Pineapple Juice Administration and Gastric Ulcer in Wistar Rats

O.A. Oyesola, T.O. Oyesola and A.I. Izagbo

Fruits have been part of the human diet and supplements. Present study was conducted to investigate pineapple juice (PJ) effects on Gastric Ulcer (GU), since dietary substances or supplements may predispose someone to GU. Thirty-six male wistar rats, weighing 180-200 g were used. They were divided into six groups A-F. Groups A served as control. Group B was not given PJ. Groups C-F received 0.5, 1.0, 1.5 and 2 mL of PJ, respectively. After 30 days, GU was induced with indomethacin. Ulcers Indices (UI) were scored, Total Protein Content (TPC), Catalase (CAT), Superoxide Dismutase (SOD) activities and Lipid Peroxidation (LPP) were determined from stomachs’ tissue. Significances were taken at p<0.05. UI showed significant increases when groups B-F were compared with control. A significant reduction was seen in TPC when groups B-F were compared with control. Comparison of CAT activities between control and groups B-F showed significant reduction in groups B-F and a significant increase in SOD in groups B-F. Activities of anti-oxidant enzymes in group B were compared with groups C-F. Observation showed that CAT activity increased significantly in groups C and D and SOD activity reduced significantly in groups D-F. Observation from LPP in the gastric mucosa showed increase which was significant in group B, when control was compared with groups B-F. When group B was compared with groups C-F, significant increases were observed in groups C and D. Conclusion from results suggested that PJ consumption may predispose, consumer to gastric ulceration because of its tendency to reduce TPC, increase UI and anti-oxidant enzymes.

Key word: Pineapple-juice, gastric ulcer, antioxidant-enzymes
INTRODUCTION

Over the years fruits have been part of the human diet and food supplements. They are considered as healthy food supplements because they contain high quantity of water, carbohydrates, proteins, vitamins A, B1, B2, C, D and E, and minerals such as Ca, Mg, K, Zn and Fe (Okwu and Emenike, 2006). Fruits are recommended internationally as nutrient supplements in addition to their dietary importance. Nutritionists have advised that eating at least five portions of fruits and vegetables daily can help people to maintain good health throughout their lives, protecting them from heart disease and cancer (Wenkam, 1990, Food Commission, 2009). Fruit consumption has been reported to be beneficial to health and contribute to prevent degenerative processes, particularly lowering the incidence and mortality rate of cancer and cardio- and cerebro-vascular diseases (Rapisarda et al., 1999). A diet rich in fruit and vegetables was associated with smaller gains in body mass index (BMI) (Newby et al., 2003).

Pineapple (Ananas comosus) can also be referred as the “King of Fruit” (Bartolome et al., 1995). Hawaii, Philippines, Caribbean area, Malaysia, Taiwan, Thailand, Australia, Mexico, Kenya and South Africa have been known to cultivate pineapple extensively. Pineapple juice was found to be a rich source of vitamin C which is effective in killing parasites such as worms. It is rich in magnesium and is needed for the body to build bone and connective tissue. Pineapples have been used in traditional tropic medicine for ailments ranging from constipation to jaundice and to relieve sore throats and bronchitis. Heart patients have found pineapple useful but coagulation time has been found to increase which is not good for those with kidney and liver diseases (Nieper, 1977). In 1891, an enzyme called bromelin was isolated from the flesh or the pineapple and discovered to comprise a complex mixture of sulphydryl containing proteolytic enzymes in addition to a number of non-specific components like glycoproteins, phosphatase, peroxidases, glycosidases, cellulases and carbohydrates. The extract also contains a proteinase inhibitor consisting of inhibitors (Morita et al., 1979). Pineapple juice also destroys harmful proteins in the stomach and intestines. It promotes the digestive processes because only protein compound that have broken down into smaller components can be absorbed by the intestines and enter the bloodstream (Kelly, 1996). Pineapple juice has been found to be effective in dissolving mucus and assisting recovery from tuberculosis (Taussig and Nieper, 1979; Tochi et al., 2008).

