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Research Article
Microstructural Effects of *Celosia trigyna* Leave Extracts on the Liver and Ileum in Ethanol-induced Toxicity in Adult Wistar Rats

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Abstract

Background and Objective: *Celosia trigyna* is from the family Amaranthaceae and highly consumed by human beings and animals. The leaves are rich in antioxidants and used medicinally to treat wounds, diarrhea and mouth ulcers. The aim of this study was to evaluate the healing potential of *Celosia trigyina* leave extracts on the liver and ileum in ethanol-induced toxicity in adult Wistar rats. **Materials and Methods:** Thirty adult wistar rats were procured, acclimatized and randomly divided into 6 groups of 5 animals each. Rats in groups 2-6 were administered with 5 mL kg^{-1} of 90% ethanol while animals in group 1 were administered with equivalent volume of distilled water. After 24 h, animals in groups 3-6 were administered with leave extracts of *Celosia trigyna* (methanol, dichloromethane, ethyl acetate and hexane, respectively). The therapies went on repeatedly for 14 days in all animal groups. Twenty four h after the last administration, the liver and ileum were excised and fixed in 10% formol saline for histopathological analysis. **Results:** The result showed a significant decrease ($p<0.05$) in the weight difference of animals in group 2 when compared to the *Celosia trigyna* treated groups. Furthermore, the histopathological evaluation showed a remarkable recovery of the necrotized hepatocytes by the leave extracts of *Celosia trigyna* with the dichloromethane and hexane extracts having better remediation against ethanol induced toxicity in both the liver and ileum than the methanol and ethyl acetate extracts. **Conclusion:** In conclusion, the leave extracts of *Celosia trigyna* showed promising protection of the liver and ileum against ethanol-induced toxicity in adult Wistar rats. This protection may be due to the antioxidant potential of *Celosia trigyna.*

Key words: Liver, ileum, toxicity, histopathology, ethanol, antioxidants

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Ethanol toxicity has been on the increase in recent times[1]. Globally, there has been reported abuse of alcohol consumption. Such abuses often lead to severe health conditions such as pancreatitis, diabetes mellitus, gastrointestinal damage, liver disease and cancer[2]. Ethanol toxicity can cause infiltration of inflammatory cells, necrosis of the hepatocytes and in some cases, initiate the development of tumor in rats and mice[3].

Liver damage is a gradual process and as such may not be noticeable at the onset[4]. Ethanol has caused more harm than good as available reports showed that 20% of all liver transplants in United States are traceable to liver alcohol disease[4]. Ethanol consumption is attributed to 6.5% of all deaths in Europe[5,6]. Alcohol dependence syndrome a disorder associated with preoccupation with alcohol, adaptability to the psychotropic effects of alcohol consumption and continuous consumption of alcohol despite its deleterious effects is responsible for the alcohol-related organ damage in most patients[5]. Direct interaction of ethanol with mucosa of the gastrointestinal tracts can induce functional and metabolic changes. This interaction often leads to mucosal damage culminating into diarrhea and gastrointestinal bleeding. Functional changes and cellular damage to the mucosal affect digestion and assimilation of nutrients leading to weight loss due to malnutrition[7]. Alcohol ingestion reduces the absorption of sodium in the jejunum and ileum[8,9].

Ethanol consumption has been linked with free radical generation which can react with cellular macromolecules to cause cellular damage[10]. Antioxidants can protect the body against free radicals inflicted injury. There are basically two types of antioxidants: Enzymatic and non-enzymatic[5]. Enzymatic antioxidants such as catalase and glutathione peroxidase are capable of enhancing non enzymatic antioxidants such as vitamin E and ascobate[5]. Herbal therapies rich in antioxidants can mop up free radicals generated by alcohol thus, protecting body organs from oxidative damage. *Celosia trigyna* is from the family Amaranthaceae. It is found in some African countries such as South Africa, Democratic Republic of Congo and Nigeria. Investigation has shown that both human being and animals consume the leaves of *Celosia trigyna.* The leaves can be used medicinally to treat wounds, diarrhea, mouth ulcers and intestinal worms[11,12]. Just very recently, Ofusori *et al.*[13] reported the antiulcerogenic effects of *Celosia trigyna* plant extracts on ethanol induced gastric ulcer in adult Wistar rats.

In view of the high consumption rate of alcohol, various health challenges that accompany its use as well as the need to search for relatively more affordable and tolerable plant-derived alternatives, it was imperative to study the histopathological effects of different leave extracts of *Celosia trigyna* on the liver and ileum following ethanol toxicity with a view to ascertaining their ameliorative potentials.

MATERIALS AND METHODS

This study was conducted between May-October, 2018 at the Department of Anatomy and cell Biology, College of Health Sciences, Obafemi Awolowo University, Ile-Ife, Nigeria.

