

# The Effect Of Acute Consumption Of Energy Drink On Hepatic Function In Male Wistar Rats

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## ABSTRACT

The aim of this study is to check the potential health impact associated with excessive consumption of energy drink. Previous studies showed that energy drink contains more caffeine than a cup of coffee and a bottle of coke. Twenty inbred male wista rats weighing 120-150g were grouped into 4 groups of 5 animals each. Group A received feed and water only (positive control); group B

received 1 ml of Monster Drink; group C 2 ml of Monster Drink; and group D 3 ml of Monster Drink. The administration of Monster Drink was done for 21 days. The result showed a significant ( $p < 0.05$ ) increase in AST level in-group B while group C and D had a significant ( $p < 0.05$ ) decrease when compared to group A. There was a significant ( $p < 0.05$ ) increase in alkaline phosphatase level in group B and D while group C had a significant ( $p < 0.05$ ) decrease when compared to group A. Alanine transaminase result showed a significant ( $p < 0.05$ ) increase in-group B while group C and D had a non-significant ( $p > 0.05$ ) increase when compared to group A. Acute consumption of energy drink (for 21 days) affects the liver enzymes. There was significant increase in groups feed on low dose while those of medium and high doses significantly decreased liver enzymes. Further studies are recommended for the chronic consumption of Energy drink.

**Keywords:** Energy drink, AST, ALP, ALT, liver enzymes.

## INTRODUCTION

Energy drink first appeared in Europe and Asia in 1960 as a result of customer requirements for dietary supplements that give energy (Reissig et al, 2009). Most studies from Saudi Arabia reported that greater than 50% of the consumers were young between the ages of 13- 35 years and over 40% drank more than 3 cans per week. Currently the market has been recently flooded with energy beverages with different brand names. As of 2018, over 500 different companies that deal on different brands of energy drinks (Kutia et al, 2019). The common thing about all brands of these energy drinks is that they contain high amount of certain substances and compounds which has stimulatory effects. Some of these substances are; caffeine, guarana, taurine, vitamin B and many other. Some of these companies include some natural ingredients into their products. These substance aim at increasing athletic performance, improve concentration. A study has shown that the caffeine levels in energy drinks are between 50 and 505 mg/ can, which is much higher than the caffeine content of one can of coke which contain 34mg of caffeine (Burrows et al, 2013).

The adverse effect of these substances when consumed in high amount has been a major concern across the medical communities around the world. (Sepkowitz, 2013, MacDonald, 2013). According to the report of Zenith International's Global Energy Drinks, 2019, the number of energy drink per liter was 2 billion liters in 2003 which was increased to 3.9 billion liters in 2008. There has been a recent increase in the number of consumers of energy drinks. The global consumption rate has risen from 0.4 liters in 2003 to 0.8 liters in 2008 per person (Kutia et al, 2019). It was noted by Ali et al, 2015 that the major consumers of these energy drinks are young adults between the ages of 11- 35 years.

Although these energy drinks are certified, standardized and are considered suitable for use, forms no justification to believing that these products are totally safe especially when consumed in large quantity and abused. The scientific community has expressed some concerns about the dangers of excessive use and dependency on energy drinks.

## MATERIALS

**Location of the Study:** This study was carried out in the Department of Physiology, College of Health Sciences, Nnamdi Azikiwe University, Nnewi Campus. Ethical approval consent was obtained for the progress of this study, from the Faculty of Basic Medical Sciences, College of Health Sciences, Nnamdi Azikiwe University, Nnewi Campus.

**Materials:** Twenty-(20) inbred male wista rats, Monster Drink, distilled water, oral cannula, Automatic Water distiller (SZ-1 Search Tech Instrument), chloroform, heparinized capillary tube, Electronic weighing balance (M-Metallar M311), 5ml hypodermic sterile syringe, animal weighing balance (Camry LB11), Normal laboratory chow (Standard Pellet), and standard cage

### **Methodology:**

**Experimental Animals:** Twenty inbred male wista rats weighing 120-150g were used for the study, and housed in the animal house Department of Physiology. The animals were maintained in standard cages at ambient temperature with standard pellet and distilled water ad libitum, and acclimatized for 2-weeks before the commencement of the administration of the Monster Drink. Animals were maintained under 12-hours light and dark cycles.

**Experimental Design:** Twenty inbred male wista rats weighing 120-150g were grouped into 4 groups of 5 animals each. Group A received feed and water only (positive control); group B received 1 ml of Monster Drink; group C 2 ml of Monster Drink; and group D 3 ml of Monster Drink. The administration of Monster Drink was done for 21 days within the hours of 7am to 8am through oral garvage.

**Sample collection:** Animals were anaesthetized with chloroform in an enclosed container 24 hours after the last administered dose of the monster drink, blood was collected using heparinized capillary tube and put in a plain container (Parasuraman et al, 2010), and centrifuge using centrifuge (England) and serum were retrieved and used for serum liver enzymes (AST, ALT, and ALP).

**Alkaline Phosphatase (ALP) Estimation:** This test was carried out according to the method described by (Kind and Jegathessan, 1959).

