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Research Article

Effects of Oral Administration of Environmentally Exposed Malted Product (Maltina Brand) on Some Biochemical Parameters in Albino-Rats

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Abstract

This study was conducted to determine the effects of Oral Administration of Environmentally Exposed Malted Product (Maltina Brand) on Some Biochemical Parameters in Albino-Rats. Maltina products packaged with two different materials (metal can and plastic) were exposed to sunlight for seven days and fifteen days, while the control sample was not exposed to the environment. They were evaluated for alkaline phosphatase, alanine transferase, and aspartate aminotransferase of the albino rats; lipid peroxidation, liver function tests (albumin, direct and total bilirubin) of the albino rats. 24 adult male albino rats were used for the study. Results obtained showed that there was significant changes (P < 0.05) in concentration of AST, ALT and ALP for the period of the experiment. There was significant changes in Lipid peroxidation (P < 0.05) in all the groups. Bilirubin level was affected significantly (P < 0.05). Therefore, display/storage of food products should place premium on the environmental factors of the area.

Keywords: Environmental; Malted; Biochemical; Albino Rat

Introduction

Malt drink is a non-alcoholic beverage obtained from unfermented wort. It is a very common drink in the Caribbean Islands and Latin America, but its consumption in Nigeria has been on the increase. Historically, malt drink was used as food for children and the sick, but has since become a mainstream beverage consumed by people of all ages. More importantly, malt-based drinks have developed a reputation over the centuries for their nutritional value, a message that is attractive for manufacturers to carry across in today's climate of increasing health awareness.

The manufacturing process of fermentation for malt-based soft drinks is similar to that used in beer production, with the products containing typically malt, sugar, and hops. Fermentation has not only been useful in terms of preservation, but has helped to add flavour and texture which have been developed for Malt production of various qualities. Non-alcoholic barley malt-based beverage of reduced caloric content can be produced following boiling, cooling and filtering. Prior to packaging, the amylolytic enzyme system converts the maltose and complex carbohydrates (dextrin) present in the beverage extract to simple, sweet-tasting dextrose [1]. Maltina is a natural malt drink which is produced by mashing of grains to get sugar that is blended together with other essential ingredients which provide taste, colour, flavor, multivitamins and minerals. It is enriched with Vitamin A, B, B2, B3, B5, B6 C and calcium which makes it to be positioned as the drink that has delivers superior nourishment for life, nourishment for an active, vibrant life for all [2]. The raw materials used for production of maltina include malt, mated sorghum, raw sorghum and maize grist, hops, water with additives such as amylex, calcium chloride (cacl), maltina complex (vitamin A, B complex and C), caramelized dextrose maltase (CDM) and lactic acid [3].

Malt products typically contain water, sweetener (8 - 12%, w/v), carbon dioxide (0.3 - 0.6% w/v), acidulants (0.05 - 0.3% w/v), flavorings (0.1 - 0.5% w/v), colorings (0 - 70 ppm), chemical preservatives, antioxidants (<100 ppm), and/or foaming agents (e.g., saponins up to 200 mg/mL). Some types of soft drink use sugar substitutes. However, certain ingredients may be hazardous to health if exposed to environmental condition like the sunlight or consumed in large quantities. This study is therefore aimed to investigate the effects of environmentally exposed malt product (canned and plastic maltina) on some biochemical parameters in albino-rats.

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Materials and Methods

Sources of materials

The materials used are malt products (Maltina: Canned and Plastic). Obtained from Markurdi plant in Benue State, Product of Nigerian Breweries Limited. The Albino rats were purchased from the experimental animal house in Biochemistry Department, Faculty of Natural Sciences, Prince Abubakar Audu, Anyigba, Kogi State, Nigeria.

Experimental design

The completely randomized design (CRD) was used. The Products were exposed to adequate sunlight between 11:00 am - 4:00 pm for 15 days with corresponding daily average temperature taken for each day as follow; 31°C, 32°C, 31°C, 32°C, 30°C, 32°C, 30°C, 30°C, 29°C, 33°C, 31°C, 29°C, 30°C, 31°C, 30°C; for 15 days and average daily temperature was 30.7oC. Twenty-four adult male albino rats divided into 4 groups with 6 rats in a group was used for this Experiment. Group 1- rats were fed with normal feed and distilled water ad libitum (control group); group 2- rats were given oral administration of malt that is unexposed to the environment (unexposed group) 3ml; 1ml at a time in three separate administration/ day + normal feed and water ad libitum. Group 3 rats were given oral administration of 3ml total of exposed canned maltina (3ml; 1ml at a time in three separate administration /day) + normal feed and water ad libitum. Group 4: rats were given oral administration of 3ml exposed Plastic maltina (3ml; 1ml at a time in three separate administration/day) + normal feed and water ad libitum (Plastic maltina). The malt product was administered orally using oral gavage. Administration of the product lasted for 15 days. Once daily at 24 hours interval. Three (3) rats were sacrificed from each group 24 hours after the 7th and 15th doses. After appropriate dose and the completion of the experiment, the rats were anesthesised in large jar containing cotton wool soaked with diethyleether. The rats at subconscious stage, were brought out of the jar quickly dissected and the blood sample collected via cardiac puncture into capped non-EDTA bottles. The blood was allowed to clot for about 1 hour at room temperature and then placed in a refrigerator at 10°C for another 1 hour. The liver and kidney were homogenized in ice-cold 0.25M sucrose solution (1:5w/v). This was then centrifuged using Heraus-Christ GMBH Osterode refrigerated centrifuge at 4000rpm for 30 minutes and the serum collected using a Pasteur's pipette. This was stored frozen until required.

