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Original Article

The metabolic consequences of energy drink consumption in Wistar Albino rats- A cross sectional study.

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Abstract:

Introduction: Energy drinks (EDs) are caffeinated beverages, consumed mostly by the young individuals for the sake of getting an instant energy. They also contain some herbal extracts like guarana, ginseng and ginkgo biloba along with B-vitamins and taurine. The undesirable effects are mostly due to a high content of caffeine and glucose in it, which have both the short as well as long term implications. The increase usage of caffeinated beverages among the young population is a matter of concern.

Objectives: To estimate the metabolic consequences of energy drink consumption in Wistar albino rats.

Methodology: For this cross-sectional experimental study, thirty adult male Wistar Albino rats weighing between 250-300 grams were obtained from the animal house of BMSI. Three groups of equally divided number were made and labeled as A, B and C. Animals of Group A served as controls and were fed on a regular laboratory diet and water ad libitum. Animal of B & C groups, in addition to laboratory diet were also feed a Energy Drink in different dose orally via a gastric tube for a period of thirty days on daily basis. Fasting blood glucose and serum insulin were measured in control and treated groups at the end of experimental period.

Results: Fasting blood sugar was found to be significantly elevated in both the low-dose treated (126.80±8.69) as well as high dose treated animals (147.60±8.93) as compared to the control group animals (90.10±13.37). A dose dependent decrease in serum insulin was also observed in the experimental groups. The insulin concentration of group B & C was found to be 7.45±0.82 and 5.66 ±0.36 respectively as compared to 13.61±1.77 in group A animals.

Conclusion: Energy drink consumption can contribute to increase the risk of developing metabolic disorders by interfering the regular glucose metabolism.

Keywords: Energy drinks, glucose metabolism, metabolic disorders, serum insulin

Introduction:

Energy drinks (EDs) are caffeinated beverages, consumed mostly by the young individuals for getting instant energy. Eds also contain some herbal extracts like guarana, ginseng and ginkgo biloba along with B-vitamins and taurine[1]. A high content of glucose is a component of these drinks as well. To maximize the potential effect of caffeine on the body, many supplements are added by the manufacturing companies that greatly enhances its stimulatory potential[2].

The EDs were first marketed in USA in 1949, later they were launched in Europe in 1987. In the late 90's the

market was expanded globally and an exponential growth in terms of both its production and consumption was observed in that time period[3]. People consume EDs to seek an attentive behavior, to improve cognition, to increase the muscle strength and diminish the perceived feeling of fatigue. Manufacturers are now focusing more on the adolescents and young adults as their prime consumers with the main aim to provide energy boost following its ingestion. Gendered branding and marketing tactics have also emerged as an influential factor[4].

The undesirable effects are mostly due to the high content of caffeine and glucose, both has the short as well as long term implications[5]. A large content of caffeine and sugar i.e., about 80 mg and 28 g respectively are present in a 250ml serving of ED[6]. Caffeine is a methylxanthines that can readily cross the blood brain barrier and can be found in all body fluids. As it is a substance soluble to both water and lipid, it can target various organ systems of the body[7]. Literature has revealed significant side effects among the consumers. Tachycardia, insomnia, nervousness, tremors, increased urination and abdominal pain are some of the reported adverse outcomes following its usage. The high content of sugar is also a health hazard associated with these power boosting beverages[8]. The world is currently struggling with the epidemics of obesity and type II diabetes mellitus and the amount of glucose present in these beverages is a potential risk factor for developing both of these conditions[9].

Pancreas is a soft, lobulated organ, located retroperitoneal in the abdominal cavity. It lies in close relation with the stomach, duodenum and spleen. It acts both as an endocrine and exocrine organ. The endocrine part is associated with regulating the level of glucose in the blood while exocrine part releases the pancreatic juice, which is a combination of various enzymes that aids in the digestion and absorption of nutrients present in the ingested diet[10]. Various hormones and neurotransmitters play an integral role in the secretory activity of pancreas as it plays an essential role in maintaining the glucose homeostasis of the body. Scientific studies have proved that the caffeinated beverages alter the cytoarchitecture of the organ and resultantly would affect the functional capacity as well[11].

