Available online at www.sciencedirect.com
ScienceDirect
Nutrition Research 30 (2010) 49–56
Nutrition Research
www.nrjournal.com

White button mushroom (*Agaricus bisporus*) lowers blood glucose and cholesterol levels in diabetic and hypercholesterolemic rats

Sang Chul Jeong[a], Yong Tae Jeong[b], Byung Keun Yang[b], Rezuanul Islam[b], Sundar Rao Koyyalamudi[a], Gerald Pang[a,*], Kai Yip Cho[a], Chi Hyun Song[b]

[a]*Center for Plant and Food Science, College of Health and Science, University of Western Sydney, Penrith South DC, NSW 1797, Australia*
[b]*Department of Biotechnology, Daegu University, Gyoungbuk 712-714, South Korea*

Received 19 September 2009; revised 11 November 2009; accepted 14 December 2009

Abstract

Agaricus bisporus (white button mushroom; WBM) contains high levels of dietary fibers and antioxidants including vitamin C, D, and B_{12}; folates; and polyphenols that may provide beneficial effects on cardiovascular and diabetic diseases. The objective of this study was to examine the hypothesis that intake of the fruiting bodies of WBM regulates anticholesterolemic and antiglycemic responses in rats fed a hypercholesterolemic diet (0.5% cholesterol; 14% fat) and rats with type 2 diabetes induced by injection of streptozotocin (STZ) (50 mg/kg body weight), respectively. The STZ-induced diabetic male Sprague-Dawley rats fed the *Agaricus bisporus* powder (ABP; 200 mg/kg of body weight) for 3 weeks had significantly reduced plasma glucose and triglyceride (TG) concentrations (24.7% and 39.1%, respectively), liver enzyme activities, alanine aminotransferase and aspartate aminotransferase (11.7% and 15.7%, respectively), and liver weight gain ($P < .05$). In hypercholesterolemic rats, oral feeding of ABP for 4 weeks resulted in a significant decrease in plasma total cholesterol (TC) and low-density lipoprotein (LDL) (22.8% and 33.1%, respectively) ($P < .05$). A similar significant decrease in hepatic cholesterol and TG concentrations was observed (36.2% and 20.8%, respectively) ($P < .05$). Decrease in TC, LDL, and TG concentrations was accompanied by a significant increase in plasma high-density lipoprotein concentrations. It was concluded that *A bisporus* mushroom had both hypoglycemic and hypolipidemic activity in rats.

Keywords: *Agaricus bisporus*; White button mushroom; Rats; Diabetes; Hypercholesterolemia; Hyperglycemia
Abbreviations: ABP, *Agaricus bisporus* powder; ALT, alanine aminotransferase; AST, aspartate aminotransferase; BW, body weight; HDL, high-density lipoprotein; LDL, low-density lipoprotein; STZ, streptozotocin; TC, total cholesterol; TG, triglyceride.

1. Introduction

Cardiovascular disease and diabetes are the major causes of morbidity and mortality in Western countries and in the Asia-Pacific region. Epidemiological studies suggested that the risk factors for cardiovascular disease and diabetes include hypercholesterolemia and hyperglycemia, which are largely influenced by diet [1,2]. Laboratory and clinical studies have shown that a diet supplemented with fruit and vegetables had beneficial effects on diabetes and atherosclerosis [3-5]. *Agaricus bisporus*, commonly known as white button mushroom (WBM), constitutes the bulk of the total mushrooms consumed in most Western countries. However, little information is available on its health properties because greater attention has been primarily focused on its immune modulating and antitumor properties [6-9]. Edible mushrooms and their constitutive active compounds have been

* Corresponding author. Tel.: +61 2 9685 9987 (Office phone); fax: +61 2 9685 9915.
E-mail address: g.pang@optusnet.com.au (G. Pang).

0271-5317/$ – see front matter © 2010 Elsevier Inc. All rights reserved.
doi:10.1016/j.nutres.2009.12.003

described to have beneficial effects on hyperglycemia and hypercholesterolemia [10,11]. White button mushroom has high content of acidic polysaccharides, dietary fiber, and antioxidants, including vitamins C, B_{12}, and D; folate; ergothioneine; and polyphenol [12-15], suggesting that the mushroom may have potential antiinflammatory, hypoglycemic, and hypocholesterolemic effects. In an earlier study, rats fed a diet containing WBM dietary fiber had lower serum cholesterol levels and greater expression of hepatic low-density lipoprotein (LDL) receptor messenger RNA gene compared with rats fed a fiber-free diet [12]. However, no further detailed investigations have been conducted to determine whether the mushroom has the potential to exert a cholesterol-lowering effect in a dietary setting of high saturated fat and cholesterol.

Diets high in saturated fat and cholesterol have been shown to contribute to hypercholesterolemia and metabolic disturbances, which in turn may lead to progression toward overt hyperglycemia/type 2 diabetes in humans and animals (reviewed by Mathe [16], Kuller [17], and Layman et al [18]). Given the high dietary fiber and antioxidants in WBM [12,15], a diet high in mushroom may be advantageous, namely, in lowering the dietary glycemic load.

To date, evidences regarding the biological activities of WBM were based on the fruiting body, the common edible form of mushrooms that contains a rich source of dietary fiber, acidic polysaccharides, nutritional antioxidants, vitamins, and polyphenols [11-15]. We hypothesized that in addition to its effects on cells of the immune system [9], tumor cells [9], and hepatic LDL receptor activity [12], WBM may also affect nutrient metabolism. Specifically, we hypothesized that WBM would have cholesterol-lowering and glucose-lowering effects. Accordingly, in diabetic and hyperlipidemic rats, we evaluated the effect of ingesting WBM fruiting bodies on blood glucose and cholesterol levels.

