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Full Length Research Paper

The assessment of effect of apple cider vinegar on hepatic and renal function of albino Wistar rat

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The aim of this research is to investigate the toxic effect of apple cider vinegar on hepatic and renal function of albino Wistar rats. A total of 22 rats were randomly divided into 5 groups labeled A, B, C, D, and kept in a well-ventilated room. Group A served as and E control and these rats were treated with distilled water. Rats in groups B and C were treated with 2 different doses (1 ml and 2 ml respectively) for 7 days. Rats in groups D and E were treated with 2 different doses (1 ml and 2 ml respectively) for 14 days. The drugs were administered once daily. Animals were sacrificed 24 h after the last treatment. Blood samples were collected into heparinized sample bottles for analysis. There was no significant difference in the serum liver enzymes results obtained when compared to control. There were deaths in both 7 and 14 days of administration. The deaths were higher in those given 2 ml irrespective of duration. Pathological changes in the liver and kidney progressively increased with concentration and duration. This study demonstrates that large amounts of apple cider vinegar when used undiluted has negative effects on the liver and kidney.

Keywords: Apple cider vinegar, renal function, albino Wistar rats, serum liver enzymes

INTRODUCTION

The name 'vinegar' is derived from the French words 'vin aigre' which translates as 'sour wine. It can be easily sourced from any fermentable carbohydrate source such as dates, grains, apples, grapes, amongst others. Apple cider vinegar contains primarily organic acids and phenolic compounds. Examples of the organic acids are: acetic acid, citric acid, formic acid lactic acid, malic acid and succinic acid (Budak, 2010). The phenolic compounds are: gallic acid, catechin, catechin, epicatechin, chlorogenic acid, caffeic acid and p-coumaric acid (Budak *et al*., 2011). Acetic acid which is the volatile organic acid used in identification, is about 3-9% of the vinegar content and is the reason for the sour taste and slightly pungent odour. Several ancient Chinese medical books recorded it usefulness in the treatment of urns, cellulitis and psoriasis. Studies done recently showed antimicrobial, antioxidant, antidiabetic, antitumor, antiobesity, cholesterol lowering and antihypertensive properties, the most well known being the anti-obesity effect. Apple cider vinegar has been shown to increase satiety thus reducing the total amount of food consumed by users (Lim *et al*., 2009). This has made it to be highly recommended for persons trying to lose weight. It has been shown to kill bacteria; some parasites for example head lice, warts and nail fungal infections due to its high acetic acid content (Chang and Fang, 2007). The phenols and vitamins contained within have very high antioxidant properties (Iriti and Faoro,

2010; Fernandez-Mar., 2012; Ramadan and Al-Ghamdi, 2012). This prevents the peroxidation of lipids, proteins and DNA which is important in ageing, cancer and degenerative brain disorders development (Buonocore *et al*., 2010; Maes *et al*., 2011). Its antidiabetic activities has been accrued to its glucose lowering or anti-glycemic effect and prevention of the complete breakdown of complex carbohydrates through either increased gastric emptying or increased intake of glucose into tissues (Fushimi and Sato, 2005). The rich polyphenol content, especially chlorogenic acid, prevents lipid peroxidation in arteries. This slows down atherosclerosis and gives apple cider vinegar its cardioprotective effect (Laranjinha *et al*., 1994). Apple cider vinegar prevents the activation of angiotensin converting enzyme (ACE) therefore its antihypertensive effect (Rufi´an-Henares and Morales, 2007). The liver and the kidney are very important organs needed in the metabolism, detoxification and excretion of waste materials from the body. It has been reported that there is increased incidence of liver and kidney disease in the population due to indiscriminate use of various substances in which apple cider vinegar was claimed to be one of the culprits. This informed the interest of the researchers in evaluating the toxicity if any of this widely used product on the liver and kidney of albino rats which will be extrapolated to that of humans.

