., 2011). Damage to the liver cells causes impairment of bilirubin excretion, thus causing accumulation of bilirubin in the blood and extracellular fluid (Singh et al., 2011). The effect of CEME administration on serum total bilirubin in this present study agrees with the reports of Amani et al. (2012) on extracts of a related plant, *C. alternifolius*. The higher relative liver weight of rats in groups A and B shows that the liver was enlarged as a result of inflammation due to damage to the liver cells caused by CCl<sub>4</sub> (Bukhsh et al., 2014), and the results obtained from this study showed that treatment with the CEME at 400 and 800 mg/kg, and Silymarin at 100 mg/kg was able to significantly reduce the inflammatory enlargement which was induced by CCl<sub>4</sub> administration.

## Conclusion

From the results obtained from this study, it was concluded that the administration of *C. esculentus* methanolic tuber extract to albino rats whose livers were experimentally damaged with CCl<sub>4</sub>, protected hepatocellular integrity, enhanced hepatic bilirubin excretion and ameliorated inflammatory enlargement of the liver, and its hepatoprotective effects compared effectively with a standard hepatoprotective drug (Silymarin). These findings imply that *C. esculentus* methanolic tuber extract is hepatoprotective against carbon tetrachloride-induced liver damage.

## CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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