2. Methods and materials

2.1. Animals, diets, and induction of diabetes and hypercholesterolemia

Male 4-week-old Sprague-Dawley rats (100-120 g each) purchased from Daehan Biolink Co, Ltd (Seoul, Korea), were housed individually in stainless steel cages in a room with controlled temperature (22°C ± 2°C), a relative humidity of 55% ± 5%, and a 12-hour cycle of light and dark. Rats were maintained on a commercial pellet diet (Sam Yang Co, Seoul, Korea). The animal studies were approved by respective Animal Care and Ethics Committees of the Universities of Daegu, Korea, and the University of Western Sydney, New South Wales, Australia.

After acclimatization for 7 to 10 days, the rats were fasted for 12 hours before an intramuscular injection of streptozotocin (STZ) (Sigma-Aldrich, 50 mg/kg body weight [BW]) dissolved in citrate buffer at pH 4.5. Two days after STZ treatment, the rats were considered diabetic (as determined by nonfasting blood glucose levels of >300 mg/dL and positive glucose urine test [Glucocard Test Meter, Japan]) [19]. After induction of diabetes, the rats were maintained on a conventional diet for the duration of the experiment.

To induce hypercholesterolemia, after acclimatization for 7 to 10 days, normal rats were switched to a modified AIN-76 diet consisting of 55.5% carbohydrates, 14.0% fats (30% of total energy), 20.0% protein, 5.0% fiber, and 0.5% cholesterol by weight, as previously described [20]. Normal rats were fed the same diet without cholesterol, as shown in Table 1. The rats were maintained on their respective diets for 6 weeks to induce hypercholesterolemia and then for the whole experimental period.

2.2. Agaricus bisporus powder

Fresh fruiting bodies of *A bisporus* were obtained from mushroom growers. After freeze-drying, the dehydrated fruiting bodies were milled to a powder approximately less than 1 mm in particle size using a cyclotec grinder (Tecator, Hoganas, Sweden). The chemical composition of *Agaricus bisporus* powder (ABP) as determined by the Standard AOAC (Association of Official Analytical Chemists) methods [21] is shown in Table 2. For oral dosing, the freeze-dried ABP was reconstituted in physiologic saline. Based on our previous experience with dose-response studies of the metabolic effects of mushroom constituents [22], a dose of 200 mg/kg BW of ABP was used for administration to rats.

2.3. Experimental design

Normal, control, and experimental rats were randomly assigned accordingly to 3 groups, with each group consisting of 6 to 8 animals. The untreated groups consisted of normal (nondiabetic or nonhypercholesterolemic) rats. The experimental control groups consisted of STZ diabetic and hypercholesterolemic rats fed daily for 3 and 4 weeks, respectively, with 1 mL of physiologic saline alone administered by the intragastric route using a 21G ball-end feeding needle (Popper Instrument, Lake Success, NY). In the experimental treatment groups, the STZ diabetic and hypercholesterolemic rats were fed daily for the same period

Table 1
Ingredient composition of the AIN-76 diet with or without added cholesterol

Ingredient	Composition (g/100 g)	
	Control diet	Cholesterol diet
Casein	20.0	20.0
Dextrin	9.5	9.5
Sucrose	55.5	46.5
Lard	–	9.0
Soybean oil	5.0	5.0
AIN-mineral mix [a]	4.0	4.0
AIN-vitamin mix [a]	1.0	1.0
Cholesterol	–	4.0
α-Cellulose	5.0	1.0

[a] Mineral and vitamin mixtures.

Table 2
Proximate analysis of ABP

Component	Composition (%)
Moisture	3.09
Crude ash	8.71
Crude fat	1.51
Crude protein	32.03
Carbohydrate	36.01
Dietary fiber	18.65

with ABP in saline (200 mg/kg BW) administered intragastrically using the feeding needle. Throughout the feeding period, the nondiabetic and diabetic rats were maintained on a conventional diet, whereas hypercholesterolemic and nonhypercholesterolemic rats were maintained on AIN-76 and cholesterol-free AIN-76 diets, respectively.

Food and water intakes and body weights were recorded daily. Body weight of the rats was measured daily using an electronic balance. The measurement of food intake was carried out on individual rats. Briefly, each cage was supplied a known amount of the specific diet and the water. Each day, the remaining diet and water were measured. The difference between the 2 values represented the amount consumed in grams by the rat for that day. The remaining diets were topped up with fresh supply of diets and then weighed. Fresh drinking water was also supplied. The above processes were followed throughout the experimental period. The food efficiency ratio was calculated as the body weight gain divided by food intake per day.

After the final oral dosing, the animals were fasted for 9 to 14 hours before euthanization by intraperitoneal injection of 0.9 mL of pentobarbital (1:6.25 dilution of 325 mg/mL stock solution). Blood was collected by cardiac puncture. Plasma samples were collected from heparinized blood after centrifugation at 1100*g* for 10 minutes. After perfusion with cold saline, the kidneys, pancreas, spleen, and liver were collected, weighed, and then kept frozen at −70°C as previously described [19,20].