Plant materials: *Celosia trigyna* leaves were procured and authenticated by a taxonomist in the herbarium unit of the Department of Botany, Obafemi Awolowo University, Ile-Ife where a voucher specimen (IFE- 17466) was deposited.

Extraction process: The extraction processes were conducted in accordance to standard procedures[14]. All the leaves were air-died and grinded into powder using the squeezing and crushing machine (Daiki Rika Kogyo Co-ltd, Japan). The powdered product was percolated with methanol, dichloromethane (DCM), ethyl acetate and hexane and subjected to continuous shaking for a period of 48 h. The solution was filtered with a (Whatman No. 1) filter paper, concentrated with rotary evaporator and stored at 4°C until needed. The 4 extracts (methanol, DCM, ethyl acetate and hexane) were later constituted at a dose of 50 mg kg^{-1} b.wt., with 20% Tween 80.

Animal care: Handling and care of the animals conform to the rules and guidelines of the ethical committee of the Institute of Public Health, Obafemi Awolowo University, Nigeria. Rats were procured from Soyebo farm, Nigeria and acclimatized for one week before randomization and grouping into plastic cages at the Animal Holdings of the Department of Anatomy and Cell Biology, Obafemi Awolowo University, Ile-Ife, Nigeria.

Study design: Thirty adult female Wistar rats were used for the study. The rats were assigned into 6 groups with 5 animals[10] each: 1, 2, 3, 4, 5 and 6:

Group 1: Normal control (DDH_2O-1 mL kg^{-1} b.wt.,/day for 14 days)

Group 2: (Negative control) was given 90% ethanol (5 mL kg^{-1}) and 1 mL kg^{-1} b.wt.,/day of 20% tween 80 for 14 days

Group 3: Was given 90% ethanol (5 mL kg^{-1}) and MeOH extract (50 mg kg^{-1} b.wt.,/day in 20% tween 80 for 14 days)

Group 4: Was given 90% ethanol (5 mL kg^{-1}) and DCM extracts (50 mg kg^{-1} b.wt.,/day in 20% tween 80 for 14 days)

Group 5: Was given 90% ethanol (5 mL kg^{-1}) and $EtOA_C$ extract (50 mg kg^{-1} b.wt.,/day in 20% tween 80 for 14 days).

Group 6: Was given 90% ethanol and hexane extract (50 mg kg^{-1} b.wt.,/day in 20% tween 80 for 14 days)

Ethanol toxicity: At the start of the research, all the rats were fasted for 24 h and then a single dose of 90% ethanol (5 mL kg^{-1}) was administered orally to the test groups (2-6)[10]. Twenty four hours after this, the rats in the test groups (3-6) were commenced on their respective treatment as stated above and administered orally and repeatedly for 14 days before sacrifice.

Sacrifice of animals: Twenty four hours after the last administration, all the rats were sacrificed under slight chloroform anesthesia. Midline incisions were performed and the liver and ileum excised and washed in normal saline.

Histopathological analysis: Histological procedures as documented by Bancroft and Gamble[15] was adopted. A portion of the liver and ileum were fixed in 10% formol saline for histopathological analyses using haematoxylin-eosin (H and E) (for the demonstration of the general histoarchitecture), Verhoeff-van Gieson (VVG) (for the demonstration of elastic fibres) and Goddon and Sweets (for the demonstration of reticular fibres) staining procedures. The tissues were dehydrated through ascending grades of alcohol (50, 70, 90% and absolute), cleared in xylene and infiltrated with paraffin wax before embedding. Five micron sections were obtained on rotary microtome. The sections were subjected to different staining procedures. Thereafter, sections were dehydrated in ascending grades of alcohol, cleared in xylene and mounted in DPX. Digital images were achieved using Leica DM 750 microscope interfaced with Leica ICC_{50} digital camera.

Image analysis: The density of the reticular fibers per group was performed by measuring the stained area per 1600 mm^2 on 5 random spots per animal. The software quantifies staining intensity by measuring the stained area in each of the images. Values for each of the photomicrographs per group which represents the staining intensity were generated which was then run through a statistical package (SPSS 22).

Statistical analysis: Data were expressed as Mean±SEM. Statistical significance between the groups were determined by one way analysis of variance (ANOVA). $p<0.05$ was considered statistically significant.

RESULTS

Weight difference: There was a significant decrease in the body weight difference of animals in group 1 when compared with all other groups (Table 1). There was no significant difference when the weight differences of animals in groups 4-6 were compared across groups. Out of the four groups administered with extracts (groups 3, 4, 5 and 6), only animals in group 3 (administered with methanolic extracts) exhibited a significant ($p<0.05$) decrease in body weight difference when compared with the other three groups (4, 5 and 6) as shown in Table 1.

