Eur Food Res Technol (2008) 227:1159–1163
DOI 10.1007/s00217-008-0831-0

ORIGINAL PAPER

Cholesterol lowering activity of mango ginger (*Curcuma amada* Roxb.) in induced hypercholesterolemic rats

M. R. Srinivasan · N. Chandrasekhara · K. Srinivasan

Received: 28 September 2007 / Revised: 15 January 2008 / Accepted: 19 January 2008 / Published online: 7 February 2008
© Springer-Verlag 2008

Abstract Mango ginger (*Curcuma amada* Roxb.) is a spice commonly consumed in Asian countries. Health beneficial hypotriglyceridemic property of mango ginger has been reported earlier. In this investigation, the anti-hypercholesterolemic influence of dietary mango ginger was evaluated in experimental rats. Dietary mango ginger powder (10%) or its equivalent of 10 mg% curcumin-containing portion was fed along with 1% cholesterol supplemented diet in Wistar rats for 5 weeks. The treatment countered the liver and serum total and LDL + VLDL associated cholesterols and increased the HDL associated cholesterol while it had no influence on cholesterol levels in animals, maintained on normal diet. Dietary *C. amada* and its curcumin-free portion were effective in lowering liver cholesterol in animals, maintained on basal diet, while the curcumin-containing component of *C. amada* was ineffective. The biliary secretion of total lipids and bile acids was increased by dietary *C. amada* and both of its components. While biliary cholesterol was increased in animals fed with whole mango ginger, no such increases were noticed in groups fed with either components of mango ginger. Cholesterol absorption in ligated rat intestinal loops was not affected by mango ginger or either of its components. Thus, the present study has evidenced that the spice mango ginger possesses beneficial anti-hypercholesterolemic activity in hypercholesterolemic situation. This information is complementary to the earlier report on the health beneficial hypotriglyceridemic influence of this spice.

Keywords *Curcuma amada* · Serum cholesterol · Liver cholesterol · Bile acids

M. R. Srinivasan · N. Chandrasekhara · K. Srinivasan (✉)
Department of Biochemistry and Nutrition,
Central Food Technological Research Institute,
Mysore 570 020, India
e-mail: ksri.cftri@gmail.com

Introduction

Mango ginger (*Curcuma amada* Roxb.) is a spice having a long history of traditional uses ranging from folk medicine to several culinary preparations. It is mostly used as a basic ingredient in pickles owing to its raw mango-like flavour, and also in making preserves, candy and sauce [1]. The phytochemical, pharmacological and ethnobotanical studies of *C. amada* are reviewed [2]. It possesses several medicinal properties such as stomachic, carminative, laxative, aphrodisiac, antipyretic and is useful in the treatment of biliousness, skin diseases, bronchitis, asthma and inflammation [3]. Its antibacterial, insecticidal, antifungal and antioxidant properties have been investigated. The rhizome is rich in essential oils, and more than 130 chemical constituents with biomedical significance have been isolated from it [2]. The major volatile aroma compounds responsible for the characteristic flavour of *C. amada* have been identified to be *cis*- and *trans*- dihydroocimene, ocimene and myrcene [4].

Among the commonly used spices, turmeric (*Curcuma longa*) or its coloring principle—curcumin, red pepper (*Capsicum annuum*) or its pungent principle—capsaicin, fenugreek (*Trigonella foenum-graecum*) seeds, garlic (*Allium sativum*) and onion are known to have health beneficial hypocholesterolemic property [5]. In an isolated study, ether and ethanolic extracts of rhizomes of mango ginger are evidenced to be hypocholesterolemic in experimentally induced hypercholesterolemic rabbits [6]. An extract of *C. amada* in admixture with a few other spices which is marketed as a drug known as 'Temoe-lawak

Singer' in certain parts of Europe is claimed to be useful as a cholesterol lowering drug [7]. Our earlier studies had shown significant hypotriglyceridemic property of mango ginger in both normal and high sucrose diet induced hyperlipidemic rats [8, 9].