2.4. Biochemical analyses

Plasma glucose, total cholesterol (TC), triglyceride (TG), high-density lipoprotein (HDL) cholesterol, alanine aminotransferase (ALT), and aspartate aminotransferase (AST) levels were measured using commercial kits (Asan Pharm Co Ltd, Seoul, Korea). The LDL cholesterol, atherogenic index, and the cardiac risk factor were calculated using the following formulas:

$$\text{LDL cholesterol} = \text{total cholesterol} - \text{HDL cholesterol} - (\text{triglyceride} / 5)\ [23],$$

$$\text{Atherogenic index} = (\text{total cholesterol} - \text{HDL cholesterol}) / \text{HDL cholesterol}\ [24], \text{and}$$

$$\text{Cardiac risk factor} = \text{total cholesterol} / \text{HDL cholesterol}\ [25].$$

Lipids were extracted from liver and purified according to Folch and others [26]. Total cholesterol, TGs, and phospholipids levels were determined using commercial kits after treatment with Triton X-100.

2.5. Statistical analyses

Sample size calculations based on results from previous studies with exotic mushroom [19,20,22] were performed assuming that a 20% decrease in blood glucose and a 15% decrease in TC in rats fed the ABP would be clinically important and a 10% SD of glucose and cholesterol changes in both ABP-fed and control groups. Using a 2-tailed unpaired *t* test, 6 to 8 rats in each arm were required to obtain these effects, with an α error of .05 and β of .9. Each data set was expressed as means ± SEM. Group means were compared using a 1-way analysis of variance and LSD (Least Significant Difference) or Tamhane's test. A *P* value of less than .05 was considered significant. All data were analyzed using the SPSS 10.0 for Windows (SPSS Inc, Chicago, Ill). Power and sample size calculations were performed using the PS software version 3.0 for Windows (Vanderbilt University, Nashville, Tenn).

3. Results

3.1. Body weight gain, food and water intake, and organ weights

The BW gain of diabetic rats fed ABP and control rats were similar (Table 3). Daily food intakes of ABP-fed rats were lower than those of control rats ($P < .05$) but higher than those of normal rats ($P < .05$). The food efficiency ratios representing body weight gain relative to food intake were the same for both ABP-fed and control rats, but their ratios were significantly lower than that observed in normal rats ($P < .05$). Daily water intakes of diabetic rats fed ABP was significantly reduced compared with those of diabetic control rats ($P < .05$), whereas water intakes for both groups were significantly increased compared with normal rats ($P < .05$). Both kidney and liver weights were lower in rats fed ABP than in control diabetic rats ($P < .05$). There was no

Table 3
Effect of ingesting ABP on growth parameters in hyperglycemic rats

Parameters	Group A	Group B	Group C
BW gain (g/d)	6.20 ± 0.29^{b}	3.53 ± 0.20^{a}	3.45 ± 0.22^{a}
Food intake (g/d)	29.93 ± 0.51^{a}	47.13 ± 0.64^{c}	43.23 ± 1.03^{b}
Food efficiency ratio *	0.21 ± 0.01^{b}	0.07 ± 0.00^{a}	0.08 ± 0.00^{a}
Water intake (g/d)	42.12 ± 1.76^{a}	202.17 ± 5.98^{c}	166.73 ± 3.65^{b}
Kidney (g/100 g BW)	0.88 ± 0.02^{a}	1.32 ± 0.01^{c}	1.25 ± 0.02^{b}
Liver weight (g/100 g BW)	3.54 ± 0.08^{a}	4.85 ± 0.02^{c}	4.56 ± 0.06^{b}

Group A (nondiabetic); group B (diabetic); group C (diabetic + ABP). Values are presented as means ± SEM (n = 6). Means in columns with different superscripts are significantly different ($P < .05$).

* Body weight gain/food intake.

difference between groups with respect to spleen and pancreas weights (data not shown).

Body weights, food intakes, and food efficiencies were similar for ABP-fed and control hypercholesterolemic rats (Table 4). Except for lower food intakes, body weights and food efficiency ratios in hypercholesterolemic rats were significantly higher than those in normal rats ($P < .05$). However, hypercholesterolemic rats had higher liver weights than normal rats, but those fed ABP had lower liver weights than control rats ($P < .05$).

3.2. Effect of ABP intake on hyperglycemic and hypercholesterolemic response

Ingestion of ABP significantly reduced blood glucose concentration by 24.7% in STZ-induced diabetic rats compared with control diabetic rats (Fig. 1A) ($P < .05$). No significant difference in the TC levels among the groups was observed. However, diabetic rats fed ABP had significantly lower plasma TG levels than those in control diabetic rats (Fig. 1A) ($P < .05$). Lower plasma ALT and AST concentrations were observed in ABP-fed diabetic rats than in control diabetic rats ($P < .05$) (Fig. 1B). In addition, reduction in plasma ALT and AST levels was associated with lower liver weights in ABP-fed diabetic rats (Table 3).

Hypercholesterolemic rats fed ABP had significantly lower total plasma cholesterol and LDL levels compared with control rats (Fig. 2A) ($P < .05$). Significantly higher HDL concentrations were observed in hypercholesterolemic rats fed ABP than in control hypercholesterolemic rats ($P < .05$). However, ingestion of ABP had no significant effect on the level of TGs. A significant decrease in the liver cholesterol and TG concentrations was observed in ABP-fed hypercholesterolemic rats but not in control hypercholesterolemic rats (Fig. 2B) ($P < .05$). The atherogenic index and the cardiac risk factor of ABP-fed hypercholesterolemic rats were significantly lower than those of the hypercholesterolemic control rats (Fig. 2C) ($P < .05$).

