

Does Extract of *Pleurotus sajor-caju* affect Liver Enzymes and Histological Integrity?**Nik Norliza, N. H., T. A. Tengku Farah Adilah, M. Siti Hajar, W. A. Wan Amir Nizam and W. I. Wan Rosli****School of Health Sciences, Universiti Sains Malaysia Health Campus, 16150 Kelantan, Malaysia***ABSTRACT**

Pleurotus sajor-caju (PSC) was believed to have both antihyperlipidemic and hepatoprotective activities. The present study aimed to investigate the effect of PSC on liver enzymes and histological integrity. This study used five groups of rats fed with ghee in the ratio 32g ghee per 68g pellet to induce hypercholesterolemia and one group was fed on cholesterol free basal diet. Rats treated with 100 mg/kg of PSC for a month was found to have an effect on the liver enzymes activities since plasma alkaline phosphatase (ALP) concentration in this group showed a significant reduction ($P < 0.05$) and a higher percentage reduction (66.01%) as compared to 20 mg/kg-PSC and 200 mg/kg-PSC treatment groups. The plasma aspartate aminotransferase (AST) and alanine aminotransferase (ALT) only showed a mild increased by 35.04% and 15.00% respectively in 100 mg/kg-PSC treatment group and there was no significant increased ($P > 0.05$) found in these both AST and ALT concentrations. Atorvastatin treatment also showed reduction in ALP enzymes but no significant reduction ($P > 0.05$) as compared to 100 mg/kg-PSC treatment group. On the other hand, plasma AST and ALT in 20 mg/kg of atorvastatin treatment were increased in percentage by 275.97% and 112.50% respectively indicated the adverse effects of statin in term of elevation of plasma enzymes activities. On the histological part, there was no significant finding in the micrograph study between treatment and hypercholesterolemic (HPC) liver rat. The micrograph of rat liver treated with 100 mg/kg PSC showed smooth and clear surface of hepatocytes compared to HPC group.

Key words: antihyperlipidemic, hepatoprotective, *Pleurotus sajor-caju*, plasma enzymes

INTRODUCTION

Edible mushrooms have been world widely cultivated mainly for food, health and medicinal purposes. These fungi are commonly cultivated on decayed organic material while producing edible portion on the surface of the substrate. *Pleurotus sajor-caju* (PSC), also known as oyster mushrooms, is easily cultivated and is done so widely in the tropical regions and many other parts of the world. It is reported to possess distinctive aroma and highly palatable.

*rosliishak@gmail.com, wrosli@kck.usm.my

In South East Asian countries, oyster mushroom is widely used in the preparation of vegetable, soup, stew and other culinary products. This edible fungus is suggested to possess considerable importance in the human diet as its rich in non-starchy carbohydrates, dietary fibre, β -glucans, minerals, vitamin B while having low in fat. β -glucans, also a components of soluble or insoluble dietary fibre (SDF, IDF) is present in appreciable amounts in mushrooms and are linked to the ability to lower blood cholesterol levels and glycaemic response *in vivo* (Manzi et al., 2004), hypercholesterolemic properties and other therapeutic functions. The present study was conducted to investigate whether *Pleurotus sajor-caju* extract affect liver enzymes and histological integrity.

METHODS AND MATERIALS

Pleurotus sajor-caju

Oyster mushroom (*Pleurotus sajor-caju*, PSC) was supplied by Anjaad Industries Sdn Bhd (Serkam, Malacca State of Malaysia). PSC mushroom was already in the ground dried form. Aqueous extract of PSC was extracted using Soxhlet apparatus.

Animals

The procedure and protocol described below were approved by Animal Ethics Committee USM (USM/Animal Ethics Approval/ 2013/ (83) (446)). Eighteen male Wistar rats weighing 250-370 g were obtained from Animal Research and Service Centre of Universiti Sains Malaysia (ARASC). For adaptation, the rats were fed with a basal diet for one week before treatment begins. They were housed in rat cages at room temperature of $22 \pm 1^\circ\text{C}$ under a 12-h light-dark cycle. Rats were divided randomly into six feed groups (3 rats/group): control group was fed a basal diet; while the remaining five groups were given 32 g ghee per 68 g pellet (high-cholesterol diet). Rats were fed for two weeks and were allowed free access to water. After two weeks of high-cholesterol diet, blood was withdrawn from the rats through saphenous vein. Blood samples were then sent to BP Laboratory Pte Ltd for total cholesterol levels determination. Plasma total cholesterol was determined using automated Abbott Ci8200 Biochemistry analyzer based on enzymatic kits methods. Rat with serum cholesterol level > 2.3 mmol/L was considered hypercholesterolemic and included in this study. Group I was served as normal control (NC) rats, whereas Group II was left as a hypercholesterolemic (HPC) control rats. Meanwhile Group III,

