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LIPTON TEA AND COFFEE COMPLICATE PATHOGENESIS OF ITS IN-DUCED GASTRIC ULCER WITH FLUCTUATIONS IN AMYLASE AND PROTEIN LEVELS

Research

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CONFLICTS OF INTEREST

There are no conflicts of interest for any of the authors.

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ABSTRACT

Gastric and plasma alpha amylase and proteins concentrations were evaluated in male and female albino rats fed with Lipton tea and coffee induced gastric ulcer. A total of ninety-six male and female albino rats were studied for 28 days. There was no significant difference (p>0.5) in gastric alpha amylase concentrations between the two beverages and the male and female rats. Also there was no significant difference in plasma alpha amylase between Lipton tea and coffee, (P>0.05) and between sexes, (p>0.05). However, the plasma alpha amylase values were very low as compared to control rats without gastric ulcer. There was significant difference in plasma protein concentrations between rats fed with coffee and Lipton tea, (p<0.05) as more proteins concentrations were recorded in coffee feeding than in Lipton tea. Also there was significant difference in plasma protein concentrations in the weekly studies of 7, 14, 21, 28 days (p>0.05). And no significant differences (p>0.05) between the sexes and rats fed with caffeinated and decaffeinated coffee. Cumulatively, both gastric and plasma proteins showed no significant difference, (p>0.5) in their concentrations in Lipton tea and coffee. But there was significant difference (p<0.05) in gastric protein between caffeinated and decaffeinated coffee. Significant differences, (p<0.05) also existed in gastric protein concentrations between 7, 14, 21 and 28 days. Both Lipton tea and coffee induced gastric ulcer, reduced amylase and increased protein concentrations and such may be associated with pancereatitis, kidney disease, multiple myeloma, bone marrow disorder, chronic inflammatory conditions and amyloidosis.

KEY WORDS: Amylase, proteins, Lipton tea, coffee, gastric ulcer.

INTRODUCTION

Amylase is an important gastro-intestinal enzyme responsible for the metabolism of carbohydrate into disaccharide also called maltose and absorbable monosacharides. The enzyme is secreted by the salivary gland particularly the parotid gland. It is otherwise called ptyalin, (Saladin, 2004). Thus, amylase is one of the first steps in the digestion of starch. However, the action of amylase in the stomach is limited due to high acidity, P_H of 1.5 – 1.6, whereas that of saliva is 5.2-7.0. (Rebecea 2015). Amylase is also produced by the pancreas also for the metabolism of carbohydrate. Increase or reduction in amylase as in hyperamylasemia and hypoamylasemia are sensitive indicators in evaluating the clinical functions of the pancreas e.g. pancreatitis. Hyperamylasemia may also result from tumors as in pancreas tumors, prostate, lungs and in ovary. It could be in kidney failure i.e. hyperamylasemia, also in medication, etc (Matthew, 2016).

The normal level of amylase is 0-137U/L. The hypoamylasemia indicates low serum amylase and is linked with cardio-metabolic anomalies e.g. obesity, diabetes, due to insufficient insulin and resistance link with the pancreas (Kei, 2016). Salivary amylase could also decrease which is associated with amylase gene variation (AMYI) and also associated with insulin resistance, but the mechanism is not fully understood. It is mainly based on the dysfunctioning of the salivary glands that secret the enzyme and that which involves the pancreas is often due to inflammation of the pancreas, (Luca, 2005) and obstruction of the pancreatic duct, acinar cell damage, (Moren, 2003). The association of hyper-amylase with peptic ulcer or gastro-duodenal ulcer is due to the peritoneal lymphatic absorption of fluid which has pancreatic enzyme due to perforation and this is associated with high mortality (Frank, 1960). Gastric ulcer is the disease of the gastro intestinal system that affects mainly the stomach. The disease is associated with high level of acidity in the stomach, intake of foods which trigger the hydrochloric level, histamine and gastrin (Jimmy, 2013). It is also associated with intake of non-steroidal anti-inflammatory drugs, (Gisbert, 2004). The important morbidity which is associated with mortality is bleeding which is often internal (Tang 2013) (Ikikawa, 2005) (Ogra, 2002), (Lin 1993). Also perforated peptic ulcer is associated with high mortality rate, (Kenneth 2013) (Frank 1960). The incidence rate of the disease varies, in Nigeria 5% rate is reported in autopsy findings, (Olubuyide, 1989).