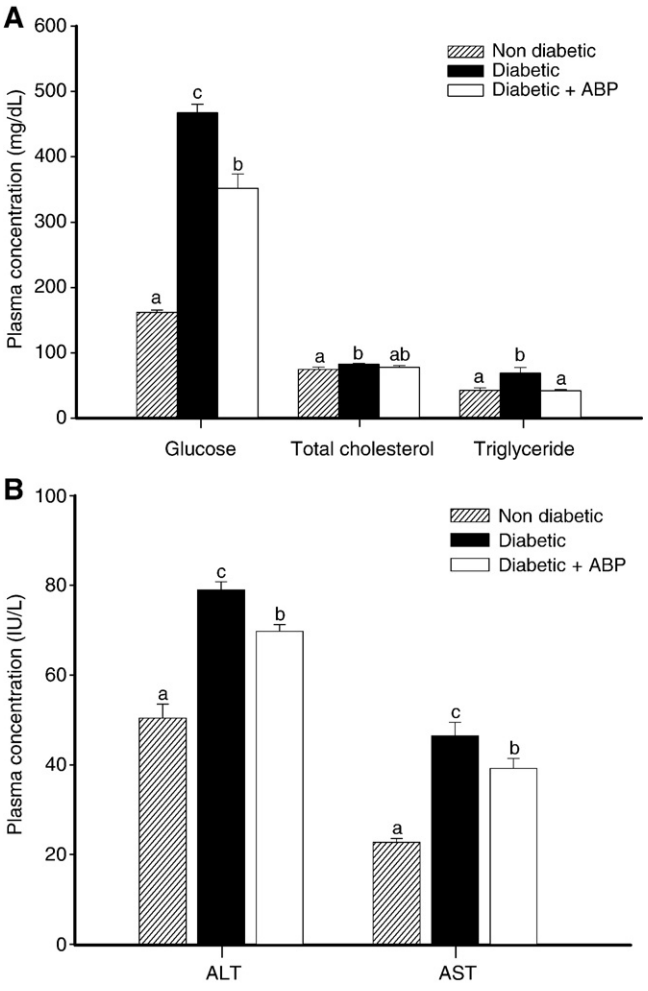

Fig. 1. Plasma glucose, TC, TG, and liver enzyme (ALT/AST) levels in diabetic rats. Nondiabetic (normal) and STZ-induced diabetic rats (control) were fed saline and diabetic rats were fed saline with ABP (200 mg/kg BW) daily for 3 weeks. Plasma glucose, TC, and TG (A) and ALT and AST (B) levels were measured using commercial kits after the 3-week feeding period. Each value on the figure represents means ± SEM of 6 rats. Comparisons between groups were performed by 1-way analysis of variance. Means for each variable with no common letters differ ($P < .05$).

4. Discussion

In the present study, we have demonstrated that the fruiting bodies of WBM have significant antihyperglycemic and antihypercholesterolemic activities in rats. The mushroom did not affect the concentration of plasma cholesterol but did lower plasma TG, ALT, and AST levels in diabetic rats. In hypercholesterolemic rats fed ABP, the reduction of plasma cholesterol is associated with the rise and the fall of plasma HDL and LDL levels, respectively. The mushroom also significantly lowered liver cholesterol and TG levels. In addition, the hypocholesterolemic effects of WBM were associated with lower atherogenic index and cardiac risk factor.

The present study found that intake of ABP resulted in lower food consumption in diabetic rats, which had a marked reduction in plasma glucose, as well as TG and cholesterol concentrations. The hypoglycemic effect of ABP on glucose response in diabetic rats can be explained by that it might have increased glucose use through promoting insulin

Table 4
Effect of ingesting ABP on growth parameters in hypercholesterolemic rats

Parameters	Group A	Group B	Group C
BW gain (g/d)	4.96 ± 0.25^{a}	6.13 ± 0.28^{b}	5.67 ± 0.19^{b}
Food intake (g/d)	26.58 ± 0.47^{b}	20.40 ± 0.47^{a}	21.16 ± 0.64^{a}
Food efficiency ratio *	0.19 ± 0.01^{b}	0.30 ± 0.02^{a}	0.27 ± 0.01^{a}
Liver weight (g/100 g BW)	3.47 ± 0.10^{a}	5.02 ± 0.15^{c}	4.56 ± 0.10^{b}

Group A (nonhypercholesterolemic); group B (hypercholesterolemic); group C (hypercholesterolemic + ABP). Values are presented as means ± SEM (n = 8). Means in the column with different superscripts are significantly different ($P < .05$).

* Body weight gain/food intake.

