ORIGINAL ARTICLE



Beneficial dietary effect of turmeric and sulphur on weight gain, fat deposition and lipid profile of serum and liver in rats

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Abstract This study was designed to investigate the effects of turmeric powder and processed sulphur on the weight gain, body fat deposition and lipid profile of serum and liver in Wistar rats. Twenty-five rats of 6 weeks old were divided into five groups with 5 rats in each group. Each group was fed different diets as follows I. common diet (CON); II. high fat diet (HFD); III. 10% turmeric powder with HFD (T); IV. 10% turmeric powder and 0.19% processed sulphur with HFD (TS); and V. 0.38% processed sulphur with HFD (S). The experimental feeding was continued for 6 weeks. The body weight gain and feed efficiency ratio (FER) in the T and TS group rats were significantly (p<0.05) lower than that of the HFD group rats. The retroperitoneal fat weights in the rats belong to T, TS and S groups were lower than that of the HFD group rats and the TS group had significant (p<0.05) reduction in retroperitoneal fat compared to the HFD group rats. The epididymal fat weights in rats of the T, TS and S groups also showed a lowering tendency compared to that of the HFD group rats. The hepatic total lipid levels in the T and

of the HFD group rats. The hepatic triglyceride level in the rats of TS group was significantly (p<0.05) lower than that of the HFD group rats. The serum total cholesterol, high-density lipoprotein (HDL) and low density lipoprotein (LDL) associated cholesterol contents in rats of the T and TS group were significantly (p<0.05) higher than that of the HFD group rats, however, there was no significant difference in serum triglyceride. The results suggest that turmeric powder along with sulphur can reduce the weight gain, body fat deposition and improve serum and liver lipid profile in rats fed with a high fat diet.

TS group rats were significantly (p < 0.05) lower than that

Keywords Turmeric powder · Processed sulphur · Weight gain · Fat deposition · Lipid profile

Introduction

The increase intake of meat and high-calorie food has become a serious concern to public health (Shin et al. 2006; Lee et al. 1999). The national health statistics in Korea (Statistics Korea 2008) indicated that the prevalence rate of obesity in Korean population has increased from 29.2% in 2001to 30.7% in 2008. Similarly the prevalence rates of diabetes and hypercholesterolemia in the population have increased from 8.6% and 9.1% in 2001 to 9.8% and 10.9% in 2008, respectively. Any medicine used to treat obesity, diabetes and hypercholesterolemia has its own side effects. Therefore, this experiment was planned to find hypolipidaemic effect, if any in natural materials and resources from Chinese medicine (Yoon and Park 2010; Choi et al. 2008).

Turmeric (Curcuma longa L.) is a perennial plant belonging to the Zingiberaceae family mainly cultivated in

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India and China (Choi 2009). Turmeric has been used since long time in the preparation of culinary dishes in India as a colourant and flavouring agent (Agarwal and Sung 2009; Sharma et al. 2005). Several studies have shown that curcumin, the phytonutrient in rhizomes of turmeric, has antioxidant (Kumar et al. 2005), anti-inflammatory (Reddy and Lokesh, 1994), anti-mutagenic (Polasa et al. 1991) and hypolipidaemic (Arafa 2005; Kim et al. 2008) effects.

In oriental medicine it is believed that sulphur protects the spirit and strengthens bones and muscles. Sulphur is also used for maintenance of homeostasis and for the treatment of cold hands and feet and neural paralysis in oriental medicine (Park et al. 2003). However, sulphur has toxicity as manifested in direct injection and therefore needs to be processed for detoxification before using as medicine (Lee et al. 2010). Sulphur is processed through heat melting of mineral sulphur followed by separation of liquid and subsequent cooling (Lee et al. 2010).

In Western medicine, sulphur is used as local stimulant and in the treatment of constipation, haemorrhoids and skin diseases (Choi and Kim 2002). Sulphur is a component of many amino acids and present in glycoprotein, glutathione, collagen and is needed for synthesis of steroid hormones and several growth factors. The studies using processed sulphur in feedstuff were carried out (Cho et al. 2005; Jang et al. 2006) to show its toxicity (Song et al. 2007; Lee et al. 2010), but no study is reported about the use of processed sulphur in food and its effects. Thus, this study was planned to investigate the effects of turmeric powder and processed sulphur separately and together as food supplements on the body weight gain, fat deposition and liver and serum lipid profile of rats fed with a high fat diet.