MATERIALS AND METHODS

Bragg's apple cider vinegar used in this study was obtained from Barata Pharmaceutical Stores which is NAFDAC approved (NAFDAC. No. 24791510: 32) and located at Rumuokuta junction along Ikwerre Road Port Harcourt, Rivers State, Nigeria. Lamivudine 300 mg: manufactured by Cipla LTD, Plot No L-139 Verna, and Goa 403722 and India for Evans Medical PLC, Km 32 Lagos-Ibadan Expressway, Lagos State. Batch No: E120365. MDF: 03/2012. EXP: 02/2014. NAFDAC Reg No: 04-7521. Specimen (animal) used for the experiment: twenty-two (22) albino rats were purchased from animal house of the Department of Biochemistry, University of Port Harcourt, Choba Park. The animals were fed with rat pellets, water and libitum. Chemicals and reagents: all chemicals and reagents used in this study were obtained from Randox Laboratories UK. Preparation of drug solution for administration: 1 ml and 2 ml of the preparation was given to the rats each day after weighing. Experimental procedure: a total twenty-two (22) albino rats of weight range (124-194g/BW) were randomly divided into five groups labeled A, B, C, D and E where group A served as control and rats (n=2 rats/dose) were treated with distilled water. Rats in groups B and C (n=5 rats/dose) were orally treated with 2 different doses (1 ml and 2 ml respectively) for 7 days. Rats in groups D and E (n=5 rats/dose) were orally treated with 2 different doses (1 ml and 2 ml respectively) for 14 days. Animals were sacrificed twenty-four (24) hours after last treatment.

Collection of blood and preparation of serum

The rats were withdrawn from the cages in each of the group twenty four (24) hours after the last administration of the drugs for 7 and 14 days and placed in a desiccator containing cotton wool soaked in chloroform to anaesthetize the rats. The blood samples were obtained by cutting the jugular vein of the rat on the neck by means of surgical blade and put in anticoagulant sample bottles smeared with lithium-heparin and fluoride oxalate. The blood samples were spun at 5000 rpm using MSE Centrifuge to obtain plasma. The animal was dissected and only the liver and kidney collected for pathological studies.

Measurement of AST (SGOT) and ALT (SGPT)

The activities of glutamic pyruvate transaminase and glutamic–oxaloacetate transaminase were analyzed according to the method specified by Asma *et al.* (2016). Measurement of ALP: Plasma alkaline phosphates activity was measured by the method of Soltan and Shehata, (2012).

Histological procedures and analysis

The liver was cut on slabs about 0.5cm thick and fixed in 10% normal saline for a day after which they were transferred to 70% alcohol for dehydration. The tissues were passed through 90% alcohol and chloroform for different durations before they were transferred into two changes of molten paraffin wax for 20 min each in an oven at 57%. Several sections of the 5 µm thick were obtained from a solid block of tissue and were stained with hematoxylin and eosin staining after which they were passed through a mixture of equal concentration of xylene and alcohols, following clearance of xylene, the tissues were oven dried. Photomicrographs were taken with a JVC colour video digital camera (JVC China) mounted on an Olympus light microscope (Olympus UK Ltd Essex, UK) to demonstrate cytoarchitecture of the liver. The same procedure was repeated for the kidney samples.

RESULTS AND DISCUSSION

Results in Table 1 shows that there was no significant change in the levels of ALT, AST and ALP in the acute group given 1 ml of the product when compared to the control. There was only one death recorded in this group.

Table 1. Acute and chronic effect of AST, ALT and ALP.

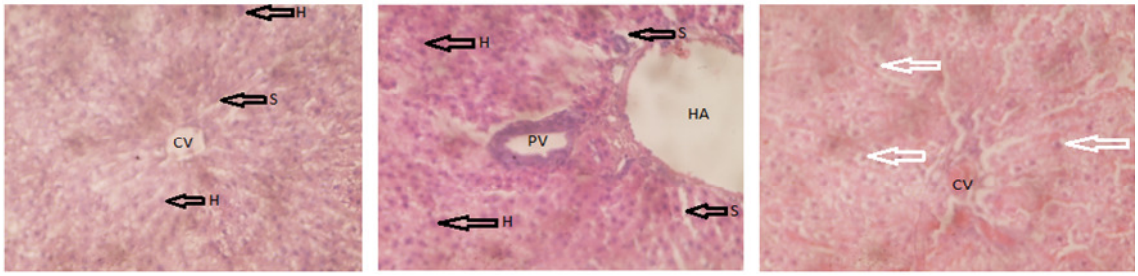
	Acute Effect			Chronic Effect		
	control	1 ml	2 ml	control	1 ml	2 ml
AST IU/L	123	120	130	125	123	132
		115	-		120	-
		118	135		-	-
		-	-		109	-
		125	-		117	
ALT IU/L	45	42	46	44	48	50
		41	-		43	-
		40	48		-	-
		-	-		45	-
		43	-		43	-
ALP IU/L	45	43	47	40	44	45
		49	-		36	-
		40	49		-	-
		-	-		41	-
		45	-		43	-

Note: (-) Represents rats that died before the day of sample collection.