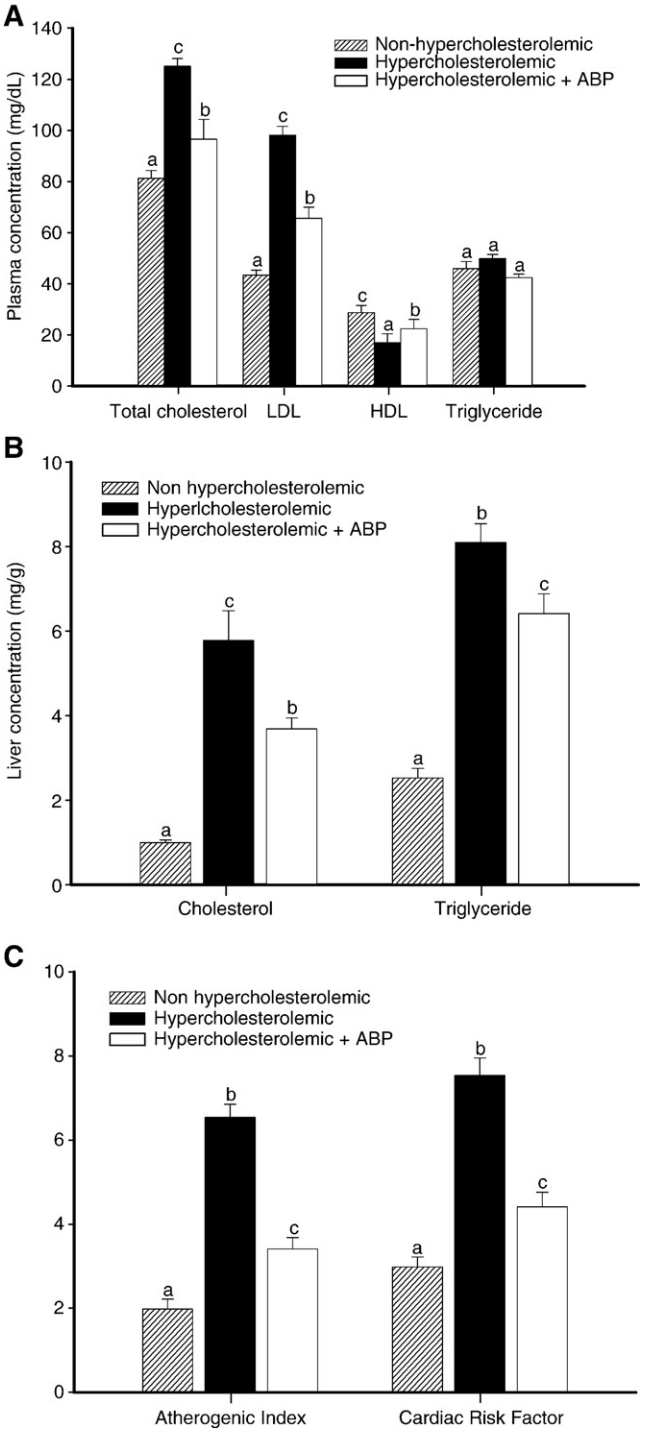

Fig. 2. Plasma and hepatic lipids, atherogenic index, and cardiac risk factor in hypercholesterolemic rats. Nonhypercholesterolemic (normal) and hypercholesterolemic control rats were fed saline and hypercholesterolemic rats were fed saline with ABP (200 mg/kg BW) daily for 4 weeks. Total cholesterol, LDL, HDL, and TG levels in plasma (A) and liver extract (B) were analyzed after the 4-week feeding period. The LDL cholesterol was calculated as the TC minus HDL cholesterol minus (TG/5). The atherogenic index was calculated as the TC minus HDL cholesterol divided by HDL cholesterol (C). The cardiac risk factor was calculated as the TC divided by HDL cholesterol (C). Values on the figure represent means ± SEM of 8 rats. Comparisons between groups were performed by 1-way analysis of variance. Means for each variable with no common letters differ ($P < .05$).

secretion [27]. The high dietary fiber content (19%) and possibly other carbohydrate components in ABP may account for its glucose-lowering effect in a way similar to other high fiber and carbohydrate-containing mushrooms [11,22,28] and single foods [29]. In obese mice, dietary nonstarch carbohydrate has been reported to stimulate increased secretion of insulin that correlated with a marked low glucose response [30]. *A bisporus* mushroom contains a lectin-like molecule that has been shown to stimulate insulin and glucagon release from islet cells [31], a key metabolic pathway involved in regulating glucose response. The high fiber content of ABP may act as a barrier to digestive enzyme action [6,32,33], thereby, contributing to lower blood glucose response. Another possible explanation for the glucose-lowering effect of ABP is that bacterial fermentation of ABP fiber, which is rich in polysaccharide and oligosaccharide, in the colon may lead to the production of short-chain fatty acids such as acetate, propionate, and butyrate [34]. Indeed, intake of *A bisporus* dietary fibers has been reported to generate propionate in rats fed a cholesterol-free diet [12]. The glucose-lowering effect of propionate has been reported to be associated with gluconeogenesis and the regulation of serum lipid levels [35,36].

We also observed lower plasma liver enzyme ALT and AST concentrations in diabetic rats fed ABP. This suggests that intake of ABP may protect against STZ-induced inflammation in the liver, which may account for the low liver weight—a finding consistent with that observed from our earlier studies on medicinal mushrooms [19,37,38].

It is well documented that elevated levels of total plasma cholesterol and TG levels are associated with complications of diabetes mellitus [39,40]. In the present study, lower plasma TC and TG concentrations were observed in diabetic rats fed the ABP, indicating that consumption of ABP had a beneficial effect in suppressing cholesterol and TG levels. Kidney and liver dysfunction is known to be associated with complications of diabetes in rodents [41,42]. Our present study showed that lower TC and TG concentrations found in diabetic rats fed ABP were associated with lower liver and kidney weights. This suggests that decrease in TG and TC concentrations may reflect a normal liver function, as demonstrated by decreased plasma ALT/AST concentrations and lower liver and kidney weights in the ABP-fed diabetic rats. It has been described that elevated blood glucose response is associated with increased serum cholesterol and TG concentrations and impaired liver function in STZ-induced diabetic rats and that, when the rats were fed mushroom extracts, the effects were reversed [19,37,38]. In this study, similar effects were observed in diabetic rats fed ABP. Although the mechanism of action is unclear, the anti-TG and anti-TC activities of ABP may be attributable to the fermentation of dietary fiber in diabetic rats fed ABP. Short-chain fatty acid such as propionate generated by bacterial fermentation of dietary fibers has been shown to inhibit hepatic cholesterol synthesis [43,44]. Furthermore, antioxidants and antiinflammatory activities of polyphenols and

ergothioneine known to occur in *A bisporus* [15] may provide protection against STZ-induced liver damage in diabetic rats [45,46], as well as influence glucose response [47]. However, detailed characterization of the metabolic pathways associated with the hypoglycemic effects of ABP will require further research.