It is known that the human gastrointestinal tract serves two main purposes; the main doorway for the nutrients and also as a barrier to the external environment. The duality of this pattern may prevent the great effect of some nutrient to preserve the integrity and function of the gastrointestinal mucosa itself (Roberfroid, 2000). The epithelial cells of the gastrointestinal tract are the primary boundary between the ingested nutrients, the blood and the lymph streams. The cells of the gastrointestinal tract that depend on the luminal and blood stream flow include, the epithelial cells, mucosal cells, enteroendocrine cells, intraepithelial lymphocytes and the multiple cells types of the laminal propria. ‘Functional food’ describes food or nutrients whose ingestion leads to physiological changes which are quite separate and distinct from those associated with their roles as nutrients (Koletzko et al., 1998).

Gastric ulcer is a form of peptic ulcer characterized by the erosion of the mucosal surface of the stomach. Peptic ulcer is due to the sum of two groups of forces (defensive and offensive) acting upon the gastrointestinal mucosa. The most aggressive factor is the hydrogen ions, the acid produced by the parietal cells in the stomach and pepsin, while the defensive force is from the mucosal wall and mucus, being the pathogenesis of gastro duodenal ulcer. A large number of people in the world are affected by gastric and duodenal ulcers. Some of the causes of the these disorders are: dietary intake (tea, pepper, beverage) stress, smoking, nutritional deficiencies and ingestion of non-steroidal anti-inflammatory drugs (Nash et al., 1994; Basil and Howard, 1995; Ibu et al., 1986; Olaeye et al., 2006; Ibronke et al., 1997).

One of the predisposing factors of gastric ulcer is dietary intake: similarly improper eating habits may result into gastro-intestinal dysfunctions (Okike, 1997), uncontrolled eating habit has brought about sickness (Amure, 1990), diet and dietary habits have been shown to be linked to most gastrointestinal disorders, however beneficiary a diet may be if proper precaution were not taken (Hyams, 1983). Due to the presence of sorbitol of fructose in excess of glucose in apple and pear juices, the malabsorption was taken as a factor in chronic nonspecific diarrhea (Thomas, 1957; Kelly, 1980). Similarly gastric emptying is highly influenced by volume of the stomach and volume ingested, calories, osmolality, the amount of acid that the stomach produced, as well as other factors associated with physiological factors e.g., splanchic blood flow, body position, posture and electrolyte balance (Moore et al., 1984; Fraser et al., 1990; Marzio et al., 1991; Horowitz et al., 1993; Amidon et al., 1991).
Pineapple is a non-citrus fruit with attractive flavor and refreshing sugar-acid balance (Bartolome et al., 1995). It is available fresh or canned and as juice (Bartolome et al., 1995). A 100 g pineapple contains 47-52 calories, 85.3-87.0 g water, 0.4-0.7 g protein, 0.2-0.3 g fat, 11.6-13.7 g carbohydrate, 0.4-0.5 g fiber, 0.3-0.4 g ash, 17-18 mg calcium, 8-12 mg phosphorus, 125-146 mg potassium, 1-2 mg sodium and 0.5 mg iron (Duke, 1983). Pineapple contains 12-15% sugars (2/3 sucrose% 1/3 glucose and fructose). Pineapple contains between 0.6 and 1.2% of acid (87% citric acid and 13% malic acid) (Samson, 1986; Adhikary et al., 1987). Pineapple has a pH of 3.71 which is acidic and the acidity percentage is 53.5%. Geographical, cultural and seasonal harvesting and processing determine the composition of pineapple juice.

In view of the composition of pineapple juice vis-à-vis gastric disorder, this study is conducted to investigate the involvement of pineapple fruit juice consumption in the etiology of ulcer because of its wide range of uses and composition.

MATERIALS AND METHODS

Preparation of pineapple fruit juice: The pineapple fruits were obtained from new Benin market in Edo State of Nigeria. The juice was extracted with a juice extractor after peeling. The coring and macerating of the pineapple fruit was done by the juice extractor. The juice was collected and stored in a clean bottle kept inside a refrigerator (deep freezer) for preservation before administration to the rats.