Determination of AST concentration/activity

Aspartate aminotransferases (AST), (EC 2.6.1.1) activity was determined using the Method of Reitman and Frankel (1957) as modified by Friedman and Young (1997) by monitoring the concentration of oxaloacetate hydrazone formed with 2, 4-dinitrophenylhydrazine as depicted in the chemical equation below:

α-oxoglutarate + L-aspartate _____AST ____ L-glutamate + Oxaloacetate.

Reagent 1 (phosphate buffer, L-aspartate and α -oxoglutarate) 0.5 ml was accurately dispensed into two clean test tubes labelled as test sample and reagent blank, containing 0.1 ml of serum and 0.1 ml of distilled water respectively. Each of the content was mixed properly and further incubated for 30 minutes at 37°C. Then 0.5 ml of Reagent 2 (2, 4-dinitrophenylhydrazine) was dispensed using a micro pipette into each test tubes, mixed and allowed to stand for 20 minutes at room temperature. Thereafter, 0.5ml of sodium hydroxide solution was accurately added and mixed properly. The absorbance was read and recorded against the blank at 540nm after 5 minutes. The concentration of AST (U/L) was then calculated.

Determination of ALT concentration/activity

Alaninetransferases (ALT), (EC, 2.6.1.2)

Alanine aminotransferase activity was determined using the Method of Reitman and Frankel, (1957) as modified by Friedman and Young (1997) by monitoring the concentration of pyruvate hydrazone formed with 2, 4-dinitrophenylhydrazine at 540 nm and 37°C as depicted by the following chemical reaction.

α-oxoglutarate + L-alanine _____L-glutamate + pyruvate

Reagent 1 (phosphate buffer, α -oxoglutarate + L-alanine) 0.5 ml was dispensed into two clean test-tubes, containing 0.1 ml of serum and 0.1mL of distilled water (blank) respectively. Each of the content was mixed very well and further incubated for 30 minutes at 37oC. Then, 0.5 ml of reagent 2 (2, 4-dinitrophenylhydrazine) was added to the set up, mixed and left for exactly 20 minutes at room temperature. Thereafter, 0.5ml of sodium hydroxide solution was added and mixed properly. The absorbance was read and recorded against the blank at 540nm after 5 minutes. The concentration of ALT (U/L) was then calculated.

Determination of ALP concentration/activity

Alkaline Phosphatase (ALP), (EC 3.1.3.1) activity was determined using the method of Wenger., et al. (1984). The alkaline phosphatase acts upon the AMP buffered sodium thymolpthalein monophosphate. The addition of an alkaline reagent stops the enzyme activity and simultaneously develops a blue chromagen, which is read photometrically.

p-Nitrophenyl phosphate+H₂O $\leftarrow ALP \\ Mg^{2+} \rightarrow$ p-Nitrophenol +Phosphate

Alkaline Phosphatase Substrate 0.5 ml was dispensed into la-

belled test-tubes of blank, sample and standard was equilibrated to 370 C for three (3) minutes. Precisely 0.05 ml (50 μ l) each of deionized water, sample and standard was added respectively. Contents were mixed. They were incubated for exactly ten (10) minutes at 370C. Alkaline Phosphatase Colour Developer 0.5 ml was added after 10 minutes. The solution was mixed very well. The absorbance

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was read and recorded against the blank at 590nm after 5 minutes. The concentration of ALP (U/L) was then calculated using the expression.

Liver function test

Albumin: Method described by Doumas., et al. [4].

Bilirubin (Total and Direct): the method described by Malloy., et al. (1937) as modified by Kaplan., et al. [5].

Results and Discussion

From the Enzyme assay of the serum samples of albino rats, the concentration of Aspartate aminotransferase (AST) was significantly high (p < 0.05) among the groups after day 7 and day 15. The concentration of Alanine aminotransferase (ALT) showed no significant difference (p > 0.05) among the control group, the unexposed group and the canned group but there were significant differences within the plastic group after day 7 and day 15. At day 15, the concentration of ALT for the control and the unexposed groups showed no significant difference between each other, but there existed significant difference between them and the exposed groups.