In view of the limited information on the cholesterol lowering potential of *C. amada*, the present study was made to substantiate the health beneficial cholesterol lowering influence of this spice in both normal and hypercholesterolemic rats. Since the hypocholesterolemic potency of the other spice—turmeric (*Curcuma longa*) is attributable to its main ingredient—curcumin [4], the present study also examines both the curcumin-free and the curcumin-containing fractions of C. *amada* for their hypocholesterolemic efficacy.

Materials and methods

Materials

Dry mango ginger powder was a freeze–dried preparation of freshly harvested rhizomes procured from local market cleaned with water, free of mud and mechanically sliced. A lipid extract was prepared from dry mango ginger powder by Soxhlet extraction with *n*-hexane for 24 h and the lipids recovered by evaporating off the solvent in vacuum. This lipid extract was 3% of dry mango ginger powder. The spent residue left over after the extraction of lipids was dried in a vacuum oven (40 °C for 8 h). Quantitation of curcumin in the mango ginger powder and its two fractions was made by rubrocurcumin reaction [10] after separation by thin layer chromatography as described by Suresh et al. [11]. The content of curcumin in the dry mango ginger powder and in the lipid-free residue ranged between 0.1 and 0.125%. The lipid extract represented the curcumin-free component of dry mango ginger powder. The curcumin-containing component of mango ginger was the dried and powdered residue obtained after lipid extraction with hexane. All reagents used in this study were of analytical grade; all solvents were distilled before use.

Animals and diets

Animal experiments were carried out by taking appropriate measures to minimize pain or discomfort in accordance with the guidelines laid down by the NIH, USA, regarding the care and use of animals for experimental procedures and with due clearance from the Institutional Animal Ethics Committee. Female Wistar rats (six per group) weighing 100–105 g and housed in individual stainless steel cages were maintained on various experimental diets ad libitum for 5 weeks. The basal diet consisted of (%): casein, 21; cane sugar, 64; NRC vitamin mixture, 1; Bernhardt-Tommarelli modified mineral mixture, 4, and refined peanut oil, 10. The high cholesterol diet consisted of 1% cholesterol and 0.125% bile salts added at the expense of an equivalent amount of cane sugar in the basal diet. The test materials, mango ginger powder or its curcumin containing portion were incorporated into the basal diet/high cholesterol diet at 10 g/100 g, substituted in place of sucrose on weight/weight basis. The curcumin-free portion of mango ginger was incorporated at levels of 0.3, 0.1 and 0.05 g/100 g as suspension in the oil portion of diet.

Collection of bile

At the end of the feeding trial, animals fed with different test materials along with normal diet were fasted overnight, and anaesthetized by an intra peritoneal injection of ethyl urethane (1.2 g/kg body weight). Laparotomy was performed and the common bile duct was cannulated with PE-10 tubing (Thomas Scientific Co., USA) [12]. Bile duct cannulation was made on the common bile duct about 2 cm from duodenum; and fresh hepatic bile was collected for two and half hours. Bile volume was measured and kept frozen until further analysis.

Analytical methods

Liver and serum lipids were extracted by the method of Folch et al. [13] and that from bile by the procedure of Bligh and Dyer [14]. Cholesterol was determined by the method of Searcy and Bergquist [15]. HDL was separated from LDL and VLDL using the heparin-manganese precipitation according to Warnick and Albers [16]. Biliary total lipids were determined by gravimetry and total bile acids by the enzymatic method of Turley and Dietschy [17].

In situ cholesterol absorption

The effect of test materials on fat absorption from the intestine was evaluated in a separate set of adult female Wistar rats weighing 200–210 g from the stock colony. The animals were fasted overnight and anaesthetized and the intestinal loops were ligated and peanut oil was instilled into the ligated loops of small intestine for 3 h as described before [8]. Whole mango ginger powder or its curcumin-containing portion (1 g) or curcumin-free portion (0.3 g) were instilled into the intestinal loop along with cholesterol as suspension in peanut oil. Cholesterol (2.5%) in peanut oil was control. The cholesterol content was determined in the recovered left over oil in the lumen after 3 h.