Animals grouping and administration of pineapple: Thirty-six male Wistar rats weighing between 180-200 g were used for this study. The animals were obtained from an animal house in Benin, Edo state Nigeria. They were transferred to the animal house of Igbinedion University Okada. They were kept in separate cages of six animals per cage constituting a group. They were allowed four weeks to acclimatize, prior to commencement of the experiment. The animals in groups were labeled from A to F. Group A and B were both control groups for this study. They were given free access to water under standard condition of temperature and humidity. Group A were animals not given pineapple juice and ulcer not induced. Group B were animals not given pineapple juice but ulcer was induced in them. Group C were animals given 0.5 mL of pineapple juice extract and latter induced ulcer. Group D were animals which were given 1 mL of pineapple juice extract and latter induced ulcer. Group E were animal which were given 1.5 mL of pineapple juice and latter induced ulcer. Group F were animal given 2 mL of pineapple juice and latter induced ulcer. Pineapple juice administration lasted for twenty-one days.

Ulcer induction and determination: Ulcers were induced with indomethacin dissolved in Sodium bicarbonate solution of 5 mg per 500 mL distilled water. Indomethacin was administered at 40 mg kg⁻¹ b.wt., after 24 h that food had been withdrawn from them. Four hours after, the animals were opened under anesthesia (Sodium Pentobarbitone) at 60 mg kg⁻¹ b.wt. intra-peritoneally and pyloric ligation of the stomach was performed. At another 4 h the stomachs were removed and opened along the greater curvature, washed in normal saline to remove debris and pinned on a cork for ulcer scoring. The wound in the glandular were located with a hand lens. Ulcers were scored using the typical criteria as shown in Table 1 (Rifat-uz-Zaman et al., 2004; Tanaka et al., 1993).

Tissue preparation: The mucosal tissue from each animal was scraped from the stomach with a blunt knife and the tissue was weighed, transferred to the ice-cooled test tube and homogenized. The homogenate was then centrifuged at 12,000 rpm at 4°C, for 10 min. Supernatant aliquot and stored at−70°C until determination of total protein, Catalase (CAT), Superoxide Dismutase (SOD) and Malondialdehyde (MDA).

Total protein determination: The protein content of the tissue samples was estimated by the method of Lowry et al. (1951) using bovine serum albumin as a standard.

Determination of catalase activity: Activity of catalase in gastric mucosa was determined according to the procedure of Goth (1991) by following the absorbance of hydrogen peroxide at 230 nm and pH 7.0.

Determination of Superoxide dismutase activity: SOD activity in the gastric mucosa was determined by measuring the inhibition of autoxidation of epinephrine at pH 10.2 at 30°C by the method of Misra and Fridovich (1972). One unit of SOD activity represents the amount of SOD necessary to cause 50% inhibition of adrenaline autoxidation.

Table 1: Criteria for scoring ulcer

<table>
<thead>
<tr>
<th>Criteria</th>
<th>Ulcer type</th>
<th>Ulcer score</th>
</tr>
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<tbody>
<tr>
<td>Normal stomach (no Ulcer)</td>
<td>0.0</td>
<td>0</td>
</tr>
<tr>
<td>Punctuated hemorrhage or Pin point Ulcer</td>
<td>0.5</td>
<td>10</td>
</tr>
<tr>
<td>Two or more small hemorrhage ulcer</td>
<td>1.0</td>
<td>4</td>
</tr>
<tr>
<td>Ulcer greater than 3mm in diameter</td>
<td>2.0</td>
<td>5</td>
</tr>
<tr>
<td>Total</td>
<td>0.0</td>
<td>5</td>
</tr>
</tbody>
</table>

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Table 2: Ulcer index, total protein content, anti-oxidant activities and lipid peroxidation upon administration of pineapple juice in wistar male rats