The variation in the incidence is based on many factors which are the causative agents and associated ones. The treatment of gastric ulcer is based on the use of antacids, H₂- blockers; cimetidine, ranitidine and the proton pump inhibitors (pp1) e.g. Omeprazole (Fong, 2015), and gastric protective agents e.g prostaglandin analogue; misoprostol (Zajac, 2013) Lipton tea and coffee have been incriminated as causative agents of gastric ulcer, (Jimmy, 2013).

The mechanism of their action is the stimulatory release of gastrin and histamine which increase the hydrochloric acid levels and hence the ulceration. The two beverages also reduce the concentration of prostaglandin which is meant to protect the gastric barrier, and the resultant ulceration.

Proteins are the building blocks of the body and associated with many funct; protection, as in immune system, metabolism as in enzymatic reaction, in coagulation, etc (Guyton, 2011). High or low level of protein signifies anomalies and basically in gastro-intestinal system, low presence may lead to kwashiorkor. Protein is acted upon by pepsin to polypeptide and the amino acids. In gastric ulcer this may be impaired as the gastric witnessed lesions with abnormal lining. The aim of the study was to find out any relationship in the concentration of amylase and protein in Lipton tea and coffee induced gastric ulcer. This will aid in the differential diagnosis of this disease associated with the two beverages and enhance in its management.

MATERIALS AND METHODS

A total of ninety-six (96) male and female swiss albino rats were used for both Lipton and coffee induced gastric ulcer i.e. 48 in each group. The animals were kept in well ventilated university of Jos animal house, fed with pellets and clean water and were cared for according to the regulation of Institute for animal Ethical Committee (IAEC), and all the ethical standard laid down in 1964 declaration of Helsinki were observed.

PREPARATION OF LIPTON TEA AND COFFEE

A sachet of Lipton that weighed 2.38g was used. It was dissolved in 250ml of hot water and brewed (kept) for 5 mins. 1160mg/kg was obtained after the shaft weight was deduced from the sachet weight. The caffeine concentration in the Lipton was determined using high performance liquid chromatography. This same method was applied in coffee study. 0.0lml of both tea and coffee were arrived at which translated to 0.3mg/ml of caffeine per weight of the animals from the average weight of man; 70kg.

ADMINISTRATION OF LIPTON TEA AND COFFEE FOR GASTRIC ULCER INDUCEMENT

After the tea and coffee had stayed for 20 - 40 mins for heat reduction, 0.0lml/kg was administered orally using canula to the animals per body weight, for the period of 7, 14, 21 and 28 days.

Confirmation of Ulcer: At 7, 14, 21 and 28 days after tea/coffee administration the rats were anaesthetized with chloroform and the stomach removed. It was opened, the contents stored for other analysis. The stomach were washed with clean water and ulceration observed using hand lens.

Amylase Test: Gastric contents were obtained from the stomach of killed rats and the methods of Kruse (1989) and (Klein, 1986) were used in determining gastric amylase and protein. The procedure of the supplier for plasma analysis was used but modified for gastric amylase. Plasma was also analyzed for amylase using the supplier

procedure.

Preparation of Sample: 0.02ml of the test sample i.e. gastric content was mixed with 1ml of reagent buffer and substrate in a cuvette. The mixture was placed in water bath for 3min to initiate enzyme reaction with the substrate. After 3mins the mixture in the cuvette was introduced into spectrophotometer and the concentration of amylase read at 405nm wave-length at 1min interval for 4mins and the results obtained as average of the time interval. The same procedure was done with 0.02ml of plasma from 5ml of blood from each rat.

Total protein Assay: The methods of Beckman Unicel (2011 – 2012) were used. The reagent was supplied by Randox cat. No. J.P. 245.

Gastric Contents: 0.02ml gastric contents was used, reagent blank was 0.02ml as distilled water, standard, 0.02ml in three cuvettes, 1,0ml of sodium hydroxide was added to the test sample in cuvette and other 2 cuvettes. The mixture was allowed to stand for 30 minutes at 25°c and the absorbance of the test sample and the standard was measured against that of the reagent blank. The results were recorded for each animal for 7, 14 21 and 28 days. Same procedure was applied for 0.02ml of plasma sample from 5ml of blood from each rat.

Calculation: Total protein = $\underline{\text{Absorbance of sample/g/dl}}$

Absorbance of standard

Statistical Analysis: Analysis of variance was used (ANOVA)

RESULTS

Effects of Lipton and Coffee on Gastric amylase and Protein in Rats with Gastric Ulcer

Male and female rats fed with Lipton tea showed no significant difference in their gastric protein, p>0.05, at p=0.4. Male and female rats fed with Lipton tea and coffee (caffeinated and decaffeinated) showed no significant difference in gastric amylase between the beverages p>0.05, p=8.6 table I. Also there was no significant difference between the male and female rats in gastric amylase fed with Lipton tea and coffee respectively, p>0.05, p=0.7, p>0.05, p=0.03 table I.