Histopathological analysis: The histopathological results showed evidence of necrosis of the hepatocytes of animals in group 2. Also, noticeable were neutrophils infiltration and fat droplets when compared with the normal control (Fig. 1). The ileum of rats in group 2 presented epithelial damage and disorganization of the mucosa. Also, the villi were compromised when compared

Table 1: Weights (g) of animals exposed to ethanol toxicity and treated with different extracts of *Celosia trigyna*

Groups	Initial weight	Final weight	Weight difference
1	163.09±5.87^{a}	175.76±5.59^{b}	12.67±1.90^{c}
2	163.92±5.69^{a}	126.93±16.97^{a}	-39.49±13.45^{a}
3	163.68±4.02^{a}	152.26±10.60^{ab}	-11.42±9.19^{b}
4	166.57±15.50^{a}	174.14±12.29^{b}	7.96±5.62^{bc}
5	169.29±13.92^{a}	162.14±10.97^{b}	-7.15±4.23^{bc}
6	165.45±10.76^{a}	169.21±11.70^{b}	5.24±2.06^{bc}

Group 1: Normal control, group 2: Negative control, group 3: Treated with methanol extract, group 4: Treated with dichloromethane extract, group 5: Treated with ethyl acetate extract, group 6: Treated with hexane extract, means with same superscript letters within the column indicate no significant difference while means with the different letters indicate significant difference ($p<0.05$, Duncan *Post hoc* multiple range test comparisons)

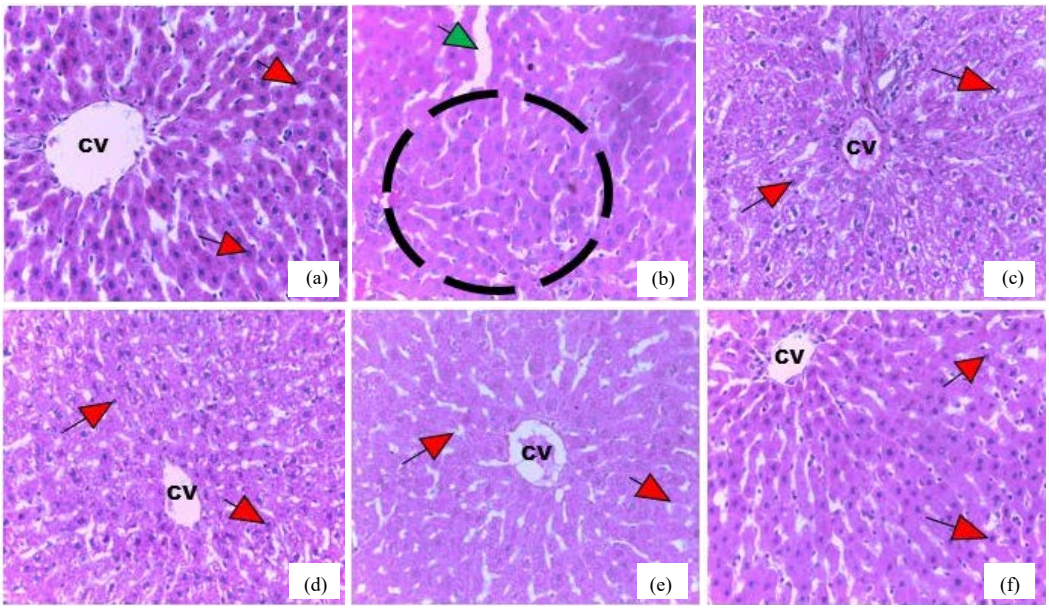

Fig. 1(a-f): Photomicrographs of the Liver in (a) group 1-normal control (intact histoarchitecture), (b) Group 2-negative control (disorientation of the hepatocytes (Black circle) characterized with distortion of the sinusoids (Green arrow), (c) Group 3-treated with methanol extract (note the partly disorganized histoarchitecture), (d) Group 4-treated with dichloromethane extract (note the similarity in the histoarchitecture compared with group 1), (e) Group 5-treated with ethyl acetate extract (note the partly disorganized histoarchitecture) and (f) Group 6-treated with hexane extract (note the similarity in the histoarchitecture compared with group 1 and 4)
CV: Central vein, Red arrow: Hepatocyte, H and E 100×