Ingestion of ABP was effective in significantly lowering LDL and raising HDL concentrations in hypercholesterolemic rats. The beneficial effects of ABP were also reflected in the atherogenic index and the cardiac risk factor, both of which were lower in ABP-fed hypercholesterolemic rats than in control rats. Taken together, these results are in agreement with those reported in earlier studies with exotic mushrooms [19,20,48]. The reduction in plasma LDL and elevated HDL concentrations were associated with lower liver weight as well as lower liver TC and TG concentrations in hypercholesterolemic rats fed ABP. The observed beneficial effects of *A bisporus* on hypercholesterolemia are likely to be complex, probably involving a combination of bioactive components in the mushrooms, including short-chain fatty acids generated by bacterial fermentation of fibers in the colon. For example, numerous studies have shown that propionate generated from dietary fibers containing polysaccharides exhibits hypocholesterolemic effects and offsets acetate generation, which tends to increase serum cholesterol via a mechanism probably involving liver lipogenesis [43,49,50]. Attempts to elucidate the mechanism involved in the cholesterol-lowering effect of dietary mushroom fiber have been reported [10,47,51,52]. It has been suggested that mushroom dietary fiber might bind bile acids to reduce their entry into enterohepatic circulation, which then leads to an increase in gut bile acid secretion [10]. As a result, the liver responds by increasing hepatic conversion of cholesterol into bile acids, thus, reducing its circulating levels.

In diabetes, hypertriglyceridemia and hypercholesterolemia are associated with the consequences of hyperinsulinemia, insulin resistance, and glucose intolerance [1]. In the current study, both diabetic and hypercholesterolemic disease rats had significantly increased plasma TG concentration compared with normal nondisease rats. These elevated plasma TG concentrations were attenuated by oral ingestion of ABP. Although the activity of antihypercholesterolemia of ABP was observed in hypercholesterolemic rats, it was not the case in diabetic rats. The reason for the difference in antihypercholesterolemic activity of ABP between the 2 disease models is not known. The inconsistent effects of ABP may be because of the different severity of diseases associated with the 2 models. Because the level of hyperglycemic control is a determinant of TC [27], it is possible that the antihyperglycemic activity of ABP may not be sufficient to effect a significant reduction in TC in our diabetic rats.

Although our data are compatible with the thesis advanced in this study, they do not conclusively prove that WBM may exert antidiabetic and antihypercholesterolemic effects in a dietary setting where, for example, ABP is incorporated into a diet given ad libitum to rats. Corroboration of our data will be necessary and multiple additional studies need to be performed using animals other than rats that may not have features of cholesterol and glucose metabolism similar to those of humans in response to dietary fat manipulations and diabetes. The absence of an appropriate "placebo" group and the use of a single dosage form of ABP may introduce a possible bias and a type 1 error. Furthermore, the mechanism(s) of action by which ABP modulates glucose and cholesterol metabolism was not addressed. Although soluble dietary fiber is the most likely candidate, other constituents, such as antioxidants (polyphenols, vitamin C, and ergothioneine), proteins, and polysaccharides may play an important role. Further research is necessary to investigate the effects of these constituents. Nonetheless, despite the limitations, our study indicated that WBM could potentially be a health beneficial food for diabetic and hypercholesterolemic individuals.

To conclude, the data presented in this study support our primary hypothesis that WBM possesses antiglycemic and antihypercholesterolemic effects. Moreover, it has a positive influence on lipid metabolism and liver function. Based on studies of other mushrooms, the glucose- and cholesterol-lowering effects of WBM are likely the result of a number of mechanisms involving dietary fiber and other active components in the mushroom acting alone or in combination. Nonetheless, the marked metabolic effects seen in our animal models suggest a potential nutritional role of dietary WBM in the increasing rates of cardiovascular disease, obesity, and type 2 diabetes in humans. Nutritional studies are needed to examine the potential benefits of the compounds in mushrooms in carefully controlled clinical trials.

Acknowledgment

This work was supported, in part, by a grant from the Australian Mushroom Growers' Association and Horticulture Australia.

References

[1] Kaur J, Singh P, Sowers JR. Diabetes and cardiovascular diseases. Am J Ther 2002;9:510-5.
[2] Tourlouki E, Matalas AL, Panagiotakos DB. Dietary habits and cardiovascular disease risk in middle-aged and elderly populations: a review of evidence. Clin Interv Aging 2009;4:319-30.
[3] Adams MR, Golden DL, Chen H, Register TC, Gugger ET. A diet rich in green and yellow vegetables inhibits atherosclerosis in mice. J Nutr 2006;136:1886-9.
[4] Bazzano LA, Li TY, Joshipura KJ, Hu FB. Intake of fruit, vegetables, and fruit juices and risk of diabetes in women. Diabetes Care 2008;31:1311-7.
[5] Mirmiran P, Noori N, Zavareh MB, Azizi F. Fruit and vegetable consumption and risk factors for cardiovascular disease. Metabolism 2009;58:460-8.
[6] Yu L, Fernig DG, Smith JA, Milton JD, Rhodes JM. Reversible inhibition of proliferation of epithelial cell lines by *Agaricus bisporus* (edible mushroom) lectin. Cancer Res 1993;53:4627-32.