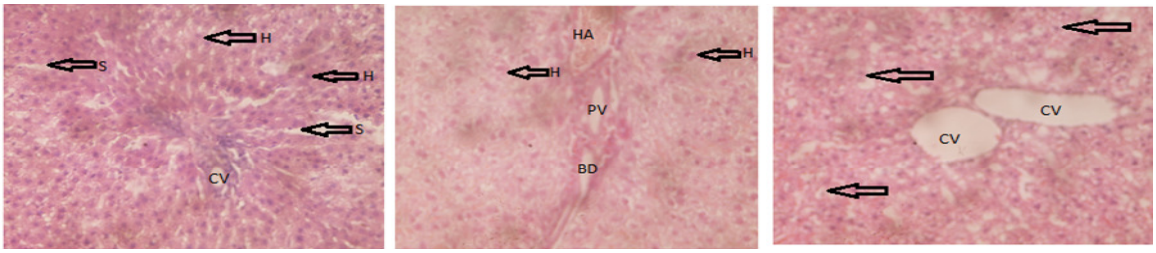
For those given 2 ml in the acute group, there was only a slight increase in the AST, ALT and ALP levels when compared to the control. There were 3 recorded deaths in this group after exposure and before the day of sample collection. In the chronic group, those given 1ml of apple cider vinegar showed no significant change in liver enzymes when compared to the control. There was only one death in this group before sample collection. Those given 2 ml of the solution only had slight increase in liver enzymes levels as compared to the control. Worthy of mention is the significant 4 deaths that were recorded in this group. Histology of the liver for the acute group showed normal cytoarchitecture at 1 ml dose and mild tissue distortion and microvascular steatosis (intracytoplasmic fat deposition) at 2 ml of administration. Amongst the chronic group there was mild tissue distortion, microvascular steatosis and congestion of the hepatic artery at 1 ml dose and mild tissues distortion and microvascular steatosis at 2 ml of administration. The results obtained from the histology of the kidney for the acute group showed a normal cytoarchitecture at 1ml of administration and distorted kidney tissue and obliterated bowman's capsular spaces at 2 ml of administration. The chronic group showed distorted kidney tissue and obliterated bowman's capsular spaces at both 1ml and 2 mls of administration (Figures 1 to 12). The photomicrograph of liver tissue showed histologically normal sinusoids, patent central vein and cords of hepatocytes (control for 7 days) (Figure 1). The photomicrograph of liver tissue treated with 1 ml of apple cider vinegar for 7 days showed histologically normal sinusoids, portal artery and vein and cords of hepatocytes (Figure 2). The photomicrograph of liver tissue treated with 2 ml of apple cider vinegar for 7 days showed mildly distorted tissue, congested central vein and hepatocytes microvesicular steatosis (shown by arrows) (Figure 3). The photomicrograph of liver tissue showed histologically normal sinusoids, central vein and cords of hepatocytes (control for 14 days) (Figure 4). The photomicrograph of liver tissue treated with 1ml of apple cider vinegar for 14 days showed mildly distorted tissue, portal triad with congested hepatic artery, portal vein and bile duct and hepatocytes with microvesicular steatosis (shown by arrows) (Figure 5). Figure 6- photomicrograph of liver tissue treated with 2 ml of apple cider vinegar for 14 days showed mildly distorted tissue, central vein and hepatocytes with microvesicular steatosis (shown by arrows).

The photomicrograph of kidney tissue showed histologically normal kidney tissue with normal glomeruli, renal tubules and patent bowman's capsular spaces (control at 7 days) (Figure 7). The photomicrograph of kidney tissue treated with 1ml of apple cider vinegar for 7 days showed histologically normal kidney tissue with normal glomeruli, renal tubules and patent bowman's capsular spaces (Figure 8). The photomicrograph of kidney tissue treated with 2ml of apple cider vinegar for 7 days showed histologically distorted kidney tissue with obliterated bowman's capsular spaces (shown by arrows) (Figure 9).The photomicrograph of kidney tissue showed histologically normal kidney tissue with normal glomeruli, renal tubules and patent bowman's capsular spaces (control at 14 days) (Figure 10). The photomicrograph of kidney tissue treated with 1ml of apple cider vinegar for 14 days showed histologically distorted kidney tissue with obliterated bowman's capsular spaces (shown by arrows) (Figure 11). The photomicrograph of kidney tissue treated with 2ml of apple cider vinegar for 14 days showed histologically distorted kidney tissue with obliterated bowman's capsular spaces (shown by arrows) (Figure 12).