The increase usage of caffeinated beverages among the young population is a matter of concern. Rich taste, aggressive marketing and easy accessibility are the key factors that make EDs a choice of consumption. The risks of developing potential side effects needed to be addressed.

Objective:

To explore the effects of ED on the blood sugar metabolism in the Wistar Albino rats.

Methodology:

This cross-sectional experimental study was carried out in the department of Anatomy; basic medical sciences institute BMSI, Jinnah postgraduate medical center Karachi in October'2018 for a period of four weeks. The ethical approval was taken from the ethical committee of BMSI (Letter No. F.1-2/2018/BMSI-E.COMT/069/JPMC dated 28.09.2018). Thirty adult male Wistar Albino rats weighing between 250-300 grams were obtained from the animal house of BMSI. They were kept in well ventilated cages and a 12-hour day and night cycle was maintained. They were observed one week prior to the experimentation and fed on a regular laboratory diet and water ad libitum. Three groups of equally divided number were made and labeled as A, B and C. [12]

- Group A served as control
- Group B received ED at a dose of 7.5ml/day equivalent to 10mg/kg of body weight
- Group C received ED at a dose of 15ml/day equivalent to 20mg/ kg of body weight (Akande and Banjoko, 2011).

A commonly available ED in commercially packaged cans, each of 250ml were obtained from the local market (identity has been kept hidden for the legal purpose). The route of administration was oral via a gastric tube to all the animals of group B and C for a period of thirty days on daily basis. The animals were monitored for appetite, behavior and general well-being throughout the duration of study.

The fasting blood glucose (FBG) was recorded at the completion of experimental period, by using the glucostrips of glucometer (Accu Chek Active, Roche). It has a lancing device equipped with a lancet and a metered sensor with the provision of inserting a test strip. While measuring the blood glucose levels of experimental animals, the blood was drawn from the tip of the tail, with the help of the lancet, a drop of blood was applied on the test strip and then the metered device was turned on. The result was displayed in 30 seconds. Later on, the rats were anaesthetized by chloroform in a closed chamber. The animals were then dissected by giving a

midline abdominal incision. The blood samples were collected through cardiac puncture by using sterile syringes and kept in non-heparinized sample tubes. The serum was collected by centrifuging the samples at 3000 rpm and stored for biochemical analyses. Insulin was measured by using ELISA kit Thermo Scientific 7335 USA. The Thermo Scientific Pierce Rat Insulin ELISA Kit is an enzyme linked immunosorbent assay for measuring rat insulin in serum, plasma and cell culture media.

The statistical software SPSS v 20 was used for data analysis. All the data were presented as mean± S.D. The statistical analysis was done by using one-way analysis of variance (ANOVA) to evaluate the significance between mean values of control and treated groups. Values less than 0.05 were accepted significant statistically.

Results:

Of To study the effects of ED on the fasting blood glucose and serum insulin levels, the experimental animals were treated with two different doses of caffeinated beverage. Fasting blood glucose and serum insulin were measured in control and treated groups (table-I, table-II and figure-I respectively) after finishing the experimental period.

The fasting blood glucose (FBG) of low dose treated animals was found to be significantly higher (126.80±8.69) as compared to the control group animals (90.10±13.37). The control group animals remain active and healthy throughout the course of study; they gradually gained weight while their appetite and behavior remained normal. The low-dose treated group also showed no significant change in their behavior and activities with the exception that they slightly lose their appetite.

The animals treated with high dose of ED significantly showed an increase in FBG (147.60±8.93). The animals in this group were restless and showed agitated behavior. They had very poor appetite with remarkable weight loss. They had lax skin with lusterless falling hair from their bodies. Two animals in this group become severely morbid and died in the 2nd and 3rd week respectively.