<table>
<thead>
<tr>
<th>Groups</th>
<th>Treatments</th>
<th>Total protein (μg g⁻¹)</th>
<th>Catalase (CAT) (unit g⁻¹)</th>
<th>Superoxide dismutase (SOD) (unit g⁻¹)</th>
<th>Malondialdehyde (MDA) (μmol g⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>No Ulcer, No Juice</td>
<td>2.23±0.18</td>
<td>5.49±0.14</td>
<td>2.80±0.13</td>
<td>2.15±0.28</td>
</tr>
<tr>
<td>B</td>
<td>Ulcer Induced, No Juice</td>
<td>1.40±0.78</td>
<td>2.45±0.49*</td>
<td>5.80±0.11*</td>
<td>3.03±0.36</td>
</tr>
<tr>
<td>C</td>
<td>0.5 mL Juice, Ulcer Induced</td>
<td>1.65±0.13</td>
<td>3.21±0.24*</td>
<td>4.31±0.25*</td>
<td>4.48±0.46*</td>
</tr>
<tr>
<td>D</td>
<td>1.0 mL Juice, Ulcer Induced</td>
<td>1.4±0.11</td>
<td>4.08±0.51*</td>
<td>5.00±0.50*</td>
<td>3.58±0.20*</td>
</tr>
<tr>
<td>E</td>
<td>1.5 mL Juice, Ulcer Induced</td>
<td>1.62±0.05</td>
<td>3.69±0.52*</td>
<td>4.34±0.59*</td>
<td>3.33±0.47*</td>
</tr>
<tr>
<td>F</td>
<td>2.0 mL juice, Ulcer Induced</td>
<td>1.41±0.11</td>
<td>4.08±0.51*</td>
<td>5.00±0.50*</td>
<td>3.58±0.20*</td>
</tr>
</tbody>
</table>

*Shows significance at p<0.05 when groups were compared with animals in group A. †Shows significance at p<0.05 when groups were compared with animals in group B.

**Malondialdehyde determination:** Lipid peroxidation of gastric mucosa was determined spectrophotometrically at 533 nm and MDA concentration was quantified by using the molar extinction coefficient, 1.56×105 mol L⁻¹ cm⁻¹ (Buege and Aust, 1978; Nair et al., 2008).

**Statistical analysis:** All values presented in tables are expressed as Mean±SEM. The appropriate comparisons between groups were made using students‘ t-test. The differences between the groups were taken to be significant at p<0.05.

**RESULTS**

From the study, groups C, D, E and F were compared with groups A and B independently. Table 1 showed the criteria that were used to score ulcer in this study as used by Rüfiz-Zaman et al. (2004) and Tanaka et al. (1993). From the experimental results showed in Fig. 1, Ulcer indices scored in the various rats groups were statistically compared. The results showed that ulcer inductions in the various groups of rats were significantly increased in group C (15.75±0.50), D (24.92±0.82), E (12.17±0.59) and F (11.08±0.83) when the groups of rats, not given pineapple juice (groups A (0.00) and B (10.25±0.50)) were compared with the groups of rats that were given pineapple juice. But the highest value of significance was noticed in group D rats where 1 mL of pineapple juice was administered.

This is in agreement with the observed trends in ulcer model with respect to total protein contents in groups B (1.40±0.78), C (1.65±0.13), D (1.4±0.61), E (1.41±0.11) and F (1.62±0.05) when compared with group A (2.38±0.18). The results showed reductions that are not statistically significant. Similarly animals in groups C, D, E and F were compared with group B animals, the result showed a slight increase not higher than group A. But in all, no statistical significance was recorded. Other biomarkers used to justify the recorded ulcer indices were catalase, superoxide dismutase and Malondialdehyde. They were all shown in Table 2.

Catalase activities in the groups of the rats, showed a general reduction when group B (2.45±0.49), C (5.40±0.67), D (3.21±0.24), E (4.08±0.51) and F (3.69±0.50) were compared with group A (5.49±0.14). Significant reductions were recorded in group B, D, E and F. Comparison of groups C, D, E and F with group B showed the activity of Catalase to increase with significant increase in group C and E.

Results from superoxide dismutase activity in the rats were compared. Groups B (5.80±0.11), C (6.37±0.84), D (4.31±0.25), E (5.00±0.50) and F (4.34±0.59) were compared with group A (2.80±0.13). The observed result showed significant increases. In the other way, group C, D, E and F were compared with group B. Recorded observations were reductions in activities with groups D, E and F with significance in groups D and F. The increase observed in group C was not significant.

