## ASSESSMENT OF THE NEPHROTOXIC AND HEPATOTOXIC EFFECTS OF COMBINED ARTIFICIAL FOOD SEASONINGS USING WISTAR ALBINO RATS.

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#### Abstract

This study investigated the possible effects of the combination of two most commonly used imported artificial food seasonings used in many delicacies during occasions/events in the South Eastern part of Nigeria on the liver and kidney function. Twenty (20) wistar albino rats were assigned to four groups (A,B,C and D) of five animals per group. The combined seasonings (coded - BM) was administered in graded doses; group A(Control), group B(200mg/kg), group C(400mg/kg) and group D(800mg/kg). The study lasted for two weeks and BM was administered once daily through oral intubation. The animals were anaesthetized with chloroform and sacrificed on the 15<sup>th</sup> day. Blood samples were collected by cardiac puncture into plain tubes for analysis after an overnight fast. Alanine aminotransferease(ALT), aspartate aminotransferase(AST) and alkaline phosphatase(ALP) were determined spectrophotometrically using enzymatic methods. Urea and creatinine levels were evaluated using Berthelot and Jaffe Slot methods respectively. The plasma/serum electrolytes were determined using Convergy Ion Selective Electrodes Electrolyte Analyser. The result showed a significant (p<0.05) increase in the urea (6.03  $\pm$ 20.87 $\mu$ mol/l) concentration of the group C when compared with group A(5.07 ± 32.14 $\mu$ mol/l). There are also significant increases in the AST(14.67  $\pm$  1.25  $\mu$ /l) and ALP(284.70  $\pm$  154.11 $\mu$ /l) activities of the groups administered with the highest doses of BM when compared with control(13.07  $\pm$  0.06 and 118.07  $\pm$  19.82  $\mu$ /l). This suggest that caution should be applied in the consumption of some imported artificial food seasoning as chronic usage could predisposed the consumers to kidney or liver related diseases.

Keywords: Artificial food seasoning, spices, herbs.

#### Introduction

Food seasonings are ingredients or additives added to food to enhance its taste, flavor and appearance. They are primarily made up of spices and herbs, can also include salt, sugar, vinegar, oils, sauces, and other animal products. Spices as component of food seasoning have been reported to serve as food preservatives and for their health-enhancing properties for centuries (Kaefer and Milner, 2011). In the world, spices stimulate appetite and create visual appeals to foods (Opara and Chohan, 2014). The use of food seasoning in culinary activities antecedes its recorded history and has been reported to be essential component of local dishes in South Asia and Middle East since 2000BCE (Tapsell et al., 2006). According to a report, In Mesopotamia, where agriculture began, there was evidence that humans were using thyme for health benefits as far back as 5000 BC and grew garlic as early as 3000 BC (Singletary, 2016). Food seasoning could be natural (derived from plant or animal source) or artificial (designed to mimic the potentials of natural ingredients). Natural food seasoning has been reported to posses some medical and pharmacological attributes because they contain some plant secondary metabolites (Newman and Cragg, 2012). Previously, natural food seasonings were used mainly for their organoleptic and preservative properties; but recent studies on their medicinal and nutritional attributes have exposed new prospects in the fields of nutraceuticals and functional foods (Ogbunugafor et al., 2017). Artificial food seasonings which may likely have high amounts of salt or other chemicals/additives are not as healthy as traditional/natural condiments in preparing foods. In Nigeria, there are different brands of artificial food seasonings which are locally manufactured or imported as bouillon cubes or packaged in powdered forms. Their major constituents are salt, monosodium glutamate (MSG), hydrogenated oil (trans fat) and other variable spices of animal or plant origin as indicated by the manufacturers and are widely used in preparation of food in homes, restaurants, in and out-door events/ceremonies (Ugbabe et al., 2022; Otunola et al., 2010; Akpanyung, 2005; Nwinuka et al., 2005). The safety of artificial food seasonings should be a major concern to health authorities and the general public owing to the influence of these food seasonings on the health of the consumer. In Nigeria, food seasoning appears to be an inevitable component of culinary activities, manifesting in the rise in demand for cheaper condiments that would enhance cooking skill and boost the desired result. In the South Eastern part of Nigeria, two artificial food seasoning (coded B and M) are the major food seasonings used in combination in variety of food preparations, especially at ceremonies, thereby increasing their consumption/utilization and which necessitated the research into the subject matter.