Table-II is demonstrating the serum insulin response of rats treated with variable doses of ED and their comparison with control group animals. The mean value of serum insulin control group A was found to be 13.61±1.77 while a decrease in the mean value of serum insulin in low dose treated group B was noted i.e.,7.48 ±0.82.

Table: No 1. Comparison of fasting blood glucose (mg/dl) in different groups of Albino Rats.

Groups (n=10)	Treatment Received	Blood glucose (mg/dl)		P-value
		Mean	± SD	
A	Control	90.10	± 13.37	<0.05*
B	Treated – low dose	126.80	± 8.69	<0.05*
C	Treated – high dose	147.60	± 8.93	<0.01*

Statistical Comparison within groups	
Group B and A	0.001 **
Group C and A	0.001 **
Group C and B	0.001 **
P < 0.05 (*) statistically considered significant P < 0.01 (**) statistically considered highly significant	

Where n is the number of albino rats
Data is presented as Mean ± S.D (Standard Deviation)

Table: No 2. Comparison of serum insulin levels (µl/ml) in different groups of Albino Rats.

Groups (n=10)	Treatment Received	Serum insulin (µl/ml)		P-value
		Mean	SD	
A	Control	13.61	±1.77	<0.01*
B	Treated – low dose	7.45	±0.82	<0.01*
C	Treated – high dose	5.66	±0.36	<0.01*

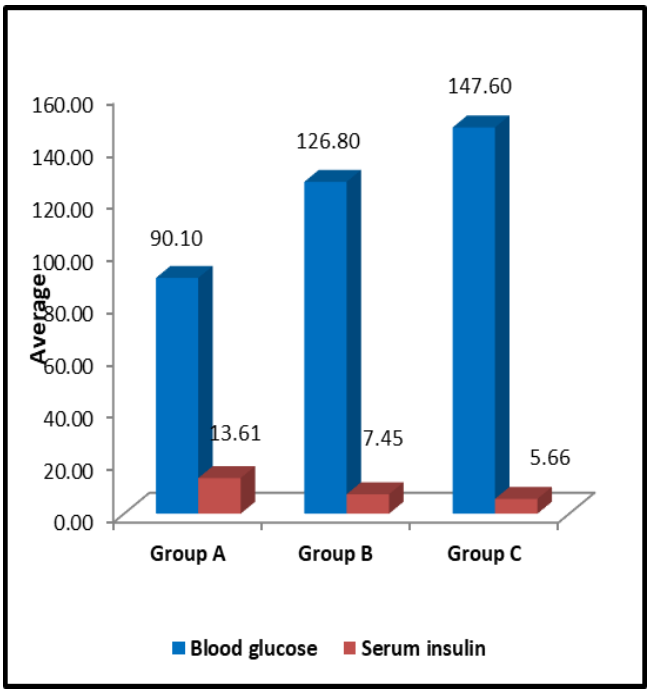
Statistical Comparison within groups	
Group B and A	0.001 **
Group C and A	0.001 **
Group C and B	0.005 **
P < 0.05 (*) statistically considered significant P < 0.01 (**) statistically considered highly significant	

Where n is the number of albino rats
Data is presented as Mean ±S.D (Standard Deviation)

Group C animals were kept on high dose of ED throughout the study duration, their blood sugar levels reflected profound alteration in serum insulin levels

that was found to be 5.66±0.36. The data revealed a significant decrease in insulin concentration in group C animals when compared with group A and group B respectively (P-value <0.001).

Figure: No 1. Comparison of blood glucose (mg/dl) and serum insulin levels in different Groups of Albino Rats.



Group C animals were kept on high dose of ED throughout the study duration, their blood sugar levels reflected profound alteration in serum insulin levels which was found to be 5.66±0.36. The data revealed a significant decrease in insulin concentration in group C animals when compared with group A and group B respectively (P-value <0.001).