[7] Chen S, Oh S, Phung S, Hur G, Ye JJ, Kwok SL, et al. Anti-aromatase activity of phytochemicals in white button mushrooms (*Agaricus bisporus*). Cancer Res 2006;66:12026-34.
[8] Ren Z, Guo Z, Meydani SN, Wu D. White button mushroom enhances maturation of bone marrow-derived dendritic cells and their antigen-presenting function in mice. J Nutr 2008;138:544-50.
[9] Adams LS, Phung S, Wu X, Ki L, Chen S. White button mushroom (*Agaricus bisporus*) exhibits antiproliferative and proapoptotic properties and inhibits prostate tumor growth in athymic mice. Nutr Cancer 2008;60:744-56.
[10] Cheung PCK. The hypocholesterolemic effect of two edible mushrooms: *Auricularia auricula* (tree ear) and *Tremella fuciformis* (white jelly-leaf) in hypercholesterolemic rats. Nutr Res 1996;16:1721-5.
[11] Lo HC, Tsai FA, Wasser SP, Yang JG, Huang BM. Effects of ingested fruiting bodies, submerged culture biomass, and acidic polysaccharide glucuronoxylomannan of *Tremella mesenterica* Retz: Fr on glycemic responses in normal and diabetic rats. Life Sci 2005;78:1957-66.
[12] Fukushima M, Nakano M, Morii Y, Ohashi T, Fujiwara Y, Sonoyama K. Hepatic LDL receptor mRNA in rats is increased by dietary mushroom (*Agaricus bisporus*) fiber and sugar beet fiber. J Nutr 2000; 130:2151-6.
[13] Koyyalamudi SR, Jeong SC, Cho KY, Pang G. Vitamin B12 is the active corrinoid produced in cultivated white button mushrooms (*Agaricus bisporus*). J Agric Food Chem 2009;57:6327-33.
[14] Koyyalamudi SR, Jeong SC, Song CH, Cho KY, Pang G. Vitamin D2 formation and bioavailability from *Agaricus bisporus* button mushrooms treated with ultraviolet irradiation. J Agric Food Chem 2009;57: 3351-5.
[15] Mattila P, Könkö K, Eurola M, Pihlava JM, Astola J, Vahteristo L, et al. Contents of vitamins, mineral elements, and some phenolic compounds in cultivated mushrooms. J Agric Food Chem 2001;49: 2343-8.
[16] Mathe D. Dyslipidemia and diabetes: animal models. Diabete Metab 1995;21:106-11.
[17] Kuller LH. Nutrition, lipids and cardiovascular disease. Nutr Rev 2006;64:S15-26.
[18] Layman DK, Clifton P, Gannon MC, Gannon MC, Krauss RM, Nuttall FQ. Protein in optimal: heart disease and type 2 diabetes. Am J Clin Nutr 2008;87:1571-5.
[19] Yang BK, Jeong SC, Lee HJ, Sohn DH, Song CH. Antidiabetic and hypolipidemic effects of *Collybia confluens* mycelia produced by submerged culture in streptozotocin-diabetic rats. Arch Pharm Res 2006;29:73-9.
[20] Yang BK, Park JB, Song CH. Hypolipidemic effect of an exo-biopolymer profuced from a submerged mycelial culture of *Hericium erinaceus*. Biosci Biotechnol Biochem 2003;67:1292-8.
[21] Conniff P. Official methods of analysis. 16th ed. Arlington (Va): Association of Official Chemists; 1995.
[22] Kim DH, Yang BK, Jeong SC, Park JB, Cho SP, Das S, et al. Production of a hypoglycemic extracellular polysaccharide from the submerged culture of the mushroom, *Phellinus linteus*. Biotechnol Lett 2001;23:513-7.
[23] Friedeward WT, Levy RI, Fredrickson DS. Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. Clin Chem 1972;18:499-502.
[24] Hostmark AT, Berg JE, Osland A, Simonsen S, Vantne K. Lipoprotein-related coronary risk factor in patients with angiographically defined coronary artery disease and control: improved group separation by indexes reflecting the balance between low and high-density lipoprotein. Coron Artery Dis 1991;2:679-84.
[25] Kim DH, Sho KS. Experimental study of *Pinus densiflora* et zuccarini on hyperlipidemia and lipid in rats. K P I 2007;10:109-19.
[26] Folch J, Lees M, Sloane-Stanley GH. A simple method for the isolation and purification of total lipids from animal tissues. J Biol Chem 1957; 226:497-509.
[27] Gray AM, Flatt PR. Insulin-releasing and insulin-like activity of *Agaricus campestris* (mushroom). J Endocrinol 1998;157:259-66.
[28] Cheung PCK. Dietary fiber content and composition of some cultivated edible mushroom fruiting bodies and mycelia. J Agric Food Chem 1996;44:468-71.
[29] Giacco R, Parillo M, Rivellese AA, Lasorella G, Giacco A, D'Episcopo L, et al. Long-term dietary treatment with increased amounts of fiber-rich low-glycemic index natural foods improves blood glucose control and reduces the number of hypoglycemic events in type 1 diabetic patients. Diabetes Care 2000;23:1461-6.
