Histologic Effects of Carbonated Drinks on Rat Kidney

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Abstract

Background: To study the histologic effects of carbonated drinks on rat kidneys

Methods: In this experimental animal study histological changes on rat kidney caused by chronic consumption of carbonated drinks were observed. Thirty adult Wistar rats with average weight of 200g were randomly assigned into three groups, ten per group. Group A and B served as treatment groups while group C served as the control. The rats in group A were administered a randomly selected carbonated drink ad libitum for thirty days. Rats in group B were given ad libitum access to carbonated drink for sixty days and animals in group C were given water. The rats in group A and B were sacrificed in CO₂ and Chloroform chamber on day 30 and 60 respectively. The abdominal region was quickly opened and the kidney dissected out and fixed in 10% formal saline. Deparaffinized sections were stained routinely with haematoxylin and eosin (H & E).

Results: H and E stain study revealed glomeruli congestion and inflammatory cell infiltrate in group A specimens. The specimens of group B showed glomeruli congestion along with tubular necrosis and nuclear cell disturbance.

Conclusion: Long term consumption of carbonated drinks adversely affects histology of kidney, and the deterioration of cells increases with increase in the consumption of beverage.

Key Words: Carbonated drink, Wistar rats, kidney, histology.

Introduction

Carbonated drinks are the second most consumed beverages in the world. These beverages are already notorious for their adverse effects on teeth, bone, liver and heart vessels. Soft drinks are carbonated drinks that are non-alcoholic. Carbonated soft drinks are also referred to as soda pop, soda or tonic. Carbonated drink first appeared in the market in the 17th century as a mixture of lemon juice and water, sweetened with honey. The term “soft drink” was originated to distinguish the flavored drinks from alcoholic liquor, or spirit. Coca-Cola, the first cola drink was invented in Atlanta by John Pemberton in 1886. Today, products of the Coca-Cola Company are consumed at the rate of more than one billion drinks per day. For more than a century, carbonated drink industry has been under political issues. “Pure food and drug act” was passed in 1906. A series of legislative acts were introduced in the congress to monitor and take actions against carbonated drinks and their ingredients which were proven harmful from time to time. There have been a number of serious health issues associated with regular consumption of soft drinks. According to a peer-reviewed study, 25 separate harmful effects have been reported due to consumption of carbonated soft drinks. Carbonated drinks are considered to be associated with the development of cardiovascular disease, diabetes mellitus, dental/bone problems and obesity, all of which are strongly liked with kidney health. There is also the associated risk of formation of kidney stones.

Materials and Methods

An experimental animal study was conducted at the Post Graduate Medical Institute, Lahore to observe histological changes on rat kidney caused by chronic consumption of carbonated drinks. All animals used in this study were handled with the international, natural and institutional guidelines for the care and use of laboratory animals in biomedical research as promulgated by the National Research Council. Thirty adult Wistar rats of both sexes with average weight of 200g were randomly assigned into three groups: A, B and C of ten per group (Table 1). Group A and B served as treatment groups while group C served as the control. The rats in group A were administered a randomly selected carbonated drink ad libitum for thirty days. Rats in group B were given ad libitum access to carbonated drink for sixty days and animals in group C were given water. The rats in group A and B were sacrificed in CO₂ and Chloroform chamber on day 30 and 60 respectively. The abdominal region was quickly opened and the kidney dissected out and fixed in 10% formal saline for routine histological techniques. The tissues were dehydrated in an ascending grade of alcohol (ethanol), cleared in xylene and embedded in paraffin wax. Consecutive sections...
of 5 microns thick were obtained using a rotary microtome. The deparaffinized sections were stained routinely with haematoxylin and eosin (H & E). Photomicrographs of the results were obtained using research photographic microscope in the Department of Anatomy, Post Graduate Medical Institute Lahore. Data were analyzed by using two-paired student's t-test for quantitative differences between experimental groups and control group at the 5% level of significance. A p-value of equal to or less than 0.05 was considered statistically significant.