#### Sample collection/preparation

The two most commonly used artificial food seasoning (B and M), in combination, purchased from Eke Awka, a popular market in the South Eastern part of Nigeria were used for the study. The seasonings were weighed of equal proportion with a sensitive digital weighing balance and mixed together in the ratio of 1:1 (5g per seasoning). The mixture (BM) was dissolved in distilled water 193

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and the various doses administered in milliter were obtained using the below dose control formular;

Dose =	Dose mg/kg	Х	Weight of rat	
	1000		Concentration (mg/ml)	

#### **Experimental Animal**

Wistar albino rats, used for the study were procured from a reputable research center in Awka Anambra State, Nigeria. They were maintained and housed in cages and allowed to acclimatize with the environment for one week before the experiment with access to both food and water *ad libitum*.

Study design

A total of twenty wistar albino rats were grouped into four groups of five rats per group as follows;

Group A:	Normal control
Group B:	200mg/kg of BM
Group C:	400mg/kg of BM
Group D:	800mg/kg of BM

The study lasted for two weeks with BM administered once daily through oral intubation. The animals were anaesthetized with chloroform and sacrificed on the fifteenth day and blood samples were collection after an overnight fast into plain sample containers for analysis via cardiac puncture.

## **Biochemical Analysis**

The Alanine aminotransferease(ALT) and aspartate aminotransferase(AST) were assayed spectrophotometrically using Randox Kit purchased from Randox Laboratory, UK according to manufacturer's protocol. (Omokehinde *et al.*, 2024). Alkaline phosphatase (ALP) was determined using IFCC method as outlined by Omokehinde *et al* (2024). The urea and creatinine levels were determined Berthelot and Jaffe-slot methods respectively. The plasma/serum electrolytes were determined using Convergy Ion Selective Electrodes Electrolyte Analyser (Convergent Technology, GmbH and CO, KG, Marburg, Germany).

## Statistical analysis

Results are presented as mean  $\pm$  S.D (standard deviation of the mean). Comparisons were done between groups by use of one-way analysis of variance (ANOVA) followed by post hoc tests (Least Square Deviation). *P* < 0.05 was declared as significant statistically.

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#### Results

Table 1 shows the effect of the combined artificial food seasoning on the some parameters for assessing liver function. The group D animals administered with the highest dose showed the highest enzyme activities for ALT, AST and ALP. The AST and ALP activities of group D, 14.67  $\pm$  1.25 and 284.70  $\pm$  154.11 respectively were significantly higher than the control (13.07  $\pm$  0.06 and 118.07  $\pm$  19.82).

# Table 1: showing the results of ALT, AST and ALP activities of the various groups of rats administered with the graded doses of BM

Groups	ALT (µ/l)	AST (µ/l)	ALP $(\mu/l)$
А	$13.57 \pm 0.55$	$13.07\pm0.06$	$118.07 \pm 19.82$
В	$14.17 \pm 1.53$	$13.96 \pm 0.76$	$119.60 \pm 26.61$
С	$13.77 \pm 067$	$13.57\pm0.49$	$82.37 \pm 19.35$
D	$15.60 \pm 2.10$	$14.67 \pm 1.25 **$	284.70 ± 154.11**

\*\* considered significant at p<0.05

From table 2, the creatinine and urea concentration of the group administered with the highest dose were insignificantly higher when compared with group A, although group C exhibited a significant increase in urea concentration  $(6.03 \pm 20.87)$  compared with group A  $(5.07 \pm 32.14)$ .