Statistical evaluation

Significance of differences between groups were evaluated by using Student's *t* test [18].

Results and discussion

In an earlier study on rabbit, hypocholesterolemic activity was observed with ethereal and alcoholic extracts from *C. amada* while the effect of whole *C. amada* rhizomes is not evaluated [6]. The dose of *C. amada* employed in this study works out to 100–200 g/day. Moreover, these authors have inferred the beneficial hypocholesterolemic activity of *C. amada* based on observations made in three animals per treatment. The lipid profile of liver and other tissues were not examined in this earlier study. In view of these limitations, and in continuation of our studies on beneficial metabolic effects of *C. amada*, the blood and liver cholesterol status was evaluated in both normal diet and high cholesterol diet fed rats. The intake of mango ginger when fed at the level of 10% in the diet in the current study works out to around 0.8–1.0 g/day in the rat. The highest level of curcumin-free portion of mango ginger studied here, namely 0.3 g/100 g diet corresponds to 10 g% of whole mango ginger examined in this study.

Effect of whole mango ginger, curcumin-free and curcumin containing portions of mango ginger on serum and liver cholesterol profile

As seen from Table 1, the cholesterol supplemented diet caused increases in serum and liver total cholesterol by 223 and 127%, respectively, as compared to animals fed basal diet. When 10% *C. amada* powder or its equivalent 10% curcumin-containing portion of *C. amada* was included in the diet, it increases the blood and liver cholesterol was significantly decreased. The increase in serum cholesterol in cholesterol supplemented animals was predominantly in LDL + VLDL associated fraction, thus resulting in a sharp fall in HDL-cholesterol:total cholesterol ratio. Dietary *C. amada* decreased serum total cholesterol by 24%. The anti-hypercholesterolemic influence of dietary *C. amada* was even more pronounced in LDL + VLDL fraction with the decrease being as much as 50%. There was a concomitant increase in serum HDL-cholesterol with a resultant significant increase in HDL-cholesterol:total cholesterol ratio. The curcumin-containing portion of *C. amada* also exerted anti-hypercholesterolemic activity producing a reduction in serum total and LDL-cholesterol by 28 and 41% respectively. The curcumin-containing portion of *C. amada* also increased serum HDL-cholesterol and the ratio of HDL-cholesterol:total cholesterol. The curcumin-free portion of *C. amada* countered the increase of LDL + VLDL associated cholesterol in high cholesterol diet fed animals, only when fed at 0.3% in the diet. The curcumin-free portion of *C. amada* increased serum HDL-cholesterol and hence HDL-cholesterol:total

Table 1 Effect of mango ginger, curcumin-free and curcumin containing portions of mango ginger on serum and liver cholesterol profile

