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Supplement of bamboo extract lowers serum monocyte chemoattractant protein-1 concentration in mice fed a diet containing a high level of saturated fat

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Abstract

Monocyte chemoattractant protein-1 (MCP-1) is an inflammatory chemokine up-regulated in obese subjects, contributing to the development of type 2 diabetes. The present study investigated the inhibitory effect of an ethanol–water extract from bamboo (BEX, *Phyllostachys edulis*) on the blood concentration of MCP-1. C57BL/6J mice were fed a standard diet or a high-fat diet with or without the BEX supplement (11 g dry mass/17 000 kJ) for 6 months. A total of ten mice were used in each group. Body weight and food consumption were measured weekly. After euthanisation, the weight of visceral fat and circulating MCP-1 concentration were measured. In comparison with the standard control group, the high-fat control group had increased body weight, abdominal fat storage and serum MCP-1 concentration by 60 % ($P<0·001$), 266 % ($P<0·001$) and 180 % ($P<0·01$), respectively. In comparison with the high-fat control group, the high-fat BEX group showed a 3 % decrease in body weight ($P<0·01$), 24 % decrease in mesenteric fat depot ($P<0·01$) and 49 % decrease in serum MCP-1 concentration ($P<0·05$). The present study suggests that the BEX supplement in the high-fat diet ameliorates elevated MCP-1 concentrations in the blood, and whether this is related to modulated endocrine properties of the visceral fat is to be studied.

Key words: Monocyte chemoattractant protein-1: High-fat diet: Bamboo extract: Mesenteric fat

Monocyte chemoattractant protein-1 (MCP-1/chemokine (C–C motif) ligand 2) is a member of the C–C chemotactic cytokine (chemokine) family, produced by multiple cell types constitutively or after induction[1]. The circulating level of MCP-1 was found to be approximately 50 % higher in obese mice[2] and in human subjects with type 2 diabetes[3] in comparison with controls. MCP-1 recruits monocytes into adipose tissues and enhances obesity-associated chronic inflammation[4] and insulin resistance[5]. It also facilitates the expansion and remodelling of the adipose tissue during the development of obesity through an angiogenic effect on the endothelial cells[6]; furthermore, this chemokine can decrease the liposynthesis ability of adipocytes[2] and subsequently elevate the NEFA level in the circulation[4], exerting lipotoxicity in the periphery[7]. Therefore, inhibiting MCP-1 overproduction has become a preventive strategy for obesity-induced type 2 diabetes.

Our previous study has shown that an ethanol–water extract from bamboo (BEX, *Phyllostachys edulis*) efficiently protected murine muscle C2C12 cells from lipotoxicity[8], an obesity-related condition leading to inflammation and insulin resistance[9,10]. In the present study, it is further revealed that the BEX as a dietary supplement significantly decreased the circulating level of MCP-1 in mice treated with a diet containing a high level of saturated fat, with concurrence of decreased weight of the mesenteric fat depot.

Experimental methods

Bamboo extract

The BEX used in the present study was provided by Golden Basin LLC (Kailua, HI, USA). It is prepared from fresh leaves and small branches of bamboo (*P. edulis*) in Hunan Province, China, through a patented ethanol–water extraction procedure (Chinese invention patent, CN 1287848A).

Animals

Male C57BL/6J mice at 4 weeks of age were purchased from Jackson Laboratories (Bar Harbor, MN, USA). The animals were housed three to four per cage and had *ad libitum* access to water and food. The room temperature was controlled at 20°C, and lighting was turned on and off with 12 h

Abbreviations: BEX, bamboo extract; MCP-1, monocyte chemoattractant protein-1.

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intervals. Institutional and national guidelines for the care and use of animals were followed, and all experimental procedures involving animals were approved by the Institutional Animal Care and Use Committee at the University of Hawaii (Honolulu, HI, USA).

Dietary treatment

After 1 week of acclimatisation with regular rodent chow, mice were fed a standard diet (10 % energy from fat) or a high-fat diet (60 % energy from fat) with or without the BEX supplement (11 g dry mass per 17 000 kJ) for 6 months. A total of ten mice were used in each dietary group. Body weight and food consumption were measured weekly. All diets were purchased from Research Diets (New Brunswick, NJ, USA). The dietary composition is listed in Table 1.

