THE EFFECT OF SPICE PRINCIPLES ON BODY COMPOSITION AND LIPOGENESIS IN RATS

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ABSTRACT
Inclusion of a spice like red pepper fruit and kokum powder in the diet led to a lowering of total lipids, particularly triglycerides in the liver. The total body fat was lowered in animals fed red pepper or capsaicin but not in animals fed kokum powder which had less effect on the total animal body weight content. Hyperlipogenesis and hypertriglyceridemia caused by fructose feeding were significantly decreased in capsaicin-fed animals. Lipogenesis was decreased as reflected by the reduced activities of the key lipogenic enzymes observed in albino rats.

KEYWORDS: Red pepper; Kokum powder; Lipogenesis.

INTRODUCTION
Earlier studies showed that red pepper or its active principle ‘capsaicin’ partly counteracted the accumulation of fat in the liver of rats fed high-fat or choline-deficient diets \cite{18} and in methionine or carbon tetrachloride-induced fatty livers. \cite{23} The effect of capsaicin may be due to its reduction of hepatic lipogenesis, enhanced transport of lipids to serum or increased oxidation of lipids in the tissues. This communication describes findings about the influence of red pepper and kokum (\textit{Garcinia indica}) powder which contains garcinol on total body fat and lipogenic enzyme activities in the liver of albino rats.

MATERIALS AND METHODS
Materials
Red pepper (\textit{Capsicum frutescens}) was obtained locally and ground to 30 mesh size. Kokum powder was obtained by grinding the dried rinds of \textit{Garcinia indica} to the same mesh size as red pepper. Capsaicin in red pepper and Garcinol content in kokum powder were determined...
according to \cite{8, 23} to be 0.3 and 0.005\%, respectively.

**Animals**

Male albino rats of Wistar strain weighing about 250g were used in all the experiments. The basal diet fed to rats consisted of 18\% protein (casein), 10\% sucrose, 10\% fat (groundnut oil), 59\% corn starch, 2\% salt mixture \cite{11} and 1\% vitaminised starch \cite{3} In addition, rats daily received adequate quantities of vitamins A, D and E. Experimental diets were prepared by incorporating 5\% red pepper, 5\% kokum powder or 15 mg\% capsaicin in the basal diet in place of corn starch. At the end of the feeding period (5 weeks), the rats were sacrificed and various tissues collected for analysis.

**Body Composition**

For determining body composition the weighed carcass (free from blood, liver and the intestinal contents) was cut into small pieces and dried at 100\°C until a constant weight was achieved. The difference in weight denoted the water content. The dried carcass was used for the estimation of fat after extraction by gravimetry, \cite{6} protein by micro-Kjeldahl (nitrogen content × 6.25) and ash by the A.O.A.C procedure (A.O.A.C., 1975).

**Fructose Induced Hypertriglyceridemia**

Two groups of animals were fed either the basal or experimental diet containing 5\% capsaicin or 5\% kokum powder for 5 weeks. At the end of the feeding period, in addition to the diet, instead of drinking water both groups of rats received 10\% fructose solution for a period of 7 days.

**Lipid Analysis**

Total lipids in liver and serum were extracted and purified according to \cite{6} and estimated by Gravimetry. Cholesterol, phospholipids and triglycerides were determined using methods described by, \cite{19, 5, 15} respectively.

**Enzyme Assays**

Rats were killed by decapitation and the livers were removed quickly and washed with cold 0.15 KCl. 1g of liver tissue was weighed, homogenised in cold 0.15 KCl and the homogenate was centrifuged at 1000g at 0\°C for 15min. The supernatant was re-centrifuged 100000g for 1h in a Beckman ultracentrifuge and the resulting supernatant was used for enzyme assays. Glucose-6-phosphate dehydrogenase and 6 phosphogluconate dehydrogenase were assayed by
the method of [7] with modifications as described by. [13] Malic enzyme and citrate cleavage enzyme were assayed as described by [16, 4] with modifications as suggested by. [22] The protein content of homogenates was determined by the method of. [14] The enzyme activities were expressed as units/mg protein and one unit is defined as the amount of enzyme which catalyzes the utilization of one nmol of substrate per min.

Statistical Analysis
Data were analyzed by Student ‘t’ test and levels of significance denoted in tables by the letters a-p<0.05, b-p<0.02, c-p<0.01, d-p<0.002 and e-p<0.001. [20]

RESULTS

Body Composition
The effect of spice principle from red pepper and kokum powder in the diet significantly reduced the weight of liver compared to controls (Table 1).

Lipid Analysis
Total lipid levels were reduced only in rats receiving red pepper or capsaicin and kokum containing diets. The triglyceride component of the total lipids was lowered significantly, without any change in the phospholipid or cholesterol content. Supplementation of the kokum powder containing diet with Garcinol brought about similar changes (Table 1).

The carcass composition of rats fed different experimental diets is presented in Table 2. A significant decrease in total body lipid with a concomitant increase in moisture was found in rats fed pepper. Kokum powder in contrast with red pepper, did not alter the body fat content. There were no differences observed in the protein and ash content among rats fed the different diets.