**Principle:** Alkaline Phosphatase in Alkaline medium hydrolyses Phenyl phosphatase in 15minutes at 37°C and pH of 10 to release phenol which in the presence of potassium ferricyanide reacts with 4-aminophenazone to give a red-pink color which is measured Spectrophotometrically at 510nm wavelength. The intensity of the color indicates ALP activity in the sample.

**Aspartate Amino Transferase (AST) Estimation:** This test was carried out according to the method described by (Reitman and Frankel, 1957).

**Principle:** The substrates in the reaction are alpha ketoglutaric acid and L- Aspartate. The products formed by enzyme action are glutamate and oxaloacetate. Addition of 2, 4 dinitrophenyl hydrazine results in the formation of hydrazine complex with ketoacids. A red color is produced on the

addition of sodium hydroxide. The intensity of color is related to the enzymatic activity and this can be measured at 550 nm wavelength using Spectrophotometer.

**Alanine Amino Transferase (ALT) Estimation:** This test was carried out according to the method described by (Reitman and Frankel, 1957).

**Principle:** The substrates in the reaction are alpha ketoglutaric acid and L- Aspartate. The products formed by enzyme action are Glutamate and Pyruvate. Addition of 2, 4 dinitrophenyl hydrazine results in the formation of hydrazine complex with ketoacids. A red color is produced on the addition of sodium hydroxide. The intensity of color is related to the enzymatic activity and this can be measured at 550 nm wavelength using Spectrophotometer.

**Data analysis:** Data obtained were subjected to SPSS version 25. ANOVA was used to analyzed the serum level of liver enzymes (AST, ALT, and ALP) followed by multiple comparism using post HOC Turkey HSD. Values were presented as MEAN±STD, and data were considered significant at p<0.05.

## RESULT

**Table 4.1 Effect of Monster Drink on Aspartate Transaminase and Alanine Transaminase level**

Groups	Aspartate Transaminase (IU/L)	Alanine Transaminase (IU/L)
	MEAN±STD	MEAN±STD
Group A (Control)	105.00±1.00	41.50±4.22
Group B (1 ml of Monster Energy)	144.50±7.50***	44.50±32.24*
Group C (2 ml of Monster Energy)	62.00±1.00***	42.00±2.00NS
Group D (3 ml of Monster Energy)	90.50±0.50**	42.50±1.00NS

Data was analyzed using ANOVA followed by Post-Hoc Turkey HSD and values were considered significant at p<0.05. p<0.001\*\*\*, p<0.01\*\*, and p<0.05\*

Table 4.1 result showed a significant (p<0.05) increase in AST level in-group B while group C and D had a significant (p<0.05) decrease when compared to group A.

Alanine transaminase result showed a significant (p<0.05) increase in-group B while group C and D had a non-significant (p>0.05) increase when compared to group A.

**Table 4.2 Effect of Monster Drink on Alkaline Phosphatase activity**

Groups	Alkaline Phosphatase (IU/L)
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	<b>MEAN±STD</b>
Group A (Control)	121.87±1.04
Group B (1 ml of Monster Energy)	287.06±1.05***
Group C (2 ml of Monster Energy)	100.63±2.35***
Group D (3 ml of Monster Energy)	393.66±6.18***

Data was analyzed using ANOVA followed by Post-Hoc Turkey HSD and values were considered significant at  $p < 0.05$ ,  $p < 0.001$ \*\*\*,  $p < 0.01$ \*\* , and  $p < 0.05$ \*

Table 4.2 result showed a significant ( $p < 0.05$ ) increase in alkaline phosphatase level in group B and D while group C had a significant ( $p < 0.05$ ) decrease when compared to group A.

## DISCUSSION

Energy drinks are usually nonalcoholic drinks that contain high amount of caffeine amongst other substances. About 50- 505 mg of caffeine is contained in a bottle of energy drink, an amount that is 3 times or more, greater than the caffeine in the usual soft drinks (Marinoni et al, 2022). Energy drink can pose a harmful effect on the health particularly when consumed in excess. Study by Bawazeer et al, 2013 has linked the consumption of energy drink with an increased risk of obesity, hypertension and cardiac problems.

Effects of acute consumption of energy drink on liver enzymes (AST, ALT and ALP) were evaluated in this study. There was significant increase in groups feed on low dose while those of medium and high doses significantly decreased liver enzymes. Studies done by Mansy et al, 2017, showed that there was significant increase in the level of AST, ALT and ALP showing some evidence of liver damage following chronic consumption of energy drink. Similar increase was also reported by Bukhari et al, 2012. It has also been demonstrated that rats fed with energy drink alone or in combination with alcohol showed much elevation in the levels of serum total bilirum, ALT, ALP and AST than in the control group (Ugwujaja, 2014).

The effect of the consumption of energy drink has been observed in almost all organs and system of the body especially the central nervous system, peripheral nervous system (Persad, 2011), cardiovascular system, (Wassef et al, 2017), urinary system (Salih et al, 2018), gastrointestinal system (Ayuob and El Beshbeishy, 2016) and liver (Mansy et al, 2017).

Acute consumption of energy drink (for 21 days) affects the liver enzymes. There was significant increase in groups feed on low dose while those of medium and high doses significantly decreased liver enzymes. Further studies are recommended for the chronic consumption of Energy drink.

## DISCLOSURES

There was no conflict of interest.

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