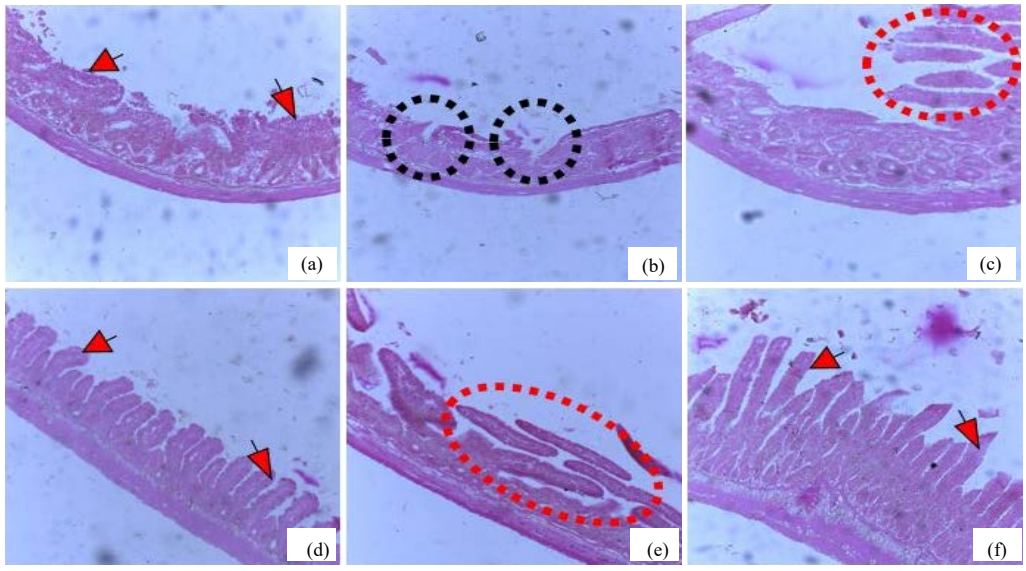

Fig. 2(a-f): Photomicrographs of the Ileum in (a) Group 1-normal control (arrow showing intact epithelium), (b) Group 2-negative control (disruption of the epithelium-Black circle), (c) Group 3-treated with methanol extract (note the pattern of orientation and sloughed villi) (Red circle), (d) Group 4-treated with dichloromethane extract (intact epithelium) (arrow), (e) Group 5- treated with ethyl acetate extract (note the pattern of orientation and sloughed villi) (Red circle) and (f) Group 6-treated with hexane extract (note the intact epithelium)
H and E 400×

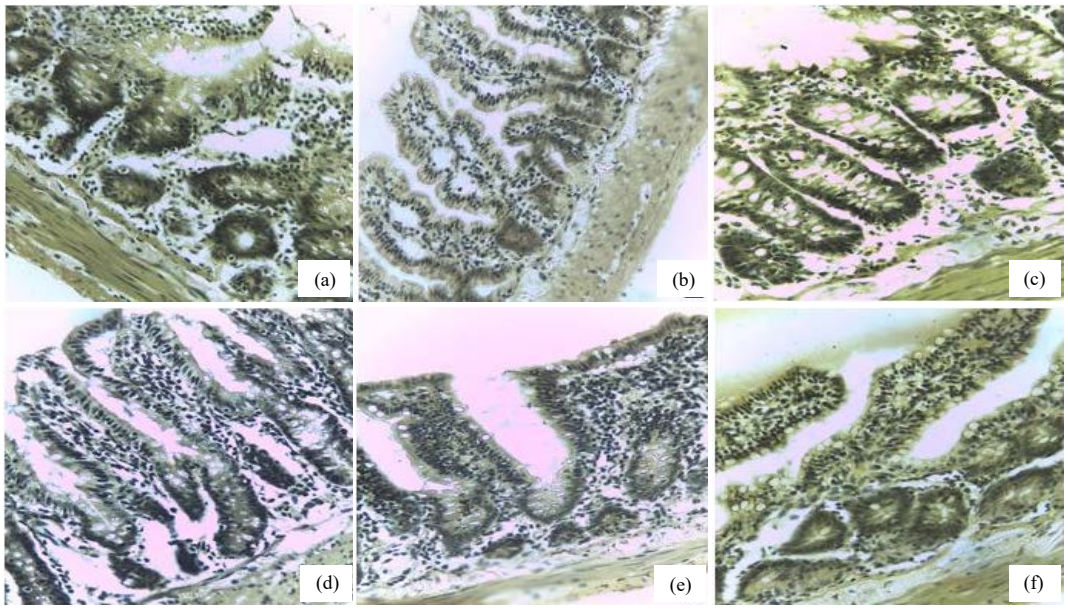

Fig. 3(a-f): Photomicrographs of the Ileum in (a) Group 1-normal control, (b) Group 2-negative control, (c) Group 3-treated with methanol extract, (d) Group 4-treated with dichloromethane extract, (e) Group 5-treated with ethyl acetate extract and (f) Group 5-treated with hexane extract
Elastic fiber: Black VVG 1000×