Measurement of the abdominal fat pads

After euthanisation, epididymal fat, perirenal fat and mesenteric fat were collected and weighed.

Serum monocyte chemoattractant protein-1 quantification

Blood was collected through cardiac puncture. MCP-1 concentrations in the sera were measured using a Cytometric Bead Array – Mouse MCP-1 Flex Set (BD Biosciences, Bedford, MA, USA).

Statistical analysis

Statistical analysis was performed by two-way ANOVA with Bonferroni's *post hoc* test using the software Prism 4.0a (GraphPad Software, Inc., La Jolla, CA, USA) and Stata 11.0 (StataCorp LP, College Station, TX, USA) (Table 2). The weekly record of body weight was analysed using linear regression with Huber correction and random-effects regression to account for multiple measurements per mouse. $P \leq 0{\cdot}05$ was considered statistically significant.

Results

Table 2 summarises the major findings of the present study. During the 6 months of treatment, the high dietary fat content increased the daily energy intake by approximately 30 % ($P < 0{\cdot}0001$). The BEX supplement did not affect the energy intake. The body weight of mice at both start and end points is shown. High-fat diets resulted in an average of 60 % increase in body weight ($P < 0{\cdot}0001$) at the end point. When the weekly record of the body weight (data not shown) was analysed, the BEX supplement in the high-fat diet was found to slightly decrease (−3 %, $P < 0{\cdot}01$) the weight gain of mice.

The high dietary fat content also increased the total weight of the abdominal fat by approximately threefold. When the weight of the individual fat depot was analysed, the BEX was found to increase the epididymal fat by 20 % (0·37 g, $P < 0{\cdot}05$) and decrease the mesenteric fat by 24 % (0·52 g, $P < 0{\cdot}01$), but did not affect the total weight of the visceral fat. An interaction between the fat content and the BEX was found to play an important role in regulating the weight of the mesenteric fat depot ($P < 0{\cdot}01$).

Most interestingly, the BEX supplement dramatically decreased high-fat, diet-induced elevated MCP-1 concentration in the serum (−49 %, $P < 0{\cdot}05$), and whether this is related to modulated endocrine properties of the visceral fat, especially the mesenteric fat, is to be studied.

Table 1. Composition of the diets used in the present study

Diets...	Standard control		Standard BEX		High-fat control		High-fat BEX	
	g	kJ	g	kJ	g	kJ	g	kJ
Ingredients								
Casein, 80 mesh	200	3349	200	3349	200	3349	200	3349
L-Cystine	3	50	3	50	3	50	3	50
Maize starch	315	5275	315	5275	0	0	0	0
Maltodextrin 10	35	586	35	586	125	2093	125	2093
Sucrose	350	5862	350	5862	68·8	1151	68·8	1151
Cellulose, BW200	50	0	50	0	50	0	50	0
Soyabean oil	25	942	25	942	25	942	25	942
Lard	20	754	20	754	245	9232	245	9232
Mineral mix S10026	10	0	10	0	10	0	10	0
Dicalcium phosphate	13	0	13	0	13	0	13	0
Calcium carbonate	5·5	38	5·5	38	5·5	38	5·5	38
Potassium citrate, 1 H_2O	16·5	0	16·5	0	16·5	0	16·5	0
Vitamin mix V10001	10	167	10	167	10	167	10	167
Choline bitartrate	2	0	2	0	2	0	2	0
Bamboo extract (dry mass)	0	0	11	0	0	0	11	0
Water from the bamboo extract	0	0	11	0	0	0	11	0
FD&C Yellow Dye no. 5	0·05	0	0·025	0	0	0	0·025	0
FD&C Red Dye no. 40	0	0	0	0	0	0	0·025	0
FD&C Blue Dye no. 1	0	0	0·025	0	0·05	0	0	0
Total	1055·1	16 986	1077·1	16 986	773·9	16 986	795·9	16 986

BEX, bamboo extract.