IV, V and VI were treated with 20 mg/kg PSC, 100 mg/kg PSC, 200 mg/kg PSC and 20 mg/kg atorvastatin respectively.

Plasma enzymes analysis

At the end of experiment, the rats were anaesthetized with 6% pentobarbital and blood was collected through cardiac puncture and sent to the BP Laboratory for liver function tests (LFTs) analysis. Plasma liver enzymes were determined using automated Abbott Ci8200 Biochemistry analyzer based on photometric IFCC (International Federation of Chemistry) methods.

Histological analysis of liver by using scanning electron microscope (SEM)

Liver tissues were rapidly dissected, removed and fixed in McDowell-Trump Fixative at 4°C for 24 hours. The fixed tissues were then cut, washed, post-fixed and dehydrated. The sample was then proceed with critical point drying step, coated with gold and lastly viewed under Quanta FEG 450 Scanning Electron Microscope (SEM) by using XTm Product Version 4.1.7.2095 viewer software.

Statistical analysis

All values used in analysis are presented as means \pm SEM. Comparisons among the different groups were performed by one-way analysis of variance (ANOVA), followed by Bonferroni multiple comparisons test and differences were considered significant when $P < 0.05$.

RESULTS AND DISCUSSION

Effects of PSC on plasma liver enzyme profile

Table 1 summarizes the values for all liver enzyme analysis before and after treatments. There were no significant differences found in total protein, albumin, globulin, and albumin/globulin (A/G) ratio for all experimental groups. AST level in 20mg/kg-PSC, 200mg/kg-PSC and HPC group was increased in percentage changes by 127.50%, 85.39% and 147.22% respectively. Like AST, there was an increased in ALT by 65.79%, 23.40% and 53.49% respectively for 20mg/kg-PSC, 200 mg/kg-PSC treatment and HPC group of rats. The least amount of increase in AST and ALT were found in 100 mg/kg-PSC treatment group (35.04% and 15.00% respectively). In contrast, the highest increased in AST and ALT were found in atorvastatin treatment group

(275.97% and 112.5% respectively). ALP level in 20mg/kg-PSC, 200mg/kg-PSC and 20mg/kg-atorvastatin treatment group was increased in percentage changes by 49.40%, 52.95% and 54.97% respectively. A significant reduction by 66.47% and 66.01% was found in HPC and 100mg/kg-PSC treatment groups respectively. There were no changes observed before and after treatment in both total bilirubin and GGT enzymes.

Lower plasma ALP concentration in hypercholesterolemic rats was assessed to significantly ($P < 0.05$) reduced by the supplementation of diet with 100 mg/kg PSC (Figure 1). Treatment with 20 mg/kg and 200 mg/kg of PSC were not significantly ($P > 0.05$) differed in their impact on ALP enzyme. Thus, by comparing to other treatment groups, dose of 100 mg/kg of PSC was the effective dose of PSC to lower the plasma ALP. However, this finding was not significant as HPC group was also found to have significantly reduction in plasma ALP ($P > 0.05$). The reason for such result may be due to the modulation of mechanism or physiological functions in hypercholesterolemic rats which cannot be controlled. ALP is a marker enzyme of the plasma membrane and can be found mainly in liver and bone origin. The underlying mechanism for reduction of plasma ALP enzymes by mushroom is not clearly understood. However, study by Mishra and Singh (2010), had comes out with a finding that mushroom (*Pleurotus* sp.) restores the changes in ALP, AST and ALT activities due to antioxidant effects and their ability as a radical scavenger, thus, protecting membrane permeability.