The concentration of alkaline phosphatase (ALP) showed no significant difference (p > 0.05) between the control and the unexposed group both at day 7 and day 15 but no significant difference between the canned and the plastic groups. Since high or low concentration of these biomarkers (AST, ALT and ALP) is an indicator that there is liver and kidney damage; this therefore implies that there were damages done to the livers and the kidneys of the affected groups.

The exposed plastic maltina group was significantly higher than the other groups both at day 7 and 15. This indicates that the exposed plastic maltina group generates more lipid peroxides (free

Sample type		AST (U/L)	ALT (U/L)	ALP (U/L)	Lipid Peroxidation U/mg
Control	[day 7]	70.47 ± 5.08^{b}	19.67 ± 2.67 ^b	42.16 ± 1.30^{a}	$6.77 \pm 1.75^{\circ}$
Unexposed		74.28 ± 8.22 ^c	21.00 ± 2.31 ^b	48.10 ± 3.85°	6.19 ± 1.25^{a}
Exposed canned		76.42 ± 1.68^{d}	15.00 ± 3.00^{a}	47.06 ± 6.46^{b}	5.91 ± 1.05^{a}
Exposed plastic		67.32 ± 7.1^{a}	15.33 ± 1.67 ^a	46.22 ± 4.20^{b}	10.61 ± 2.82^{b}
SEM		1.49	1.52	1.30	1.10
Control	[day 15]	79.59 ± 2.87 ^d	$19.67 \pm 2.67^{\rm b}$	52.01 ± 1.16^{b}	10.25 ± 1.73^{b}
Unexposed		77.12 ± 2.27 ^c	19.67 ± 2.27^{b}	54.49 ± 1.41°	10.33 ± 1.19^{b}
Exposed canned		75.25 ± 7.28^{b}	19.67 ± 1.33 ^b	44.88 ± 0.10^{a}	9.61 ± 1.62^{a}
Exposed plastic		71.87 ± 2.73^{a}	23.67 ± 1.33^{a}	46.70 ± 1.93^{a}	11.39 ± 2.65°

Table 1: Effect of oral administration of unexposed, exposed Canned and Plastic Maltina on Serum levels of AST, ALT and ALP.

Values are expressed as mean ± SEM (n = 3). Values with the same alphabets are not statistically different at (P < 0.05).

radicals).

Serum levels of Albumin, total Bilirubin and direct Bilirubin are essential indices for testing liver functions. From table 2, at day 7 and 15, albumin values for exposed canned and exposed plastic groups are significantly low compared to the unexposed and the control groups. This hypoalbuminemia result indicates some damages to the liver and the kidney. For the total bilirubin, the value for the exposed canned maltina only was significantly higher than the other groups at day 7, while at day 15, both the exposed canned and exposed plastic are significantly high (p < 0.05) from other groups. While for the direct bilirubin, at day 7 the values showed no significant difference in all the groups. But at day 15, both exposed canned and plastic maltina groups were significantly different from other groups. This implies that there were damages done

Sample type	Duration	Albumin (g/dl)	T. Bilirubin (mg/dl)	D. Bilirubin (mg/dl)
Control		4.57 ± 0.12^{b}	0.63 ± 0.05^{a}	0.19 ± 0.02^{a}
Unexposed		4.27 ± 0.08°	0.64 ± 0.18^{a}	0.19± 0.05ª
Exposed Canned	[day 7]	4.23 ± 0.06 ^a	0.67 ± 0.16^{b}	0.20 ± 0.05^{a}
Exposed plastic		4.10 ± 0.18^{a}	0.64 ± 0.21^{a}	0.19 ± 0.06^{a}
SEM		0.04	0.01	0.01
Control	[day 15	4.17 ± 0.41^{b}	0.37±0.08ª	0.11±0.02ª
Unexposed		4.58 ± 0.36^{b}	0.34 ± 0.03^{a}	0.10 ± 0.01^{a}
Exposed Canned		4.47 ± 0.33ª	0.44 ± 0.08^{b}	0.13±0.03 ^b
Exposed Plastic		4.45 ± 0.17^{a}	0.48±0.14°	0.14 ± 0.04^{b}

Table 2: Effect of oral administration of unexposed, exposed canned and Plastic Maltina on Serum levels of Albumin, total Bilirubin and

 Direct Bilirubin.

Values with the same alphabets are not statistically different at (P < 0.05). Values are expressed as mean ± SEM (Values n = 3).

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to the liver.

Conclusion

In conclusion, this study revealed that exposure of canned and plastic maltina to sunlight would result into chemical changes that could cause damage to organs such as liver and kidney. Canned and plastic maltina should therefore be stored in environments that are not exposed to sunlight.

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