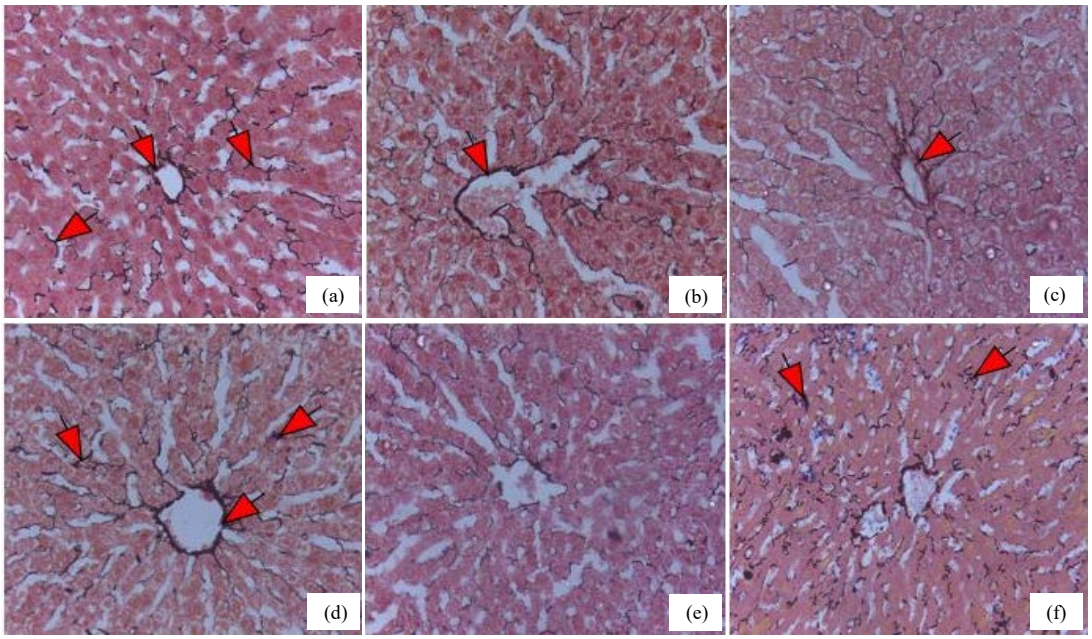

Fig. 4(a-f): Photomicrographs of the liver in (a) Group 1-normal control, (b) Group 2-negative control, (c) Group 3-treated with methanol extract, note the scanty reticular fibers, (d) Group 4-treated with dichloromethane extract, (e) Group 5-treated with ethyl acetate extract, Note the scanty reticular fibers and (f) Group 6-treated with hexane extract, note the reticular fibres within the perisinusoidal space and around the wall of the central vein in groups 1, 4 and 6
Reticular fibres: Black Goddon and Sweets 100×

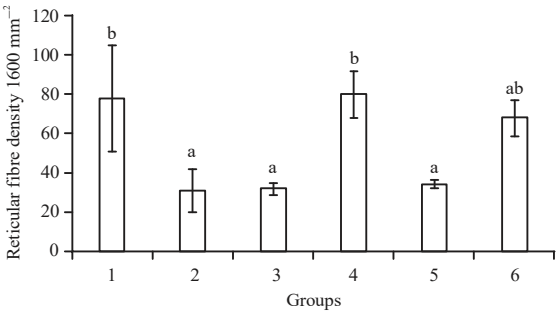

Fig. 5: Mean reticular fibre density 1600 mm^{-2}

Values are given as Mean±SEM, means with different letters differs significantly at p<0.05 while means with the same letters does not differ significantly at p<0.05, Group 1: Normal control, group 2: Negative control), group 3: Treated with methanol extract, group 4: Treated with dichloromethane extract, group 5 Treated with ethyl acetate extract, group 6: Treated with hexane extract

with group 1 (Fig. 2). There were various degrees of remediation in the groups treated with the leave extracts of *Celosia trigyna.* Remarkable improvements were noticed in the groups treated with DCM and hexane extracts.

Elastic fibers were appreciably deposited in the mucosa and muscularis externa of the ileum in groups 4 and 6 which is comparable with group 1. There were only mild traces of elastic fibers in group 2, 3 and 5 when compared with group 1 (Fig. 3).

The reticular fibers were sparsely stained in group 2 animals when compared with normal control (Fig. 4). There were improvement in the deposition of reticular fibers in groups 4 and 6 when compared with groups 3, 5 (Fig. 4). The reticular fibers in the untreated group was significantly lower ($p<0.05$) when compared with the normal control. The groups treated with methanol and ethyl acetate extracts were significantly different when compared with the normal control but the group treated with DCM extract was not significantly different from the normal control. The untreated group and the groups treated with methanol and ethyl acetate were not significantly different from each other as shown in Fig. 5.