### Table 1: Detail of animal groups

<table>
<thead>
<tr>
<th>Groups</th>
<th>Number of animals</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>10</td>
<td>Experimental group (30 day treatment)</td>
</tr>
<tr>
<td>B</td>
<td>10</td>
<td>Experimental group (60 day treatment)</td>
</tr>
<tr>
<td>C</td>
<td>10</td>
<td>Control group</td>
</tr>
</tbody>
</table>

### Results

The parameters which were evaluated were glomerular congestion, tubular necrosis and inflammatory cell infiltration. The microanatomy of the kidney in the control group (C) showed normal histological features (Figure 1; Table 2). The section indicated a normal cortical parenchyma and stroma, while the renal corpuscles appeared as dense rounded structures with the glomerulus surrounded by a narrow Bowman's spaces.

### Table 2: Comparison of Number of inflammatory cells and Number of congested glomeruli between groups on Day 30

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Group A</th>
<th>Group C</th>
<th>Number of Animals</th>
<th>p-Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of inflammatory cells</td>
<td>175.67±14.71</td>
<td>56.67±4.07</td>
<td>10</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Number of glomeruli congestion</td>
<td>79.37±2.08</td>
<td>20.42±3.09</td>
<td>10</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

Experimental group A and B showed distorted appearance of glomerular apparatus and tubular damage and necrosis along with inflammatory cell infiltrate. In group A, there was congestion of glomeruli and a perivascular infiltrate was seen of inflammatory cells. All the specimen showed signs of glomerular congestion and inflammatory cell infiltrate (Figure 2; Table 2). In group B however, the proximal convoluted tubule, distal convoluted tubule and collecting tubules showed areas of necrosis and collecting tubules showed areas of necrosis and

### Table 3: Comparison of Number of inflammatory cells and Number of congested glomeruli between groups on Day 60

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Group B</th>
<th>Group C</th>
<th>Number of Animals</th>
<th>p-Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of inflammatory cells</td>
<td>203.63±12.21</td>
<td>56.67±4.07</td>
<td>10</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Number of glomeruli congestion</td>
<td>113.47±8.28</td>
<td>20.42±3.09</td>
<td>10</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

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Figure 1: Normal histology of rat kidney of group C

Figure 2: Histology of rat kidney of group A at day 30. Congestion of glomeruli is visible (arrow)

Figure 3: Histology of rat kidney of group B at day 60. Tubular necrosis is visible (arrows) distorted architecture (Figure 3). The cell lining of the tubules was also disturbed as seen by irregularity of
nuclei and cell shape. Out of the total specimen slides of experimental group 80% showed compromised kidney histology.

Discussion
This study revealed the histological effects of carbonated drinks on kidney of adult Wistar rats. In group A, carbonated drink caused glomerular congestion and inflammatory cell infiltration. The results obtained in this experiment are in consonance with the work carried out by Enaibe et al. They reported that administration of camphor resulted in glomerulonephritis and mild edema, glomerular lobulation and congestion of blood cell in the kidney of rabbit. A study by Adjene also supported the current results. The consumption of carbonated drink for 30 days caused varying degree of distortion and disruption of the cytoarchitecture of the renal cortex, diffuse glomerulonephritis with some congestion.

In group B, carbonated drink caused necrosis of tubular cells and cellular distortion of kidney cells. According to a study, when damiana(Tunera diffusa) was administered to adult Wistar rats, it caused reduced size and number of renal corpuscles, distortion of renal structures and some degree of necrosis in kidney histology. Hence the results of this study were in concordance with the previous work.

The findings implicated that carbonated beverages were capable of precipitating kidney disease most probably by causing congestion and tubular necrosis of kidney. According to a study accidental or pathologic cell death was regarded necrotic, which could result from an extrinsic insult to cells as a thermal, osmotic, toxic or traumatic effect. The process of cellular necrosis involved disruption of membranes, as well as functional and structural integrity. In this experiment, the carbonated drinks may have acted as toxin to the cells of the kidney resulting in the distortion and disruption, congestion and glomerulonephritis. Alteration in histology of kidney would definitely result in compromised kidney function.

This study has not looked into the mechanism of renal damage. The report is limited simply to a general statement about the overall architecture of the tissue. The hypothesis of possible functional changes that may be detrimental to the health status of the animals would require further studies, including quantification of the structural changes as well as urine and microscopic examinations.

Conclusion
1. Chronic consumption of carbonated adversely affects the anatomy of kidney. It could cause glomerular congestion, edema in tubules and tubular necrosis.
2. Work is required to assess the reversibility of these changes induced by carbonated drink.

References