Diet group	Liver (mg/g)	Serum (mg/dl)			
		Total	LDL + VLDL	HDL	HDL/total
HCD + nil	12.4 ± 0.71	271.0 ± 18.6	223.0 ± 19.2	48.0 ± 4.22	0.18 ± 0.02
HCD + 10% whole mango ginger	8.10 ± 0.19*	205.5 ± 14.4*	110.0 ± 3.53*	89.0 ± 7.60*	0.42 ± 0.04*
HCD + 0.05% curcumin-free portion	14.1 ± 1.40	276.0 ± 20.4	215.0 ± 16.8	61.0 ± 7.10	0.22 ± 0.02
HCD + 0.1% curcumin-free portion	14.7 ± 1.02	290.5 ± 20.4	183.0 ± 17.1	107.5 ± 13.2*	0.37 ± 0.03*
HCD + 0.3% curcumin-free portion	13.9 ± 0.48	266.5 ± 11.5	144.0 ± 6.34*	129.0 ± 6.61*	0.46 ± 0.05*
HCD + 10% curcumin containing portion	9.82 ± 0.48*	194.0 ± 14.3*	131.0 ± 10.6*	63.0 ± 4.50*	0.33 ± 0.05*
Basal diet + nil	5.91 ± 0.61	84.0 ± 4.61	59.3 ± 6.58	25.7 ± 3.48	0.31 ± 0.04
Basal diet + 10% whole mango ginger	4.21 ± 0.30*	98.8 ± 6.88	73.5 ± 6.58	25.4 ± 2.91	0.26 ± 0.03
Basal diet + 0.05% curcumin-free portion	3.42 ± 0.29*	75.6 ± 3.38	49.4 ± 2.53	26.4 ± 2.04	0.35 ± 0.03
Basal diet + 0.1% curcumin-free portion	3.58 ± 0.22*	75.0 ± 3.42	49.1 ± 2.58	26.1 ± 4.38	0.35 ± 0.05
Basal diet + 0.3% curcumin-free portion	4.28 ± 0.32*	98.3 ± 10.5	71.2 ± 9.40	27.0 ± 3.47	0.28 ± 0.04
Basal diet + 10% Curcumin containing portion	5.30 ± 0.68	83.0 ± 6.42	48.9 ± 2.50	30.1 ± 4.21	0.39 ± 0.05

HCD High cholesterol diet

Values are mean ± SEM of six rats in each group

* Significantly different ($P < 0.05$)

Table 2 Biliary total lipid, bile acid and cholesterol in rats fed mango ginger, curcumin-free and curcumin containing portions of mango ginger

Diet group	Bile(mg/ml)		
	Total lipid	Total bile acids	Cholesterol
Basal diet + nil	7.30 ± 0.41	4.28 ± 0.26	0.28 ± 0.01
Basal diet + 10% whole mango ginger	10.4 ± 0.38*	7.30 ± 0.16*	0.35 ± 0.02*
Basal diet + 0.3% curcumin-free portion	10.4 ± 0.46*	7.38 ± 0.25*	0.29 ± 0.02
Basal diet + 10% curcumin containing portion	8.58 ± 0.29*	6.02 ± 0.25*	0.27 ± 0.02

Values are mean ± SEM of six rats in each group

* Significantly different ($P < 0.05$)

cholesterol ratio at both 0.1 and 0.3% dietary levels (Table 1). The increase in the ratio of HDL-cholesterol:total cholesterol brought about by *C. amada* and its curcumin-free portion even surpassed this value found in basal diet fed animals. While dietary *C. amada* and its curcumin-free and curcumin-containing fractions exerted beneficial anti-hypercholesterolemic property in cholesterol fed animals, they did not have any influence on serum cholesterol levels in rats maintained on basal diet (Table 1).

The decrease in LDL + VLDL associated cholesterol and increase in the HDL-cholesterol caused by the curcumin-free portion of *C. amada* was almost proportionate to the dose employed. The relatively lower efficacy of curcumin-containing portion is probably due to low curcumin content in mango ginger [8].

Dietary *C. amada* and its curcumin-containing portion were effective in lowering liver cholesterol in animals maintained on high cholesterol diet, while the curcumin-free component of *C. amada* was ineffective (Table 1). Dietary *C. amada* and its curcumin-free portion were effective in lowering liver cholesterol in animals maintained on basal diet, while the curcumin-containing portion of *C. amada* was ineffective. The hepatic cholesterol lowering by this curcumin-free portion of *C. amada* was effective at all the three dietary levels examined viz., 0.05, 0.1 and 0.3%. Liver and serum cholesterol concentrations are reported to be unaffected by dietary curcumin in rats fed normal diet [19].