[30] Platt PR, Bailey CJ. Role of dietary factors in the hyperinsulinemia of genetically\obese hyperglycaemic (ob/ob) mice. J Nutr 1982;112: 2212-6.
[31] Ahmad N, Bansal R, Rastogi AK, Kidwai JR. Effect of PHA-B fraction of *Agaricus bisporus* lectin on insulin release and $45Ca^{2+}$ uptake by islets of Langerhans in vitro. Acta Diabetol Lat 1984;21:63-70.
[32] Estruch R, Martínez-González MA, Corella D, Basora-Gallisá J, Ruiz-Gutiérrez V, Covas MI, Fiol M, et al, PREDIMED Study Investigators. Effects of dietary fibre intake on risk factors for cardiovascular disease in subjects at high risk. J Epidemiol Community Health 2009;63:582-8.
[33] Talpur NA, Echard BW, Fan AY, Jaffari O, Bagchi D, Preuss HG. Antihypertensive and metabolic effects of whole Maitake mushroom powder and its fractions in two rat strains. Mol Cell Biochem 2002; 237:129-36.
[34] Wong JMW, De Souza R, Kendall CW. Colonic health: fermentation and short chain fatty acids. J Clin Gastroenterol 2006;40:235-43.
[35] Todesco T, Rao AV, Bosello O. Propionate lowers blood glucose and alters lipid metabolism in healthy subjects. Am J Clin Nutr 1991;54: 860-5.
[36] Yamashita K, Kawai K, Itakura M. Effect of fructo-oligosaccharides on blood glucose and serum lipids in diabetic subjects. Nutr Res 1984; 4:961-6.
[37] Yang BK, Jeong YS, Song CH. Hypoglycemic effects of *Ganoderma applanata* and *Collybia confluens* exo-polymers in streptozotocin-induced diabetic rats. Phytother Res 2007;21:1066-9.
[38] Hwang HJ, Kim SW, Lim JM, Joo JH, Kim HO, Kim HM, et al. Hypoglycemic effect of crude exopolysaccharides produced by a medicinal mushroom *Phellinus baumii* in streptozotocin-induced diabetic rats. Life Sci 2005;76:3069-80.
[39] Aas AM, Ohrvik J, Malmberg K, Rydén L, Birkeland KI. Insulin-induced weight gain and cardiovascular events in patients with type 2 diabetes. A report from the DIGAMI 2 study. DIGAMI 2 investigators. Diabetes Obes Metab 2009;11:323-9.
[40] Feldstein AC, Nichols GA, Smith DH, Stevens VJ, Bachman K, Rosales AG, et al. Weight change in diabetes and glycemic and blood pressure control. Diabetes Care 2008;31:1960-5.
[41] Yao HT, Lin P, Chang YW, Chen CT, Chiang MT, Chang L, et al. Effect of taurine supplementation on cytochrome P450 2E1 and oxidative stress in the liver and kidneys of rats with streptozotocin-induced diabetes. Food Chem Toxicol 2009;47:1703-9.
[42] Doi K, Ishida K. Diabetes and hypertriglyceridemia modify the mode of acetaminophen-induced hepatotoxicity and nephrotoxicity in rats and mice. J Toxicol Sci 2009;34:1-11.
[43] Chen WJ, Anderson JW, Jennings D. Propionate may mediate the hypocholesterolemic effects of certain soluble plant fibers in cholesterol-fed rats. Proc Soc Exp Biol Med 1984;175:215-8.
[44] Hara H, Haga S, Kasai T. Fermentation products of sugarbeet fiber by cecal bacteria lower plasma cholesterol concentration in rats. J Nutr 1998;128:688-93.
[45] Ey J, Schömig E, Taubert D. Dietary sources and antioxidant effects of ergothioneine. J Agric Food Chem 2007;55:6466-74.
[46] Fernandez-Panchon MS, Villano D. Antioxidant activity of phenolic compounds: from in vitro results to in vivo evidence. Crit Rev Food Sci Nutr 2008;48:649-71.
[47] Thompson LU, Yoon JH, Jenkins DJA, Wolever TMS, Jenkins AL. Relationship between polyphenol intake and blood glucose response in normal and diabetic individuals. Am J Clin Nutr 1984;39:745-51.
[48] Hossain S, Hashimoto M, Choudhury EK, Alam N, Hussain S, Hasan M, et al. Dietary mushroom (*Pleurotus ostreatus*) ameliorates

atherogeneic lipid in hypercholesterolaemic rats. Clin Exp Pharmcol Physiol 2003;30:470-5.

[49] Cook SI, Sellin JH. Short chain fatty acids in health and disease. Aliment Pharmacol Ther 1998;12:499-507.

[50] Thacker PA, Salamons MO, Aherne FX. Influence of propionic acid on the cholesterol metabolism of pigs fed hypercholesterolemic diets. Can J Anim Sci 1981;61:969-75.

[51] Fukushima M, Ohashi T, Fujiwara Y, Sonoyama K, Nakano M. Cholesterol-lowering effects of maitake (*Grifola frondosa*) fiber, shiitake (*Lentinus edodes*) fiber, and enokitake (*Flammulina velutipes*) fiber in rats. Exp Biol Med (Maywood) 2001;226:758-65.

[52] Preuss HG, Echard B, Bagchi D, Perricone NV, Zhuang C. Enhanced insulin hypoglycemic activity in rats consuming a specific glycoprotein extracted from maitake mushroom. Mol Cell Biochem 2007;306:105-13.