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Influence of red pepper and capsaicin on body composition and lipogenesis in rats

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Abstract. Inclusion of red pepper or its active principle 'capsaicin' 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 paprika powder which had negligible capsaicin content. Hyperlipogensis and hypertriglyceridemia caused by fructose feeding were significantly were decreased in capsaicin-fed animals. Activities of the key lipogenic enzymes were reduced as reflected by decreased lipogenesis.

Keywords. Red pepper; capsaicin; 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or choline-deficient diets (Sambaiah *et al.,* 1978) and in ethionine or carbon tetrachloride induced fatty livers (unpublished work).

The effect of capsaicin may be due to (a) its reduction of hepatic lipogenesis; (b) enhanced transport of lipids to serum or (c) increased oxidation of lipids in the tissues. This communication describes findings about the influence of red pepper and capsaicin on total body fat and lipogenic enzyme activities in the liver of rats.

Materials and methods

Red pepper (*Capsicum frutescens*) was obtained locally and ground to 30 mesh size. Paprika (*Capsicum annuum*) powder was obtained as a gift from Lalocsa Paprika Co., Hungary. Capsaicin content in these samples was determined (Govindarajan and Ananthakrishna, 1974) to be 0.3 and 0.005% respectively. Male rats of Wistar strain weighing about 200 g 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 (Hubbel *et al.,* 1937) and 1% vitaminised starch (Chapman *et al.,* 1959). In addition, rats daily received adequate quantities of vitamins A, D and E. Experimental diets were prepared by incorporating 5% red pepper, 5% paprika powder or 15 mg% capsaicin (Fluka AG, Switzerland) in the basal diet in place of corn starch. At the end of the feeding period, 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 to constant weight at 100°C. The difference in weight denoted the water content. The dried carcass was used for the estimation of fat after extraction by gravimetry (Folch *et al.,* 1957), protein by micro-Kjeldahl (nitrogen content × 6.25) and ash by the A.O.A.C procedure (A.O.A.C., 1975).

Fructose induced hypertringlyceridemia

Two groups of animals were fed either the basal or experimental diet containing 15 mg% capsaicin, for 5 weeks. At the end of the feeding period, in addition to the diet both groups of rats received 10% fructose solution instead of drinking water for a period of 7 days.

Lipid analysis

Total lipids in liver and serum were extracted and purified according to Folch *et al.* (1957) and estimated by gravimetry. Cholesterol, phospholipids and triglycerides were determined using methods described by Searcy and Bergquist (1960), Marinetti (1962) and Fletcher (1968) respectively.

Enzyme assays

Rats were killed by decapitation and the livers were removed quickly and washed with cold 0.15 M KCl. About 1 g of liver tissue was homogenised in cold 0.15 M KCl and the homogenate was centrifuged at 1000 *g* at 0-4°C for 15 min in a Sorval RC 2B centrifuge. The supernatant was recentrifuged 100000 *g* for 1 h in a Beckman L2-65B ultracentrifuge and the resulting supernatant was used for enzyme assays. Glucose-6-phosphate dehydrogenase (EC 1.1.1.49) and 6-phosphogluconate dehydrogenase (EC 1.1.1.44) were assayed by the method of Glock and McLean (1953) with modifications as described by Leveille (1972). Malic enzyme (EC 1.1.1.40) and citrate cleavage enzyme (EC 4.1.3.8) were assayed as described by Ochoa (1955), Cottom and Srere (1964) with modifications as suggested by Yeh *et al.* (1970). The protein content of homogenates was determined by the method of Lowry *et al.* (1953). The enzyme activities were expressed as units/mg protein and a 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-$p<0.001$ (Snedecor and Cochran, 1967).

Results

Influence of red pepper/capsaicin on body composition

The inclusion of capsaicin in the diet significantly reduced the weight of liver compared to me controls, whereas red pepper and paprika had no such effect (table 1). Total lipid levels were reduced only in rats receiving red pepper or

capsaicin containing diets. The triglyceride component of the total lipids was lowered significantly, without any change in the phospholipid or cholesterol content. Supplementation of the paprika containing diet with capsaicin at 15 mg% brought about similar changes (table 1).

Table 1. Effect of red pepper, capsaicin and paprika on liver lipids.

Parameters	Additions to basal diet				
	Nil	5% red pepper	15 mg% capsaicin	5% paprika	5% paprika ± 15 mg% capsaicin
Initial body weight (g)	208±3.34	207±2.78	208±3.40	208±3.25	208±2.98
Final body weight (g)	282±10.6	285±9.9	268±6.9	281±6.3	270±5.8
Weight of liver (g)	9.5±0.5	8.9±0.2	8.2[a]±0.2	8.6±0.3	8.0[a]±0.2
Liver lipi ds (mg/g)					
Total lipids	51.7±2.1	42.7[c]±2.1	42.7[a]±1.6	46.2±3.3	43.4[a]±0.8
Triglycerides	9.1±0.8	5.5[c]±0.8	5.6[a]±1.1	6.8±1.0	3.19[c]±0.3
Pospholipids	26.7±0.8	27.5±1.1	27.6±1.3	26.4±0.5	27.2±0.6
Cholesterol	3.7±0.1	4.0±0.1	3.5±0.1	4.1±0.1	4.1±0.2

Values are mean ± SEM for 5 rats fed for 8 weeks

The carcass composition of rats feed 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 or capsaicin. Paprika 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 15 mg% capsaicin, 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).