Biliary total lipid, bile acid and cholesterol in rats fed with whole mango ginger, curcumin-free and curcumin containing portions of mango ginger

Liver cholesterol is the source for biliary lipids. The biliary secretion of total lipids and bile acids was increased by dietary *C. amada* and both of its components (Table 2). While cholesterol was increased in animals fed with whole mango ginger, no such increases were noticed in groups fed with either of the two fractions of mango ginger. Dietary spice compounds-curcumin and capsaicin and spices-fenugreek, ginger, onion, mustard, cumin, coriander, ajowan, fennel and mint have been reported to increase the secretion of biliary total bile acids [20–23]. Increased biliary cholesterol in the group fed with whole mango ginger may probably be due to a higher hepatic conversion of cholesterol to bile acids.

As examined in the present study by the in situ technique, the cholesterol absorption in ligated rat intestinal loops was not affected by mango ginger or either of its components. We have also observed earlier that dietary mango ginger or its two components did not influence fat (peanut oil) absorption in ligated rat intestinal loops [8].

Conclusion

Thus, the present study evidences that the spice mango ginger possesses beneficial anti-hypercholesterolemic activity when fed to hypercholesterolemic rats. While dietary *C. amada* and its curcumin-free and curcumin-containing fractions exerted beneficial anti-hypercholesterolemic property in cholesterol fed animals, they did not have any influence on serum cholesterol levels in rats maintained on basal diet. Although the dietary levels that produced the hypocholesterolemic influence are not normally encountered, the information on the nutraceutical potency of this spice is relevant in the context of a cumulative effect or a possible therapeutic application. This information is complementary to our earlier report on the health beneficial hypotriglyceridemic influence of this spice.

References

1. Shankaracharya NB (1982) Indian Cocoa Arecanut Spices J 5:78–80
2. Jatoi SA, Kikuchi A, Gilani SA, Watanabe KN (2007) Phytother Res 21:507–516
3. Kirtikar KR, Basu BD (1984) Indian medicinal plants, vol 4, 2nd edn. Bishen Singh et al Publishers, Mumbai, pp 2422–2423
4. Rao AS, Rajanikanth B, Seshadri R (1989) J Agric Food Chem 37:740–743

5. Srinivasan K, Sambaiah K, Chandrasekhara N (2004) Food Rev Int 20:187–220
6. Pachauri S, Mukherji SK (1970) J Res Indian Med 5:27–31
7. Beynen AC (1987) Artery 14:190–197
8. Srinivasan MR, Chandrasekhara N (1992) J Food Sci Technol 29:130–132
9. Srinivasan MR, Chandrasekhara N (1993) Nutr Res 13:1183–1190
10. Ravindranath V, Satyanarayana MN, Rao MVL (1981) Indian J Chem 20:907–909
11. Suresh D, Manjunatha H, Srinivasan K (2007) J Food Comp Anal 20:346–351
12. Srinivasan K, Levine WG, Bhargava MM (1991) Xenobiotica 21:961–969
13. Folch J, Lees M, Sloane-Stanley GH (1957) J Biol Chem 226:497–509
14. Bligh EC, Dyer WJ (1958) Can .J Biochem Physiol 3:911–917
15. Searcy RL, Bergquist LM (1960) Clin Chim Acta 5:192–197
16. Warnick GR, Albers JJ (1978) J Lipid Res 19:65–76
17. Turley SD, Dietschy JM (1978) J Lipid Res 19:924–928
18. Snedecor GW, Cochran WG (1976) Statistical methods. 6th edn., Iowa State University Press, Ames, p 298
19. Subba Rao D, Chandrasekhara N, Satyanarayana MN, Srinivasan M (1970) J Nutr 100:1307–1315
20. Bhat BG, Srinivasan MR, Chandrasekhara N (1984) J Food Sci Technol 21:225–227
21. Bhat BG, Sambaiah K, Chandrasekhara N (1985) Nutr Rep Int 32:1145–1151
22. Sambaiah K, Srinivasan K (1991) J Food Sci Technol 28:35–38
23. Platel K, Srinivasan K (2000) Nutr Res 20:1493–1503