Comparisons of the lipid peroxidation of the gastric mucosa in the rats were also shown in Table 2. Groups B (3.03±0.36), C (5.10±0.21), D (4.48±0.46), E (3.50±0.20) and F (3.33±0.47) were compared with group A (2.15±0.28). General increases were recorded with a significant increase in group B. Similarly group B was compared with groups C, D, E and F while the recorded observation were increases in all the groups (groups C, D, E and F), which were only significant in groups C and D.
DISCUSSION

Despite the fact that fruits contained useful food components, minerals and vitamins (Okwu and Emenike, 2006), the results from this study clearly showed that pineapple juice has a tendency to predispose one to gastric ulcer. Analysis of the ulcer indices revealed that the various volumes of pineapple juice promoted gastric ulceration because of the significant increases indicated when they were compared as shown in Fig. 1. This is consistent with other results of earlier researchers in this field who had implicated some useful dietary food substances in the etiology of gastric ulcer (Nash et al., 1994; Basil and Howard, 1995; Ibu et al., 1986; Olaleye et al., 2006; Ibironke et al., 1997).

A reduction in total protein content, confirmed further that despite the wide medicinal usefulness of pineapple juice, it does not seem to have sufficient capability to protect the gastric epithelia layer from being eroded away. Pineapple juice has been reported to be effective in dissolving mucus and aiding recovery from the dread disease in tuberculosis patient (Taussig and Nieper, 1979; Anandam et al., 1999; Olaleye et al., 2006), similarly the juice has been reported to be acidic (Samson, 1986; Adhikary et al., 1987). In view of this, we can propose that pineapple juice may also have some interaction with the mucus content of the stomach dissolving the mucus thereby exposing the stomach to attack.

Comparison of catalase activities in group A with other groups B, C, D, E and F showed that pineapple juice can promote gastric ulceration as indicated by a reduction in activity. When control animals in groups B (2.45±0.49) were compared with the animals in groups C, D, E and F, increase in activity was noticed, but this increase did not exceed the value obtained for the control group A (5.49±0.67) and became significant in groups C (5.40±0.14) and E (3.69±0.32). These combined effects suggested that pineapple juice protective role under this condition may not be sufficient enough to prevent gastric ulceration.

When superoxide dismutase activities were compared as shown in Table 2, it revealed increased activities that were significant when group A animals were compared with animals in groups B, C, D, E and F. But the comparison of groups C, D, E and F with group B brought similar result which is consistent in a typical gastric ulcer model with antioxidant enzyme (superoxide dismutase). There was reduction in activity in all the groups except group C in which 0.5 ml of pineapple juice was administered.

Observation recorded that lipid peroxidation with malondialdeyde showed increases when groups A and B were compared with other groups (i.e., group A compared with B, C, D, E and F; group B compared with groups C, D, E and F). These observations clearly showed that gastric mucosal damage may be by hemorrhagic lesions through oxidative damage of the mucosa by the increased lipid peroxidation, decreased level of antioxidant defensive enzymes and decrease in protein content (Bandyopadhyay et al., 1999; Priya et al., 2012).

Lipid peroxidation leads to loss of membrane fluidity, ion transport and membrane integrity of the surface epithelia cells which lead to gastric lesions. The lipid content also determines the degree of resistance of mucin to peptic degradation and thus contributes significantly to mucus viscosity hydrophobicity and impedance to hydrogen ion diffusion (Bilski et al., 1987; Olaleye and Fakorede, 2006).

Characteristics model of gastric ulceration has shown reduction in total protein, increase in anti-oxidant activities and reduction in lipid per-oxidation. But results from this experimental study suggested that Pineapple juice consumption may predispose the user to gastric ulceration because it has the tendency to reduce total protein content and increases anti-oxidant enzymes activities. Although, effectiveness of the lipid per-oxidation can not be ascertained because two out of the three conditions studied clearly showed that Pineapple juice consumption can promote gastric ulcer and similarly pineapple juice was found to be effective in dissolving mucus (Taussig and Nieper, 1979; Anandam et al., 1999). Therefore, one should consume the juice with caution.

CONCLUSION

In conclusion, the results of this study clearly showed that despite many medicinal values and nutritional benefits that one can derive from eating pineapple and its juices, care must be taken because it can predispose one to gastric ulceration. However studies are going on to investigate other interactions that pineapple juice may have with our body.

REFERENCES