DISCUSSION

The liver and ileum are very vital organs in the body. While the liver sub serve so many functions among which is: Metabolism, detoxification and production of bile, the ileum on the other hand, is involved in the absorption of vitamin B_{12}, bile salt and every product of digestion that escapes the jejunum. The abuse of ethanol has been on the increase worldwide thus leading to alcoholic liver disease[5]. Alcoholic liver disease is the leading cause of morbidity and mortality as a result of liver failure, liver cancer and cirrhosis[5]. Ethanol can induced oxidative stress and by so doing, impair the antioxidant defense mechanism[16]. When the antioxidant defense mechanism is compromised, this can lead to cellular damage[10]. Cellular damage to the mucosal affect digestion and assimilation of nutrients leading to weight loss due to malnutrition[7] as observed in group 2 of this study. In a similar fashion, Olaibi *et al.*[17] in their work on the histomorphometric study of stomach and duodenum of aspirin treated Wistar rats concluded that weight loss could be due to decline digestive functions as a result of severe injuries by aspirin administration to the gastrointestinal mucosa. There was an improvement in the weight difference of the extract treated groups, this may be due to phytochemicals and antioxidant potentials of *Celosia trigyna* as reported by Ofusori *et al.*[18]. *Celosia trigyna* contains proteins, amino acids, minerals and vitamins[19]. In addition, *Celosia trigyna* is highly rich in magnesium, zinc, calcium, iron and vitamins A, B, C, E, K[20,21]. Herbal therapies that contain high level of antioxidants are capable of safeguarding body viscera from oxidative damage by reducing free radicals produced by toxicants such as ethanol. Antioxidants are found in majority of herbs consumed by man and animals. In most cases, the antioxidant component of a particular plant may have to work in concert in order to achieve a medicinal potential.

The hepatocellular damage characterized by steatosis, neutrophils infiltration and fat droplets in the liver of the untreated group were gradually been restored to near normal in the extracts treated groups when compared with the normal control. The extracts of *Celosia trigyna* ameliorated the epithelial damage and disorganization of ileum mucosa when compared with group 1. Better remediation was achieved in the groups treated with DCM and hexane extracts than those treated with methanol and ethyl acetate extracts. This observation is similar to the pattern of events in the study documented by Ofusori *et al.*[13], where they observed that the DCM and hexane extracts ameliorated the gastric lesion induced by ethanol better than the methanol and ethyl acetate extracts. The reason for the effectiveness of DCM and hexane extracts may be due to the rich antioxidant properties. Ofusori *et al.*[13] isolated the following compounds: Lutein, chondrillasterol acetate, chondrillasterol and pheophytin a from the combined hexane and DCM extracts of *Celosia trigyna*. Lutein which is the most abundant constituent of *Celosia trigyna*[13] has a wide range of beneficial activities which ranges from antioxidant, antihypertensive, antiulcer, antidiabetic and anti-inflammatory and antiatherogenic[22-25]. According to Serpeloni *et al.*[26], lutein scavenge reactive species and cause gene expression that is needed for improved antioxidant response, thereby enhancing oxygen transport. The positive influence of lutein in the redox state of cells could be due to its antigenotoxic and antioxidant effects.

The elastic fibres are very important in the normal functions of the gastrointestinal tract. They help in the resilient rebound of the stretched wall of the gastrointestinal tract. The gradual deposition of the elastic fibres in the muscularis externa of the ileum of the extract treated groups as against the scanty deposit in the untreated group is suggestive of enhanced mechanical movement necessary to propel the chyme in corroboration with Ofusori *et al.*[27].

Reticular fibres are mesh-like in nature and predominant in viscera such as liver. They provide supportive mechanism for the liver[28]. Some medical conditions or toxicants can elicit hepatic fibrosis. These conditions, when present, leads to the depletion of reticular fibres (a pathological feature) common in hepatic fibrosis[29]. A significant improvement observed in the deposition of reticular fibres within the perisinusoidal space and around the wall of the central vein in the extract treated groups is an indication that *Celosia trigyna* has the capability of halting the advancement of hepatic fibrosis. The mechanism for achieving this may be through the amelioration of hepatocellular degeneration caused by ethanol administration. Ofusori *et al.*[13] reported a significant cellular turn over after the administration of *Celosia trigyna* in ethanol induced gastric lesion. The synergic effects of these isolated compounds (chondrillasterol, lutein, chondrillasterol acetate and pheophytin a) may have as well been responsible for these observations as reported by Ofusori *et al*[13]. Alcohol liver disease is the cause of most death in adult consumers and it is characterized by inconstant degrees of cirrhosis, inflammation and progressive fibrosis[30]. Cubero *et al.*[31] confirmed that hepatic stellate cells, the cell type implicated in liver fibrosis are always triggered by toxins such as alcohol due to the elicitation of reactive oxygen species-induced damage as observed in this study. Before the development of alcoholic cirrhosis (the end-stage liver disease), abnormalities such as alcoholic hepatitis, steatosis and hepatic fibrosis are the observable features[32] characterized by decrease in reticular fibres as noticed in this study. The administration of graded doses of aqueous extract of *Camellia sinensis* by Lodhi *et al.*[33] in ethanol-induced liver damage led to regeneration of the hepatocytes with little or no evidence of microvesicular steatosis in a similar fashion displayed in this study. Stickel *et al.*[5] and Stickel[34] called for great concern in the growing incidence of hepatocellular carcinoma which occurs in about 1-2% of alcoholic cirrhotics per year. Ethanol consumption is deleterious to the liver and ileum as seen in this study and should be avoided or taken with caution, herbal products such as *Celosia trigyna* is capable of restoring the compromised viscera to near normal and thus recommended for further studies aimed at corroborating our findings.