Plasma AST and ALT was found to be elevated in all groups. However, there was no significant increased ($P > 0.05$) found for both enzymes (Figure 2 and 3). Increased in plasma AST and ALT was more prominent in 20mg/kg PSC-treatment, atorvastatin treatment and hypercholesterolemic control groups. From the result, we found that 20mg/kg of PSC-treatment was the least effective treatment compared to the other two PSC-treatment groups. On the other hand, plasma AST and ALT concentrations for 100mg/kg PSC treatment was increased by 35.04% and 15.00% respectively which is the smallest percentage of increased in concentration as compared to HPC and other PSC-treatment groups.

In this study, atorvastatin treatment served as positive control group, thus, the results should be almost similar with PSC treatment group. However, by comparing with HPC and PSC treatment groups, we found another finding in which AST and ALT levels in 20 mg/kg atorvastatin treatment group showed a significant increased by 275.97% and 112.50% respectively. Thus, we can postulate that increased in these two liver enzyme parameters may be due to statin adverse effects. There is a correlation exists between statin treatment and hepatic adverse effects. However, the underlying mechanism of this effect is still not clearly understood. Previous study had postulated that statin causes changes in the lipid components of the hepatocyte membrane and this will leads to an increase in its permeability and result in an increased of liver enzymes (Rossana *et al.*, 2010).

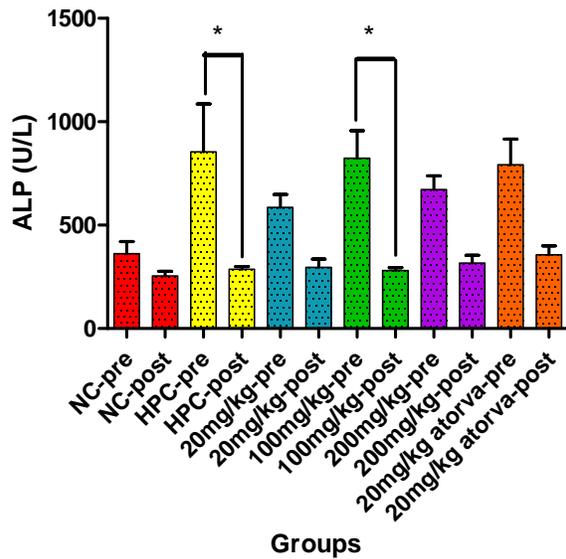
Figure 3 - 7 showed no significant different ($P > 0.05$) in plasma total protein, albumin, globulin and A/G ratio respectively for all treatments. There were also no changes in concentration of pre- and post-study for total bilirubin and GGT concentration (Table 1). In addition, there were also no significant differences of total protein within groups ($P > 0.05$) in both pre and post-treatment of PSC and atorvastatin on total protein level of HPC rats (Figure 4). On the other part, the effect of pre and post-treatment of PSC and atorvastatin on plasma albumin of HPC rats resulted reduction in albumin values for all groups, however there were no significant decreased of albumin (Figure 5) within groups ($P > 0.05$). In addition, globulin level was decreased for 20 mg/kg and 100 mg/kg-PSC treatment groups, but increased for other groups. However, there were no significant differences of globulin levels (Figure 6) within groups ($P > 0.05$). In addition, albumin/globulin ratio (A/G) of HPC rats' ratio was reduced in normal, HPC, 200 mg/kg-PSC and 20 mg/kg atorvastatin treatment groups, whereas other groups showed no changes in values (Figure 7). There were no significant differences of A/G ratio within groups ($P > 0.05$).

Table 1 Effect of *Pleurotus sajor-caju* on liver enzymes of hypercholesterolemic rats