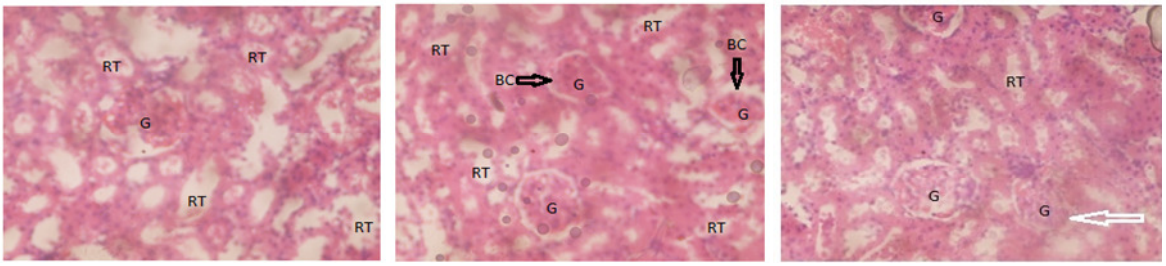
The biochemical analysis for the liver enzymes gave



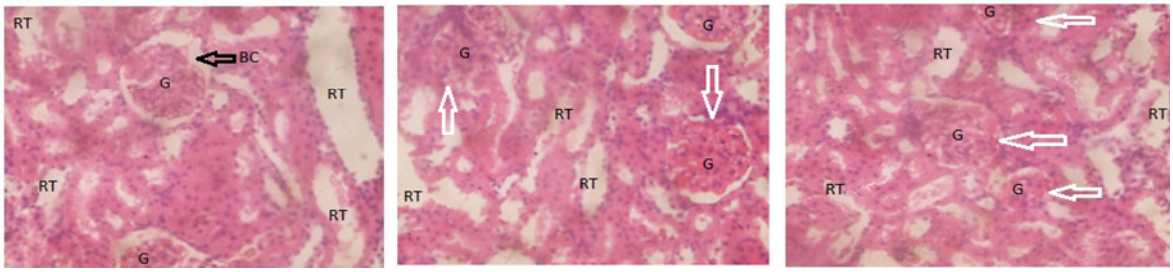
Figures. 1, 2 and 3 (L-R).



Figures. 4, 5 and 6 (L-R).



Figures. 7, 8 and 9 (L-R).



Figures. 10, 11 and 12 (L-R).

results that were not significant when compared to the control values as also seen in (Asma *et al.,* 2016). This has been ascribed to the high concentration of phenolic compounds (Asma *et al.,* 2016) found in apple cider vinegar. Also this study was carried out on healthy rats. However worthy of note are the significant deaths that occurred in members of both groups given 2 ml of apple cider vinegar irrespective of duration. The deaths were also more in the chronic group than the acute group. Unfortunately autopsy was not done to confirm the actual cause of death but it can be extrapolated that the concentration and duration of use of this product is important in the side effects experienced. Pathological changes were seen in the liver and this is in accordance

with the work done by Asma *et al.* (2016), which showed changes in the groups of animals exposed to apple vinegar as compared to the control (Asma *et al.,* 2016). From this research there were also some histopathological changes in the kidney.

Conclusion

Apple cider vinegar as a result of its antioxidant activity is said to be protective to both the liver and kidney although this was not the monitored in this work. This work shows that it also causes some degree of damage to the above named organs when it is used undiluted and continuously. This damage is not readily observed with measurement of the liver enzymes but is very clear when histology is done. The result also shows that more damage is done on increased exposure either in volume or in duration of use. It is on this premise that the product is advised to be diluted with enough water prior to use. Pathological changes of the liver and kidney progressively increased with concentration and duration. This study demonstrates that large amounts of apple cider vinegar when used undiluted has negative effects on the liver and kidney.

Authors' declaration

We declared that this study is an original research by our research team and we agree to publish it in the journal.

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