However, there was significant difference in gastric protein between caffeinated and decaffeinated coffee, p<0.05, at p = 0.001, with more gastric protein in caffeinated than decaffeinated coffee. There was significant difference in gastric protein between Lipton tea and coffee, p<0.05, at P = 0.0009 with more gastric protein in Lipton tea feeding than coffee generally. A significant difference also existed between, 7, 14, 21 and 28 days in gastric protein p<0.05 at P = 0.013, table 2.

| | <u>Day 7</u> | | <u>Day 14</u> | | <u>Day 21</u> | | <u>Day 28</u> | |
|--------------------|------------------|------------------|------------------|------------------|-------------------|------------------|------------------|------------------|
| Beverages | <u>M</u> | <u>F</u> | <u>M</u> | <u>F</u> | <u>M</u> | <u>F</u> | <u>M</u> | <u>F</u> |
| <u>Lipton tea</u> | <u>13.8</u> | <u>13.21</u> | <u>14.21</u> | <u>15.01</u> | <u>16.89</u> | <u>16.26</u> | <u>18.85</u> | <u>18.72</u> |
| | <u>p>0.05</u> | <u>p>0.05</u> | <u>p>0.05</u> | <u>p>0.05</u> | <u>p.>0.05</u> | <u>p>0.05</u> | <u>p>0.05</u> | <u>p>0.05</u> |
| | | | | | | | | |
| Coffee | 125.28 | 125.26 | 126.05 | 126.01 | <u>131.18</u> | 131.26 | 133.54 | 133.99 |
| (caffeinate d | <u>p>0.05</u> | <u>p>0.05</u> | <u>p>0.05</u> | p>0.05 | <u>p>0.05</u> | p>0.05 | <u>p>0.05</u> | <u>p>0.05</u> |
| Coffee | <u>125.06</u> | 125.01 | 122.71 | 122.82 | <u>126.18</u> | <u>126.18</u> | <u>129.07</u> | <u>129.21</u> |
| (decaffeina ted | <u>p>0.05</u> | <u>p>0.05</u> | p>0.05 | <u>p>0.05</u> | <u>p>0.05</u> | <u>p>0.05</u> | | |

Values expressed as mean n = 24. Control value = 76.5 - 127.5U/L.

Table 1: Effect of Lipton tea and coffee on gastric alpha amylase (U/L) in male and female rats in gastric ulcer

SIFT DESK

| | Day 7 | | Day 14 | | Day 21 | | Day 28 | |
|-----------------|--------|--------|---------|--------|--------|--------|--------|--------|
| Beverages | M | F | M | F | M | F | M | F |
| Lipton tea | 2.6 | 2.6 | 4.2 | 4.2 | 5.8 | 5.7 | 6.5 | 5.9 |
| | p>0.05 | p>0.5 | p.>0.05 | p>0.05 | p>0.05 | p>0.05 | p>0.05 | p>0.05 |
| Coffee | 2.3 | 2.4 | 3.6 | 3.6 | 3.6 | 3.5 | 4.2 | 4.2 |
| (caffeinated | p>0.05 | p>0.05 | p>0.05 | p>0.05 | p>0.05 | p>0.05 | p>0.05 | p>0.05 |
| Coffee | 1.7 | 1.7 | 2.0 | 2.0 | 2.8 | 2.7 | 3.1 | 3.1 |
| (decaffeinat ed | p>0.05 | p>0.05 | p>0.05 | p>0.05 | p>0.05 | p>0.05 | p>0.05 | p>0.05 |

Table 2: Effects of Lipton tea and coffee on gastric protein in male and female rats with gastric

Effect of Lipton tea and Coffee on Plasma Alpha Amylase and protein in Rats with Gastric Ulcer

Male and female rats fed with Lipton tea and coffee showed no significant difference in plasma alpha amylase between the two beverages, p<0.05 at p = 58 table 3. Also there was no significant difference in plasma alpha amylase between male and female rats fed with Lipton tea only, p<0.05 at p = 0.65. There was no significant difference in plasma alpha amylase between male and female rats fed with caffeinated coffee only, p<0.05, at p = 0.27, and decaffeinated coffee; p<0.05, at p = 3.11. The plasma amylase concentrations were very low compared with control rat without gastric ulcer.

| | Day 7 | | Day 14 | | Day 21 | | Day 28 | |
|---------------------|--------|--------|--------|--------|--------|--------|--------|--------|
| Beverages | M | F | M | F | M | F | M | F |
| Lipton tea | 6.75 | 7.21 | 26.7 | 25.8 | 28.1 | 29.0 | 36.75 | 35.8 |
| | p>0.05 |
| Coffee (caffeinated | 32.7 | 32.9 | 33.6 | 33.7 | 34.1 | 33.1 | 35.9 | 34.6 |
| | p>0.05 |
| Coffee | 12.6 | 11.8 | 14.2 | 13.8 | 14.7 | 15.2 | 15.7 | 14.9 |
| (decaffeinated | p>0.05 |

Values expressed as mean = 24.Control values = 76.5 - 127.5m/l.