Group	Treatment	TP (g/L)	Alb (g/L)	Glo (g/L)	A/G	TB (μ mol/L)	SGOT/AST (U/L)	SGPT/ALT (U/L)	ALP (U/L)	GGT (U/L)
I (Normal control, NC)	Before	61.5 \pm 1.5	24.0 \pm 2.0	37.5 \pm 0.5	0.70 \pm 0.05	1.7	129.0 \pm 7.0	40.0 \pm 1.5	362.0 \pm 58.0	4
	After	61.5 \pm 2.5	23.0 \pm 0.5	39.0 \pm 2.0	0.60 \pm 0.0	1.7	215.0 \pm 80.0	61.0 \pm 15.5	254.0 \pm 23.0	4
	% changes	-	-4.17	+4.00	-14.29	-	+66.67	+52.50	-29.83	-
II (HPC)	Before	60.0 \pm 1.5	24 \pm 1.0	35.5 \pm 0.5	0.70 \pm 0.05	1.7	126.0 \pm 17.0	43.0 \pm 2.0	854.5 \pm 23.5	4
	After	57.5 \pm 0.7	22 \pm 0.5	36.0 \pm 1.0	0.6 \pm 0.0	1.7	311.5 \pm 169.5	66 \pm 18.5	286.5 \pm 12.5	4
	% changes	-4.17	-8.33	+1.41	-14.29	-	+147.22	+53.49	-66.47*	-
III (20 mg/kg- PSC)	Before	62.0 \pm 2.0	25 \pm 0.9	36.7 \pm 1.5	0.7 \pm 0.03	1.7	120.0 \pm 7.4	38 \pm 1.8	585.0 \pm 62.6	4
	After	58.3 \pm 2.3	23.0 \pm 1.2	35.3 \pm 1.5	0.7 \pm 0.03	1.7	273.0 \pm 65.2	63.0 \pm 10.2	296.0 \pm 39.5	4
	% changes	-5.97	-8.00	-3.81	-	-	+127.50	+65.79	-49.40	-
IV (100 mg/kg- PSC)	Before	62.0 \pm 0.3	25 \pm 0.9	37.0 \pm 1.0	0.70 \pm 0.06	1.7	122.7 \pm 11.2	40.0 \pm 2.3	823.7 \pm 133.4	4
	After	58.3 \pm 1.2	24.0 \pm 0.0	34.3 \pm 1.2	0.7 \pm 0.03	1.7	165.7 \pm 38.1	46.0 \pm 4.4	280.0 \pm 14.1	4
	% changes	-5.97	-4.00	-7.30	-	-	+35.04	+15.00	-66.01*	-
V (200 mg/kg- PSC)	Before	58.0 \pm 3.2	24.0 \pm 1.0	34.0 \pm 2.3	0.8 \pm 0.1	1.7	130.7 \pm 22.8	47.0 \pm 7.0	672.3 \pm 65.9	4
	After	61.0 \pm 1.2	24.0 \pm 0.6	37.0 \pm 1.0	0.6 \pm 0.03	1.7	242.3 \pm 84.4	58.0 \pm 13.3	316.3 \pm 37.9	4
	% changes	+5.17	-	+8.82	-25.00	-	+85.39	+23.40	-52.95	-
VI (20 mg/kg- Atorvastatin)	Before	63.7 \pm 0.9	25.0 \pm 0.6	38.7 \pm 0.7	0.5 \pm 0.1	1.7	115.7 \pm 6.2	40.0 \pm 1.8	791.3 \pm 124.7	4
	After	60.7 \pm 0.7	22.0 \pm 0.9	39.0 \pm 0.6	0.6 \pm 0.03	1.7	435.0 \pm 243.4	85.0 \pm 30.8	356.3 \pm 43.2	4
	% changes	-4.71	-12.00	+0.78	+20.00	-	+275.97	+112.50	-54.97	-

Values are expressed as means \pm SEM for each group; * indicates significantly different at $P < 0.05$. TP: total protein (g/L); Alb: albumin (g/L); Glo: Globulin (g/L); A/G: albumin/globulin ratio; TB: total bilirubin (μ mol/L); SGOT/AST: serum glutamic oxaloacetic transaminase/aspartate transaminase (U/L); SGPT/ALT: serum glutamic pyruvate transaminase/alanine aminotransferase (U/L); ALP: alkaline phosphatase (U/L); GGT: gamma-glutamyl transpeptidase (U/L).

Graph of ALP values pre- vs. post-treatment groups



Graph of AST values pre- vs. post-treatment groups

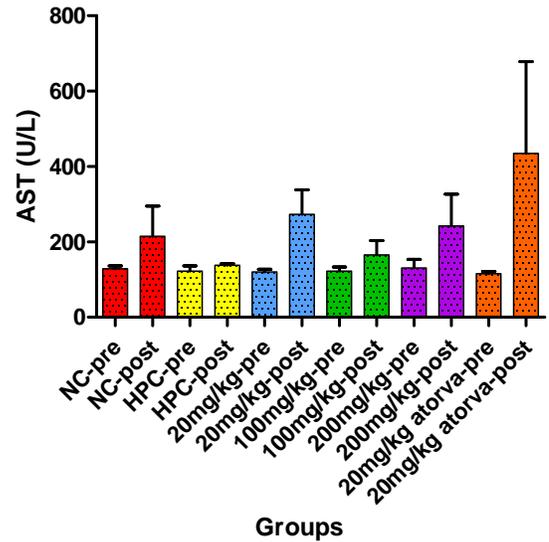


Fig 1 Effect of pre- and post-treatment of PSC and atorvastatin on plasma alkaline phosphatase (ALP) of HPC rats. Values are in means \pm SEM.