Discussion:

The current study was aimed to observe the levels of fasting blood glucose and serum insulin following the administration of ED for a period of 30 days. An animal model was created, composed up of thirty adult male Wistar Albino rats weighing between 250-300 grams They were categorized into three groups each having 10 animals, one group was receiving the regular laboratory diet while the other two kept on two different doses of ED i.e.,7.5ml and 15ml on daily basis. On completion of experimental period all the animals were evaluated for the measurement of biochemical parameters.

Studies have evaluated that consuming the EDs leads to significant structural collapse to the cyto-architecture of pancreas. The microscopic structure demonstrates histopathological alterations in both the exocrine as well as endocrine part of the organ.[13] A possible mechanism behind the damage is the release of reactive oxygen species that induces the pro-inflammatory environment in the body by decreasing the concentration of anti-inflammatory markers on the other hand the serum levels of pro-inflammatory cytokines is increased. [14] The present study was therefore hypothesized to observe the functional deficit occurred in pancreas by measuring the serum insulin and fasting blood glucose levels.

The β-cells of islets of pancreas are mainly responsible for the secretion of insulin therefore plays a pivotal role in regulating the glucose homeostasis in the body, which is the maintenance of glucose at a steady-state level.[15] Results of the current study showed a dose dependent exacerbation of fasting blood glucose levels in the animals treated with ED (P-value <0.001). These findings are in accordance with Nasira et al [16] who gave Power house energy drink to experimental animals via a gastric tube once daily for a period of 4 weeks and found a positive correlation between ingestion of energy drink and raised levels of blood glucose. A single can of 250ml of ED contains about 25 g of sugar, which results in no additional nutritional value and only aggravates the sugar concentration in the blood. [17] Sudden fluctuation in the blood glucose levels is often associated with symptoms like confusion, nervousness, seizures and rapid heart rate. Consumption of an increased uptake of glucose rich products produces deleterious effects on overall health system of the body. There's either decreased production of the insulin by the β-cells of islets of Langerhans's or a resistance for the insulin is produced by the peripheral tissues of the body that eventually results in metabolic disorders like obesity and type-II diabetes mellitus.[18]

During present study serum insulin of all the experimental animals was also measured as it is a key factor driving the metabolic syndrome. The level of serum insulin was found to be significantly decrease in the ED treated animals (p value<0.005). These results are in line with the observations made by Haroun et al.[19], who fed the experimental rats with an intra-peritoneal injection of a famous energy drink for a period of 4

week and later observed the serum insulin and blood glucose levels. They described that intake of caffeinated beverages reduces the sensitivity of insulin by peripheral tissues of the body and resultantly metabolic consequence are being observed. This finding is although in contradiction to the observation given by Nasira et al 16, who found an increase in both serum insulin level and blood glucose in their diabetic animals and has explained that this increase was due to insulin resistance produced in the body.

Many investigations have also proved that caffeine is able to enter the brain and stimulate the release of stress hormones. These hormones are known to affect the insulin and produces harmful effects on blood sugar metabolism. Excessive sugar is also likely to be associated with decrease in serotonin secretion from the brain and hampers its functions of modulating the regulation of mood and behavioral sensitivity. [20, 21]

Findings of the present study clearly showed that use of caffeinated beverages have implications on the glucose metabolism of body. An extended human trial on a larger scale needs to be done, in order to explore the potential outcomes of these beverages. As well as future researches should be conducted to understand the risk and possible interventions for stimulating protective consumption of energy drinks. Evidence based upper limit for the amount of caffeine and sugar should be established to be used by children and young adults, in order to prevent them from potential harmful effects.

Conclusion:

The consumption of energy drink has negative effect on the metabolism leading to serious health concerns. Legislation should be done to label health concerns over the label of energy drinks.

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Conflict of interest:

The authors declare that they have no conflict of interest.

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