CONCLUSION

It can be concluded that there were improvements in the histoarchitectural outline of the liver and ileum in the extract treated groups when compared with the untreated group. These improvements were more pronounced in the groups treated with DCM and hexane extracts than those treated with methanol and ethyl acetate extracts. The reason for this observation may be due to the rich phytochemical compounds present in the DCM and hexane extracts. The extract of *Celosia trigyna* has demonstrated a very good potential in ameliorating the degenerative changes in the liver and ileum of ethanol-induced toxicity in adult Wistar rats.

SIGNIFICANCE STATEMENT

This study discovered that the leave extracts of *Celosia trigyna* is capable of ameliorating ethanol induced toxicity in the liver and ileum of rats, with the DCM and hexane extracts demonstrating better remediation. Therefore, *Celosia trigyna* can be beneficial in the management of ethanol-induced complications. This study will assist researchers in the development of alternative therapeutic approach in the management of alcohol related disorders.

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REFERENCES

1. Watabiki, T., Y. Okii, T. Tokiyasu, S. Yoshimura and M. Yoshida *et al.*, 2000. Long-term ethanol consumption in ICR mice causes mammary tumor in females and liver fibrosis in males. Alcoholism: Clin. Exp. Res., 24: 117S-122S.
2. Beland, F.A., R.W. Benson, P.W. Mellick, R.M. Kovatch, D.W. Roberts, J.L. Fang and D.R. Doerge, 2005. Effect of ethanol on the tumorigenicity of urethane (ethyl carbamate) in $B6C3F_1$ mice. Food Chem. Toxicol., 43: 1-19.
3. Hassan, S.M.A., A.K. Saeed and A.J. Hussein, 2015. Ethanol-induced hepatic and renal histopathological changes in BALB/c mice. J. Nat. Sci. Res., 5: 12-20.