*indicates significantly different at $P < 0.05$.

NC = normal control, HPC = hypercholesterolemic

Fig 2 Effect of pre and post-treatment of PSC and atorvastatin on plasma aspartate aminotransferase (AST) of HPC rats. Values are in means \pm SEM.

NC = normal control, HPC = hypercholesterolemic

Graph of ALT values pre- vs. post-treatment groups

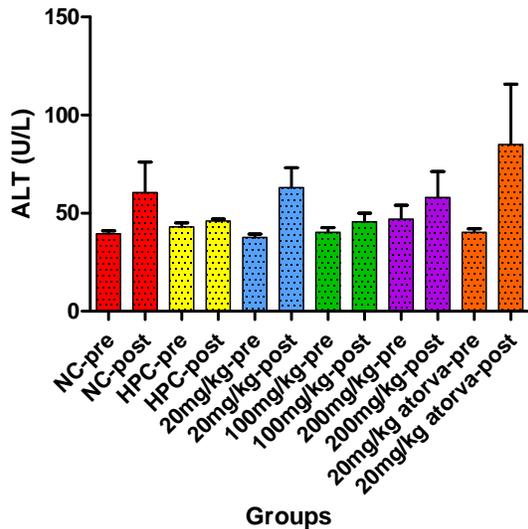


Fig 3 Effect of pre and post-treatment of PSC and atorvastatin on plasma alanine aminotransferase (ALT) of HPC rats. Values are in means \pm SEM.

NC = normal control, HPC = hypercholesterolemic

Graph of total protein pre- vs. post-treatment groups

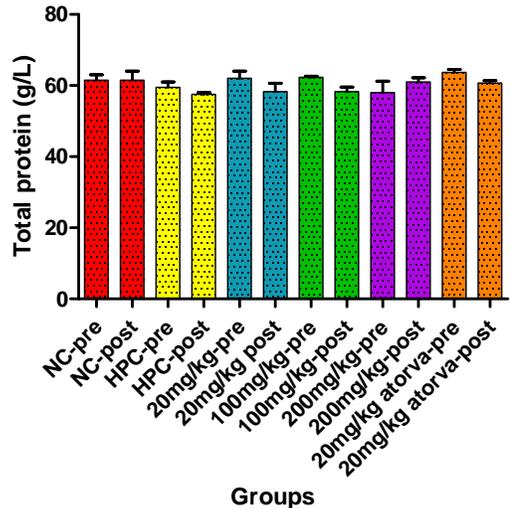


Fig 4 Effect of pre and post-treatment of PSC and atorvastatin on total protein level of HPC rats. Values are in means \pm SEM.

NC = normal control, HPC = hypercholesterolemic

Graph of albumin pre- vs. post-treatment groups

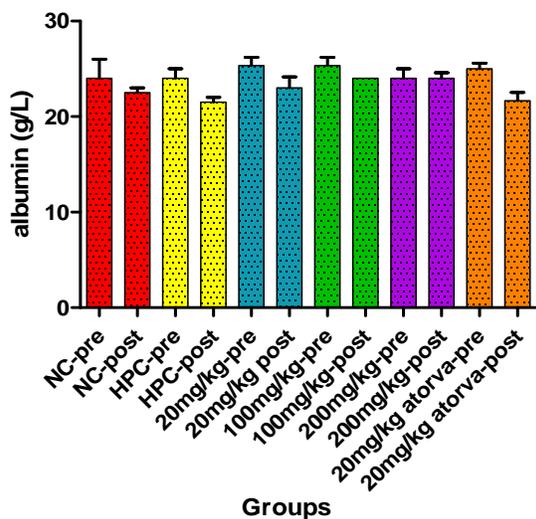


Fig 5 Effect of pre and post-treatment of PSC and atorvastatin on plasma albumin of HPC rats. Values are in means \pm SEM. NC = normal control, HPC = hypercholesterolemic