Table 3: Effects of Lipton tea and coffee on plasma alpha amylase in male and female rats in gastric ulcer

| | Day 7 | | Day 14 | | Day 21 | | Day 28 | |
|------------------------|----------------|----------------|----------------|----------------|----------------|----------------|----------------|----------------|
| Beverages | M | F | M | F | M | F | M | F |
| Lipton tea | 58.5 | 57.1 | 38.2 | 38.1 | 21.7 | 22.0 | 15.8 | 14.7 |
| | P<0.05 |
| Coffee (caffeinated | 64.6 P<0.05 | 65.1 P<0.05 | 69.2 P<0.05 | 69.2 P<0.05 | 80.1 P<0.05 | 79.1 P<0.05 | 59.2 P<0.05 | 87.8 P<0.05 |
| Coffee (decaffeinate d | 64.1 P<0.05 | 63.8 P<0.05 | 64.2 P<0.05 | 64.2 P<0.05 | 65.1 P<0.05 | 66.8 P<0.05 | 68.8 P<0.05 | 67.2 P<0.05 |

Table 4: Effects of Lipton Tea and Coffee on Plasma Protein in Male and Female Rats With Gastric Ulcer

Male and female rats fed with Lipton tea and coffee showed significant difference in plasma protein between the two beverages as more protein were induced by coffee feeding than Lipton tea, (p<0.05) at 0 = 0.001, table 4. But there was no significant difference in plasma protein in same rats fed with Lipton tea and coffee between 7, 14, 21 and 28 days (p>0.05) at p = 0.96.

Also there was no significant difference in plasma protein between male and female rats fed with Lipton tea only p>0.05, at p=0.02. There was no significant difference in plasma protein between male and female rats fed with caffeinated and decaffeinated coffee respectively, p>0.05, at p=0.16, at p=0.07. Generally, in gastric and plasma protein no significant difference was observed in male and female rats.

DISCUSSION

The study has shown that both Lipton tea and coffee affected the plasma, gastric protein and amylase concentration. And more inducement by the caffeinated coffee than decaffeinated and Lipton tea as the days of feeding progressed. High protein level may induce more gastric acidity as one of the inducers of gastric secretion (Ghyton 2011). Also, increase protein concentration may lead to more gastric release which showed impact on the histamine and HCL release (Korman, 1971).

And in this study hydrochloric acid (HCL) gastrin and histamine raised levels are associated with Lipton tea and coffee feeding. However, the effect of high plasma protein implies high circulatory protein which could precipitate with serious physiologic consequences. For example hemoglobin precipitation and blockage of renal tubules and renal shut-down (Dakshinamurth, 2009) (Emara, 2013). Raised plasma and gastric amylase in Lipton tea implies over synthesis of the enzyme which major function is to act on starch converting such to maltose, glucose, etc. (Lenninger, 2000). However, low amylase suggests, pre-eclampsia, kidney disease (healthiness, http.//healthcare).

The effects of raised amylase suggest pancreatitis (Luca, 2005) (Moren, 2003) which is an inflammation of the pancreas and over-stimulation in the gastric of rats with ulcer as the rats without gastric ulcer had normal plasma and gastric amylase. The plasma and gastric increase in protein and amylase in rats fed with Lipton tea and coffee than in those with gastric ulcer and not fed with the beverages in this study has confirmed the enhanced effect of the beverages on plasma and gastric protein and amylase. This means that it is not only the ulcer that raised the protein and amylase values, the effect of presence of causative agents of ulcer has therefore complicated the parthenogenesis of gastric ulcer. This implies that anyone with gastric ulcer should not take or drink Lipton tea and coffee for effective management of the disease to be attained. Also in the diagnosis and treatment of peptic or gastric ulcer information on the daily nutrition of the patients need to be obtained.

RECOMMENDATION: Lipton tea and coffee should be taken with caution particularly patients with peptic ulcer to avoid metabolic hinderances and complications of the disease situation.

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