Effect of capsaicin on some 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 2. Effect of red pepper, capsaicin and paprika on carcass composition in adult rats.

Parameters	Additions to basal diet				
	Nil	5% red pepper	15 mg% capsaicin	5% paprika	5% paprika ± 15 mg% capsaicin
Corrected* body weight (g)	253±8.8	256±9.1	241±5.9	255±5.9	242±5.8
Carcass composition					
% of corrected body weight					
Water	57.3±1.7	64.9[a]±1.3	65.4[a]±1.8	60.6±0.8	63.8±1.7
Protein	14.3±0.6	14.3±0.4	14.1±0.3	14.4±0.4	14.4±0.5
Ash	3.1±0.1	3.3±0.1	3.1±0.1	3.1±0.1	3.1±0.2
Lipid					
% of corrected body weight	25.3±2.4	17.5[a]±1.5	17.4[a]±2.1	21.9±0.7	18.7[a]±1.4
% of dry carcass	52.1±3.0	41.9[a]±2.7	41.2[a]±2.5	44.7±2.7	40.3[a]±2.5

Values are mean ± S EM for 5 rats fed for 8 weeks

* Corrected body weight represents body weight minus liver blood and intestinal contents. Carcas composition on fat-free basis: water, 66—70%; protein, 17-20%, ash, 9.3-10%.

Table 3. Influence of capsaicin on lipid levels in liver and serum in fructose induced hypertriglyceridemia.

	Additions to basal diet		
	Nil	Nil	15 mg% capsaicin
Treatment	Nil	Fructose* feeding	Fructose feeding
Liver lipids (mg/g)			
Total	46.3±1.8	51.4±2.5	48.1±2.1
Triglycerides	10.6±0.7	15.3±1.2	6.2±1.5[c]
Phospholipids	23.8±3.4	19.6±0.9	19.7±0.9
Cholesterol	3.6±0.1	7.4±0.4	8.4±0.6
Serum triglycerides	99.5±19.2	190.5±24.6	121.7±16.2[a]

Values are mean ± SEM 8 rats fed for 6 weeks

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

Table 4. Effect of capsaicin on some liver lipogenic enzymes[ø]

	Addition to basal diet	
	Nil	15 mg% capsaicin
Glucose-6-phosphate dehydrogenase	57.8 ± 3.3 (6)	31.4 ± 3.9[c] (6)
6-Phosphogluconate dehydrogenase	33.6 ± 1.6 (7)	26.5 ± 2.8[a] (7)
Malic enzyme	72.6 ± 4.4 (9)	45.8 ± 7.1[a] (6)
Citrate cleavage enzyme	16.1 ± 0.5 (6)	11.7 ± 0.9[d] (6)

Values are mean ± SEM with number of rats in parenthesis in each group. Period of feeding,6 weeks.

* Enzyme activity is expressed as units where one unit of enzyme is one nanomol of substrate utilized/minute/mg protein.

Discussion

The present investigations have shown that both red pepper and capsaicin feeding significantly counteract the increase in triglyceride levels in livers of rats fed normal and fructose containing diets. A decrease in serum triglyceride levels is also brought about by capsaicin feeding in fructose fed animals (tables 1 and 3).

Fructose is known to enhance hepatic lipogenesis (Zakim *et al.,*1967; Sullivan *et al.,* 1971) and cause hypertriglyceridemia (Bar-On and Stein, 1968; Hill, 1970). The former condition is likely to be brought about by an increase in lipogenic enzyme activities (Romsos and Leveille, 1974) observed in the livers of rats fed fructose rich diets.

Fatty acid synthesis involves the activities of glucose-6-phosphate dehydrogenase, 6-phosphogluconate dehydrogenase, malic enzyme and citrate cleavage enzyme, to various degrees. A reduction of these enzymes leads to decreased availability of reduced nucleotides as well as extra mitochondrial acetyl Co A both of which are required for lipogenesis. A good correlation between pentose phosphate pathway and the rate of fatty acid synthesis in rat adipose tissue was demonstrated earlier (Kather *et al.,* 1972). Malic enzyme also participates in the generation of extramitochondrial NADPH which supplements the requirement of reduced equivalents to fatty acid synthesis. The citrate cleavage enzyme is generally accepted to be of major importance in supplying the needed acetyl COA for lipogenesis (Greville, 1969). Capsaicin probably lowers lipogenesis as reflected by the reduced activity of the key lipogenic enzymes (table 4).

Analysis of the body composition of adult rats fed either red pepper or capsaicin 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, 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.

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