Table 1 Composition of the experimental diets* of rats (g/kg diet)

*AIN-93 G (Dyets Inc., Bethlehem, PA, USA)
CON: control diet; HFD: high fat diet; T: 10% turmeric powder added to HFD
TS: 10% turmeric powder, 0.19% processed sulphur added to HFD
S: 0.38% processed sulphur added to HFD

Materials and methods

The turmeric powder obtained from Haenam, Korea was cleaned and dried before use. The processed sulphur was procured from a local company (Ebada Environment-friendly Lab Co. Ltd, Seoul). Chloroform and methanol were purchased from Samchun Chemical (Seoul, Korea). All other chemicals used were of analytical grade.

Analysis of turmeric powder and processed sulphur Moisture, crude protein, crude fat, crude fiber and ash contents were analysed as per AOAC (2005). Carbohydrate content was calculated by subtracting the sub-total of moisture, crude protein, crude fat, crude fiber and ash percent from hundred. Sulphur content in processed sulphur was analysed using auto-analyzer (EA 1110, ThermoQuest CE Instrument, Germany).

Animal experiment Twenty-five 5 weeks old Wistar male rats were procured from Central Lab Animal, Seoul, Korea. The animals were housed in individual stainless steel cages and given a 7 day adjustment period before starting the experiment (ethical clearance number IACUC No. KU10035). The rats were divided into five groups with five rats in each group viz. I. Common diet group (CON); II. High fat diet group (HFD); III. Group received 10% of turmeric powder with high fat diet (T); IV. Group received 10% of turmeric powder and 0.19% of processed sulphur with high fat diet (TS) and V. Group received 0.38% of processed sulphur with high fat diet (S). The experimental feeding continued for 6 weeks (Table 1). Room temperature and relative humidity were maintained at 22±2°C, 60±5% respectively with room

Ingredient	Groups						
	CON	HFD	T	TS	S		
Casein	200	233.1	221	221	233.1		
Sucrose	100	201.4	201.4	201.4	201.4		
Dextrose	132	116.5	44	44	116.5		
Corn starch	397.5	84.8	84.8	84.8	84.8		
Cellulose	50	58.3	46.5	44.6	54.5		
Soybean Oil	70	29.1	25.5	25.5	29.1		
Lard	0	206.9	206.9	206.9	206.9		
Processed sulphur	0	0	0	1.875	3.75		
Turmeric powder	0	0	100	100	0		
Mineral mix	35	52.4	52.4	52.4	52.4		
Vitamin mix	10	11.7	11.7	11.7	11.7		
L-Cystine	3	3.5	3.5	3.5	3.5		
Choline bitartrate	2.5	2.33	2.33	2.33	2.33		
Total (g)	1000	1000	1000	1000	1000		
Total (kcal)	4000	4661	4661	4661	4661		



Table 2 Effect of turmeric and sulphur on the body weight gain and FER of the rats during six weeks feeding trial

	CON	HFD	T	TS	S
Initial body weight (g)	$186\pm3^{\mathrm{NS}}$	188±4	180±7	181±7	179±9
Final body weight (g)	330 ± 2^{c}	$381\!\pm\!1^a$	348 ± 6^{bc}	$348{\pm}20^{bc}$	$370{\pm}3^{ab}$
Body weight gain (g)	144±5°	192 ± 14^{a}	167 ± 11^{b}	167 ± 14^{b}	190 ± 8^a
Total feed intake (g)	786 ± 1^{ab}	785 ± 6^{ab}	$778 \pm 6^{\mathrm{b}}$	$783\!\pm\!6^{ab}$	$789{\pm}5^a$
Total energy intake (kcal)	3145±5°	3660 ± 29^{ab}	3627 ± 27^{b}	3649 ± 26^{ab}	3676 ± 21^{a}
FER (%)	0.18 ± 0.01^{c}	$0.25\!\pm\!0.02^a$	0.22 ± 0.01^{b}	$0.21\!\pm\!0.02^{b}$	0.24 ± 0.01^a

 $^{^{}a,b,c}$ Significantly different at p<0.05 level by Duncan's multiple range test

All the values are mean \pm S.D (n=5)