# Table 2: shows the creatinine and urea concentrations of the various groups of rats administered with the graded doses of BM

Groups	Creatinine(µmol/l)	Urea(µmol/l)	
А	$72.33 \pm 2.45$	$5.07 \pm 32.14$	
В	$61.53 \pm 22.64$	$5.07 \pm 32.14$	
С	$64.07 \pm 8.04$	$6.03 \pm 20.87 **$	
D	$79.83 \pm 15.57$	$5.20\pm10.00$	

\*\* considered significant at p<0.05

Figure 1 shows the serum electrolyte concentrations in of the various groups of animals. The serum electrolyte levels of all the various groups administered with BM were insignificantly higher than the control (group A).



# Fig 1: showing the electrolyte concentrations of the different groups of rats administered with graded doses of BM

#### Discussion

This study assessed the effect of the combination of commonly consumed artificial food seasoning (coded BM) in South Eastern Nigeria on the liver and kidney function. Liver function tests (LFTs) 196

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are categories of blood tests that provide information on the health of a patient's liver (Okechukwu-Ezike, 2024). The result of the liver function showed an increase in the ALT, AST and ALP activities, with significant increase (p<0.05) in AST and ALP activities of the animals administered with the highest dose of the seasoning when compared with the control group. As reported by Lee (2009), Alkaline Phosphatase (ALP), Aspartate Aminotransferase (AST), and Alanine Aminotransferase (ALT) are good indicators of liver injury in people and animals with some degree of intact liver function. AST and ALT are also considered to be two of the most important tests to detect liver injury (Aulbach and Amuzie, 2017; Sparling, 2016). ALT facilitates the formation of glutamate and pyruvate in the hepatocyte which is important for energy production while AST facilitates amino acid metabolism (Aulbach and Amuzie, 2017; Sparling, 2016). Alkaline Phosphatase is a hydrolase enzyme that increases a phosphate group from many molecules through phosphorylation (Okechukwu-Ezike, 2024). Increased activities of these enzymes could be indicative of hepatotoxicity and loss of liver function could result to death, considering the importance of the liver. As seen from the result of this study, the increase in the enzymatic activities of AST and ALT supports an earlier study by Singh et al., (2001) and Iweka et al., (2016). Increase in ALP activity indicates an anomaly in the liver and, as such, is also used as an index to establish the possibility of some toxicity/disease of the liver.

The result of the kidney function evaluation showed an insignificant elevation in the urea and creatinine levels of the groups administered with the highest dose, when compared with the control. There is also an insignificant increase in the electrolyte concentrations of the all the test groups when compared with the control. Plasma concentrations of creatinine and urea could be used as indicators of nephrotoxicity, hence an abnormally elevated blood creatinine and urea is diagnostic of impaired renal function. Creatinine and urea are waste products of protein metabolism that need to be excreted by the kidney, therefore a marked increase of these parameters, as observed in this study, suggests an indication of functional damage to the kidney (Panda et al., 2009). Urea level can be elevated by many other factors such as dehydration, antidiuretic drugs and diet, while creatinine is more specific to the kidney, since kidney damage is the only significant factor that increases the serum creatinine level (Jose, 2014). As aforementioned, food seasonings are made up variety of ingredients especially salt. High salt intake has been linked to the development of high blood pressure and kidney disease (Graudal et al., 2014). Therefore, the elevated creatinine and urea concentrations from this study could be attributed to the high salt composition of the food seasonings which also manifested in the slight elevation of electrolyte concentrations when compared with the control.

#### Conclusion

The significant increase and changes in the assessed liver and kidney parameters is indicative of the hepatotoxic and nephrotoxic potential of the seasonings. These have provided some crucial information on artificial food seasoning and calls for caution in their usage in order to maintain a healthier life.

#### Recommendations

From the result of this study, regulatory agencies should ensure and enforce compliance with standard to help curb the availability of sub-standard products and maintain quality. Additionally, further researches on imported artificial food seasoning should be encouraged to ascertain safety and maintain healthier living.

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