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Table 2. Energy intake, body weight, abdominal fat and serum monocyte chemoattractant protein-1 (MCP-1) concentration in mice

(Mean values and standard deviations)

Group...	Standard control		Standard BEX		High-fat control		High-fat BEX	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Energy intake (kJ/d)*	39·4[a]	4·0	37·9[a]	4·5	50·3[b]	5·3	50·9[b]	3·6
Body weight (g) (start)	17·8[a]	2·1	18·2[a]	1·8	18·0[a]	0·9	18·4[a]	1·1
Body weight (g) (end)	28·7[a]	3·1	28·3[a]	2·5	47·5[b]	2·2	44·6[b]	2·6
Epididymal fat (g)	0·65[a]	0·26	0·78[a]	0·38	1·84[b]	0·24	2·21[c]	0·37
Perirenal fat (g)	0·17[a]	0·07	0·24[a]	0·16	0·85[b]	0·31	0·74[b]	0·19
Mesenteric and omental fat (g)	0·22[a]	0·08	0·30[a]	0·14	2·17[b]	0·23	1·65[c]	0·47
Total abdominal fat (g)	1·04[a]	0·38	1·32[a]	0·66	4·86[b]	0·58	4·69[b]	0·39
Serum MCP-1 (pg/ml)	16·26[a]	6·58	19·34[a]	3·23	45·78[b]	9·56	23·37[a]	1·75

BEX, bamboo extract.
[a,b,c] Mean values within a row with unlike superscript letters were significantly different ($P<0·05$).
* Energy intake is the average of the weekly measurements.

Discussion

Using the same diet containing a high level of saturated fat, Yu *et al.*[11] treated C57BL/6 mice for 3 months and compared the MCP-1 expression and secretion in four types of adipose tissues: mesenteric, epididymal, perirenal and subcutaneous. While the amounts of MCP-1 released by the epididymal, perirenal and subcutaneous fat depots were approximately the same, this level quadruplicated in the mesenteric fat. Mesenteric adipose tissue-conditioned medium also induced the highest degree of macrophage migration and stimulated pro-inflammatory cytokine production in macrophages. These findings indicate that in comparison with the other fat depots, the mesenteric fat tissue has a more pronounced role in obesity-associated inflammation. The present study showed that the BEX supplement in a high-fat diet decreased the weight of the mesenteric fat by 0·52 g, and therefore it may attenuate MCP-1 secretion from this tissue and subsequently contribute to the decrease in MCP-1 in the circulation. Although the BEX increased the weight of the epididymal fat by 0·37 g, this may not compensate the change caused by the mesenteric fat due to the dramatic difference between the MCP-1 secretion abilities of these two fat depots. This suggests that the BEX may alter the distribution of fat storage in the visceral adipose tissues and lower MCP-1 secretion as a final result. Although white adipose tissue is a major source of MCP-1[2], this chemokine is also produced in other tissues[12–14], and therefore a systematic study is needed to evaluate the tissue-specific effect of BEX.

Other natural products that inhibit the overproduction of MCP-1 in obese/diabetic rodents and in cell culture models include traditional Asian medicine[15–18], extracts from herbs[19], spices[20,21], fruits and vegetables[22–25]. Due to the use of different models and experimental procedures, and variable purity of the extracts, it is difficult to perform an accurate comparison between the efficacy of BEX and other reported natural products. The daily dose of BEX used in the present study was 773 mg/kg body weight for mouse, and this corresponds to 63 mg/kg body weight for human subjects (3·8 g/d for a 60 kg adult) when the body surface area normalisation method is used for an allometric dose translation[26].

The BEX used in the present study consists of approximately 50 % water, 20 % saccharides, 10 % protein and 20 % other components. The active component(s) contributing to the effects described earlier are to be further determined. The extraordinary abundance of the raw material is a major advantage of this natural product. *P. edulis* is known for its fast growth, wide geographical distribution and easy propagation. The raw materials (small branches and leaves) used for BEX production are by-products of the bamboo timber industry. Therefore, the present study suggests a potential nutraceutical application of a rich and environmentally friendly natural resource.

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