CON: control diet; HFD: high fat diet; T: 10% turmeric powder added to HFD

TS: 10% turmeric powder, 0.19% processed sulphur added to HFD

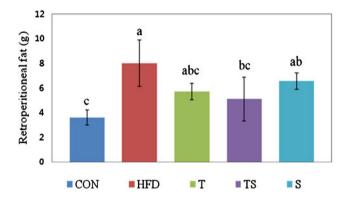
S: 0.38% processed sulphur added to HF

lighting at a 12 h light/dark cycle. *Ad-libitum* water was provided, but the feed was limited to 20 g/day/head. The body weights of the rats were noted on every 7th day. The feed intake was calculated by subtracting the amount (g) of feed remaining next day from the amount (g) of feed given previous day. The feed efficiency ratio (FER) was calculated by dividing the total body weight gain with the total feed intake in 6 weeks. At the end of experimental feeding the rats were fasted for 24 h and anesthetized using diethyl ether. Then abdominal incision was given and blood was collected from the inferior vena cava and serum was separated by centrifugation. Weight of liver, kidney, retroperitoneal fat and epididymal fat were recorded and stored at -80°C.

Liver lipid profile The total lipids in liver were analyzed using Folch method (Folch et al. 1957). Briefly, 0.5 g of liver was taken and 5 ml of chloroform-methanol (2:1 v/v) was added. Extraction was done three times using chloroform-methanol. The extracted liquid was taken in a test tube and was evaporated by nitrogen gas (Park et al. 2007). After evaporation, the test tube was weighed and the total lipid in liver was calculated. The lipid was dissolved in 1 ml chloroform-methanol from which 0.1 ml was taken and evaporated by nitrogen gas and 1 ml ethanol was added (Sale et al. 1984). From this solution 0.2 ml was taken and 0.5 ml of distilled water was added. This solution was used for the estimation of total cholesterol and triglyceride (TG) using a kit (AM 202-K & AM 157S-K, Asan Pharmaceutical, Seoul, Korea).

Serum lipid profile The serum was analyzed for the total cholesterol, high-density lipoprotein (HDL) cholesterol, low-density lipoprotein (LDL) cholesterol and triglyceride (TG) using Hitachi 7080 spectrophotometer (Hitachi, Japan).

Statistical analysis All experiments were repeated three times. Data were pooled and statistical analysis was performed using the SAS program (SAS Institute Inc 1999). The levels of significance among means were tested at p<0.05 with Duncan's multiple range tests.



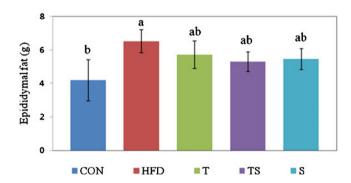
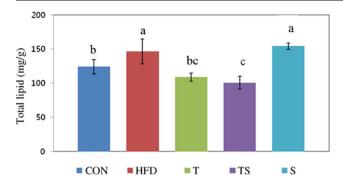
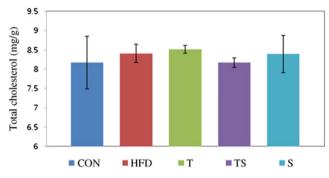


Fig. 1 Effect of turmeric and sulphur on the retroperitoneal and epididymal fat deposition in rats during 6 weeks feeding trial. ^{a, b, c} significantly different at p<0.05 level by Duncan's multiple range test. All the values are mean \pm S.D (n=5). CON: control diet; HFD: high fat diet; T: 10% turmeric powder added to HFD; TS: 10% turmeric powder, 0.19% processed sulphur added to HFD; S: 0.38% processed sulphur added to HFD







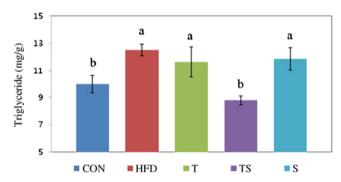


Fig. 2 Effect of turmeric and sulphur on the total lipid, total cholesterol and triglyceride in livers of rats during 6 weeks feeding trial. ^{a, b, c} significantly different at p<0.05 level by Duncan's multiple range test. All the values are mean \pm S.D (n=5). CON: control diet; HFD: high fat diet; T: 10% turmeric powder added to HFD; TS: 10% turmeric powder, 0.19% processed sulphur added to HFD; S: 0.38% processed sulphur added to HFD

Results and discussion

Composition of turmeric powder and processed sulphur The turmeric powder contained: moisture 11.3%, carbohydrate 64.33%, crude protein 10.7%, crude fat 3.2%, crude fiber 3.87% and ash 6.6%. The processed sulphur contained 100% sulphur.