Fructose Induced Hypotriglyceridemia
Triglycerides in the liver of rats receiving diet containing a combination of 15 mg% capsaicin and 5% kokum powder, were significantly reduced. Total lipids, phospholipids and cholesterol levels were not altered (Table 3). A significant reduction was also observed in serum triglyceride levels of rats fed capsaicin containing diet (Table 3).

Lipogenic Enzyme Activities in Liver
It is seen (Table 4) that all the four key lipogenic enzyme activities were significantly decreased though to different degrees. Of the three NADPH producing enzyme reactions,
glucose-6-phosphate dehydrogenase showed the maximum decrease. Feeding of capsaicin caused a significant decrease in the activity of citrate cleavage enzyme also.

**Table 1: Effect of red pepper, capsaicin and kokum powder individually and in combination on body weight and liver lipids.**

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Additional to basal diet</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Nil</td>
</tr>
<tr>
<td>Initial body wt(g)</td>
<td>248±3.5</td>
</tr>
<tr>
<td>Final body wt(g)</td>
<td>320±10.5</td>
</tr>
<tr>
<td>Wt of liver (g)</td>
<td>10.5±0.5</td>
</tr>
<tr>
<td>Liver lipids (mg/g)</td>
<td>Total lipids</td>
</tr>
<tr>
<td></td>
<td>Triglycerides</td>
</tr>
<tr>
<td></td>
<td>Phospholipids</td>
</tr>
<tr>
<td></td>
<td>Cholesterol</td>
</tr>
</tbody>
</table>

Values are mean ± SEM for 6 rats fed for 8 weeks.

**Table 2: Effect of red pepper, capsaicin and paprika on carcass composition in adult rats**

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Additional to basal diet</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Nil</td>
</tr>
<tr>
<td>Corrected*body wt(g)</td>
<td>270±8.5</td>
</tr>
<tr>
<td>Carcass Composition % of Corrected*body wt (g)</td>
<td></td>
</tr>
<tr>
<td>Water</td>
<td>61.2±1.8</td>
</tr>
<tr>
<td>Protein</td>
<td>18.1±0.8</td>
</tr>
<tr>
<td>Ash</td>
<td>5.1±0.1</td>
</tr>
<tr>
<td>Lipid % of Corrected*body wt(g)</td>
<td>30.8±5.4</td>
</tr>
<tr>
<td>% of dry carcass</td>
<td>58.1±4.0</td>
</tr>
</tbody>
</table>

Values are mean ± SEM for 6 rats fed for 8 weeks

*Corrected body weight represents body weight minus liver blood and intestinal contents.

Carcass composition on fat-free basis: water: 66—70%; protein: 17-20%; ash: 9.3-10%.

**Table 3: Influence of kokum extract on lipid levels in liver and serum in fructose induced hypertriglyceridemia.**

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Additional to basal diet</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Nil</td>
</tr>
<tr>
<td>Treatment</td>
<td>Fructose feeding</td>
</tr>
<tr>
<td>Liver lipids(mg/g)</td>
<td>55.2±1.8</td>
</tr>
</tbody>
</table>

*Corrections and edits have been made to ensure the text is clear and consistent with the guidelines.*
Values are mean ± SEM 8 rats fed for 8 weeks

* Rats were given drinking water containing 10% fructose for 7 days before they were sacrificed.

Table 4: Effect of spice principle on some liver lipogenic enzymes.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Additional to basal diet</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Nil</td>
</tr>
<tr>
<td>Glu-6-phosphate dehydrogenase</td>
<td>67.6±4.4(6)</td>
</tr>
<tr>
<td>6-phosphogluconate dehydrogenase</td>
<td>43.2±2.6(7)</td>
</tr>
<tr>
<td>Malic enzyme</td>
<td>82.8±6.2(9)</td>
</tr>
<tr>
<td>Citrate cleavage enzyme</td>
<td>22.1±0.8(6)</td>
</tr>
</tbody>
</table>

Values are mean ± SEM with number of rats in parenthesis in each group.

Duration of feeding - 8 weeks.

* Enzyme activity is expressed as units.

One unit of enzyme = 1nmol of substrate utilized/minute/mg protein.

DISCUSSION

The present investigation has shown that both red pepper/capsaicin and kokum powder feeding Analysis of the body composition of adult rats fed either red pepper or capsaicin and kokum powder in the diet revealed a significant reduction in the total body lipid levels with no change in total body protein. The reduced level of fat in the body is significant both when the fat is expressed as per cent of corrected body weight or as per cent of dry carcass (Table 2). In contrast to the reduction of fat, the change in body water was not real. Thus, although there was a significant increase in water content in the bodies of animals fed red pepper or capsaicin and kokum powder, this disappeared when the water content of the carcass was expressed on fat-free basis. These results substantiate the findings of the effect of capsaicin on fatty acid synthesis as indicated by the enzyme activities.

REFERENCES