4. Marcin, J., 2018. What's to know about alcoholic liver disease? https://www.medicalnewstoday.com/articles/215638.php
5. Stickel, F., C. Datz, J. Hampe and R. Bataller, 2017. Pathophysiology and management of alcoholic liver disease: Update 2016. Gut Liver, 11: 173-188.
6. WHO., 2010. European status report on alcohol and health 2010. World Health Organization, WHO Regional Office for Europe, Copenhagen, Denmark.
7. Bode, C. and J.C. Bode, 1997. Alcohol's role in gastrointestinal tract disorders. Alcohol Health Res. World, 21: 76-83.
8. Bode, C. and J.C. Bode, 1992. Alcohol Malnutrition and the Gastrointestinal Tract. In: Nutrition and Alcohol, Watson, R.R. and B. Watzl (Eds.). CRC Press, Boca Raton, FL., USA., ISBN-13: 9780849379338, pp: 403-428.
9. Pfeiffer, A., T. Schmidt, N. Vidon, C. Pehl and H. Kaess, 1992. Absorption of a nutrient solution in chronic alcoholics without nutrient deficiencies and liver cirrhosis. Scand. J. Gastroenterol., 27: 1023-1030.
10. Omojola, T.A., D.A. Ofusori, S.S. Edward, U. Umeaku, 2019. The histochemical and biochemical effects of coconutmilk in wistar rats with ethanol-induced gastric ulcer. Anat. J. Afri., 8: 1558-1569.
11. Okujagu, T.F., S.O. Etatuvie, I. Eze, B. Jimoh, C. Nweke and C. Mbaoji, 2006. Medicinal plants of Nigeria; South-West Nigeria. Collaboration with Lagos State Traditional Medicine Board and Botany Department, University of Lagos, Nigeria.
12. Mabona, U. and S.F. van Vuuren, 2013. Southern African medicinal plants used to treat skin diseases. S. Afr. J. Bot., 87: 175-193.
13. Ofusori, A.E., R. Moodley and S.B. Jonnalagadda, 2019. Antiulcerogenic effects of *Celosia trigyna* plant extracts on ethanol-induced gastric ulcer in adult Wistar rats. J. Tradit. Complement. Med., (In Press). 10.1016/j.jtcme.2019.11.004.
14. Zhang, Q.W., L.G. Lin and W.C. Ye, 2018. Techniques for extraction and isolation of natural products: A comprehensive review. Chin. Med., Vol. 13. 10.1186/s13020-018-0177-x.
15. Bancroft, J.D. and M. Gamble, 2008. Theory and Practice of Histological Techniques. 6th Edn., Elsevier Health Sciences, Philadelphia, PA., ISBN-13: 9780443102790, Pages: 725.
16. Albano, E., 2006. Alcohol, oxidative stress and free radical damage. Proc. Nutr. Soc., 65: 278-290.
17. Olaibi, O.K., O.M. Ijomone and A.J. Ajibade, 2014. Histomorphometric study of stomach and duodenum of aspirin treated Wistar rats. J. Exp. Clin. Anat., 13: 12-16.
18. Ofusori, A.E., R. Moodley and S.B. Jonnalagadda, 2019. Elemental distribution in the edible leaves of *Celosia trigyna* from the Western and Northern regions of Nigeria. J. Environ. Sci. Health Part B, 54: 61-69.
19. Smith, F.I. and P. Eyzaguirre, 2007. African leafy vegetables: Their role in the world health organization's global fruit and vegetables initiatives. Afr. J. Food Agric. Nutr. Dev., 7: 1-9.
20. Grabben, G.J.H. and O.A. Denton, 2004. Plant Resources of Tropical Africa, Volume 2: Vegetables. PROTA Foundation, Wageningen, Netherlands, ISBN: 90-5782-148-6, Pages: 667.
21. Mensah, J.K., R.I. Okoli, J.O. Ohaju-Obodo and K. Eifediyi, 2008. Phytochemical, nutritional and medical properties of some leafy vegetables consumed by Edo people of Nigeria. Afr. J. Biotechnol., 7: 2304-2309.
22. Miyazawa, T., K. Nakagawa, F. Kimura, Y. Nakashima, I. Maruyama, O. Higuchi and T. Miyazawa, 2013. *Chlorella* is an effective dietary source of lutein for human erythrocytes. J. Oleo Sci., 62: 773-779.
23. Johnson, E.J., 2014. Role of lutein and zeaxanthin in visual and cognitive function throughout the lifespan. Nutr. Rev., 72: 605-612.
24. Erdman, Jr. J.W., J.W. Smith, M.J. Kuchan, E.S. Mohn and E.J. Johnson *et al*., 2015. Lutein and brain function. Foods, 4: 547-564.
25. Manayi, A., M. Abdollahi, T. Raman, S.F. Nabavi, S. Habtemariam, M. Daglia and S.M. Nabavi, 2016. Lutein and cataract: From bench to bedside. Crit. Rev. Biotechnol., 36: 829-839.
26. Serpeloni, J.M., I.M. de Syllos Colus, F.S. de Oliveira, A.F. Aissa, A.Z. Mercadante, M.L.P. Bianchi and L.M.G. Antunes, 2014. Diet carotenoid lutein modulates the expression of genes related to oxygen transporters and decreases DNA damage and oxidative stress in mice. Food Chem. Toxicol., 70: 205-213.
27. Ofusori, D.A., E.A., Caxton-Martins, O.O. Komolafe, K.A. Oluyemi and O.A. Adeeyo *et al*., 2008. A comparative study of the ileum in rat (*Rattus norvegicus*), bat (*Eidolon helvum*) and pangolin (*Manis tricuspis*) as investigated using histological method. Int. J. Morphol., 26: 137-141.
28. Junqueira, L.C.U. and J. Carneiro, 2003. Basic Histology: Text and Atlas. 10th Edn., McGraw-Hill/Appleton & Lange, UK., ISBN-13: 9780071378291, pp: 332-343.
29. Oke, G.O., A.A. Abiodun, C.E. Imafidon and B.F. Monsi, 2019. *Zingiber officinale* (Roscoe) mitigates CCl_4-induced liver histopathology and biochemical derangements through antioxidant, membrane-stabilizing and tissue-regenerating potentials. Toxicol. Rep., 6: 416-425.
30. Rehm, J., A.V. Samokhvalov and K.D. Shield, 2013. Global burden of alcoholic liver diseases. J. Hepatol., 59: 160-168.
31. Cubero, F.J., R. Urtasun and N. Nieto, 2009. Alcohol and liver fibrosis. Semin. Liver Dis., 29: 211-221.
32. Prakash, O. and S. Nelson, 2002. Alcohol and liver disease. Ochsner J., 4: 241-244.
33. Lodhi, P., N. Tandan, N. Singh, D. Kumar and M. Kumar, 2014. *Camellia sinensis* (L.) Kuntze extract ameliorates chronic ethanol-induced hepatotoxicity in albino rats. Evidence-Based Complement. Altern. Med., Vol. 2014. 10.1155/2014/787153.
34. Stickel, F., 2015. Alcoholic cirrhosis and hepatocellular carcinoma. Adv. Exp. Med. Biol., 815: 113-130.