Body weight gain, feed intake and FER The body weight gain and FER in rats belonging to T and TS groups were significantly (p<0.05) lower than that of the HFD and S group rats during the 6 week period (Table 2). However, there was no significant difference in weight gain and FER between HFD and S group rats. The results indicated that turmeric powder was associated with significant (p<0.05) decrease in body weight gain and FER in T and TS group rats since similar effect was not observed in rats of S group. Our results suggest that turmeric powder has beneficial effects of reducing body weight gain even after taking high fat diet.

Organ and fat weight The liver weight of T and TS group rats were significantly (p<0.05) higher than the HFD group rats and the kidney weight in rats of T group was significantly (p<0.05) higher than that of the HFD group rats (data not shown). In the postmortem nothing unusual was found in the organs. The retroperitoneal fat weights of the T, TS and S group rats were lower than that of the HFD group rats. The TS group rats had a significant (p<0.05) reduction in retroperitoneal fat weight compared to the HFD group rats (Fig. 1). The epididymal fat weights of the T, TS and S groups rats showed a decreasing tendency than that of the HFD group rats, however, there were no significant differences (Fig.1). These data suggest that turmeric powder has beneficial effects on body fat deposition especially in peritoneal fat deposition.

Hepatic lipid profile The hepatic total lipid level in the T and TS group rats were significantly (p<0.05) lower than

Table 3 Effect of turmeric and sulphur on the serum cholesterol and triglyceride of rats during six weeks feeding trial

Item	CON	HFD	T	TS	S
Total-cholesterol (mg/dL)	53.0±2.1 ^b	54.0±1.5 ^b	78.0±1.0 ^a	82.0±10.1 ^a	58.0±5.0 ^b
HDL-Cholesterol (mg/dL)	18.8 ± 0.2^{c}	18.9 ± 1.2^{bc}	21.1 ± 1.8^{ab}	23.1 ± 1.1^{a}	19.4 ± 1.0^{bc}
LDL-Cholesterol (mg/dL)	2.8 ± 0.4^{b}	3.1 ± 0.4^{b}	5.0 ± 0.7^{a}	5.3 ± 1.2^{a}	3.5 ± 0.8^{b}
Triglyceride (mg/dL)	8.0 ± 1.2	9.0 ± 1.2	6.0 ± 1.5	9.0 ± 3.1	9.0 ± 1.0

 $^{\mathrm{a,b,c}}$ significantly different at p<0.05 level by Duncan's multiple range test

All the values are mean \pm S.D (n=5)

CON: control diet; HFD: high fat diet; T: 10% turmeric powder added to HFD

TS: 10% turmeric powder, 0.19% processed sulphur added to HFD

S: 0.38% processed sulphur added to HFD



that of the HFD group rats, however, there was no significant difference between S and HFD group rats and T and TS group rats in respect of hepatic total lipid level (Fig. 2). These data suggest that the significant reduction in the total lipid is mainly due to turmeric however, sulphur has no effect on it. The hepatic triglyceride level in the TS group rats was significantly (p<0.05) lower than that of the HFD group. Our results suggest that feeding turmeric powder has decreased the hepatic total lipid and concurrently feeding turmeric powder and processed sulphur has decreased the hepatic TG in rats.

Serum lipid profile The serum total cholesterol and LDL-cholesterol contents of the T and TS group rats were significantly (p < 0.05) higher than that of the HFD group rats (Table 3). However, no significant effect was observed in rats of S group. It appears from the result that turmeric is associated with increase in total cholesterol and LDL-cholesterol levels in rats, however, sulphur has no such effect. The serum HDL-cholesterol contents of the TS group rats was significantly (p<0.05) higher than that of the HFD and CON group rats. T group rats also had significantly (p < 0.05) higher HDL-cholesterol level than CON group rats (Table 3). Further, these results suggest that turmeric must be associated to this increase in HDL-cholesterol, since there is no significant increase in S group rats compared to CON group rats. In contrary to our results hypocholesterolemic effects of curcumin on rats fed with turmeric was reported (Arafa 2005; Kim et al. 2008). But Larry et al. (2007) reported that serum cholesterol levels of normal people were not affected by curcumin intake. Thus, increased serum cholesterol seems to be limited to the population fed with high fat diet which may not happen with the population with normal diet.

Conclusion

Based on the results it may be concluded that turmeric powder along with processed sulphur can reduce the body weight gain, feed efficiency ratio, body fat deposition, hepatic total lipid and triglyceride and increase serum HDL-cholesterol in rats even after feeding with a high fat diet. Further studies need to be carried out for better understanding the beneficial effect of turmeric sulphur combination to utilize their hypolipidaemic effect.

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