Graph of globulin pre- vs. post-treatment groups

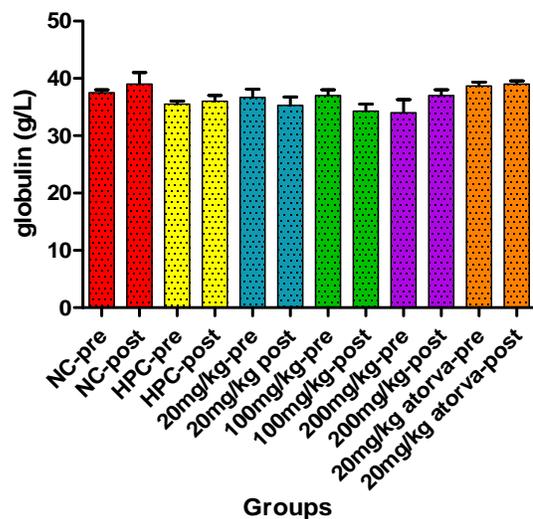


Fig 6 Effect of pre and post-treatment of PSC and atorvastatin on globulin level of HPC rats. Values are in means \pm SEM. NC = normal control, HPC = hypercholesterolemic

Graph of A/G ratio pre- vs. post-treatment groups

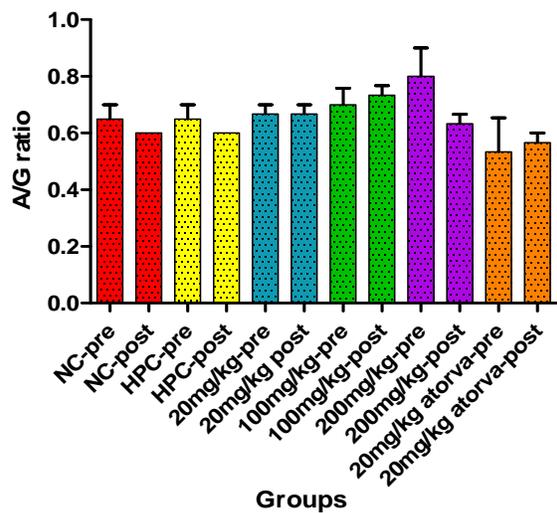


Fig 7 Effect of pre and post-treatment of PSC and atorvastatin on albumin/globulin ratio (A/G) of HPC rats. Values are in means \pm SEM. NC = normal control, HPC = hypercholesterolemic

Histological study

Scanning electron microscope (SEM) indicated three-dimensional features of intrahepatic structures in liver rat especially the surface characteristics of hepatocytes and sinusoidal endothelial cells. At 4000 x magnification, HPC rat liver treated with 100 mg/kg-PSC (Figure 8a) showed smooth surface of hepatocytes with bile canaliculi as compared to HPC rat (Figure 8c). Hepatocytes seen were in polyhedral and sharply angulated shape. However, the surface seen was not so clear due to the presence of some artifact. Figure 8b showed micrograph of HPC rat liver treated with atorvastatin in which the morphological features of liver surface resembled PSC-treatment (Figure 8a) to some extent. However, the surface of hepatocytes seen in atorvastatin treatment was not as smooth as PSC-treatment. This may be due to the incorrectly apply technique in the process of cutting the liver.

In addition, at 12 000 X magnification, rat liver after 100 mg/kg PSC treatment and atorvastatin treatment demonstrated smooth surface of hepatocytes as compared to HPC control rat (Figure 9a-9d). Round structure with whitish color may be assumed as fat storing cells or fatty infiltration but were not clearly seen. Normal control rat showed these whitish round structures but not as prominent as HPC rat. At higher magnification (30 000 X), round structure with whitish color may be assumed as fat storing cells or fatty infiltration but are not clearly seen (Figure 10a-10d). Normal control rat (Figure 10d) showed these whitish round structures measuring about 0.8 μm but not as prominent as HPC (Figure 10c) rat. A small amount of fats storing cells or fatty infiltration may present in 100 mg/kg and atorvastatin treatment rat liver but are not clearly seen and not prominent in HPC rat.

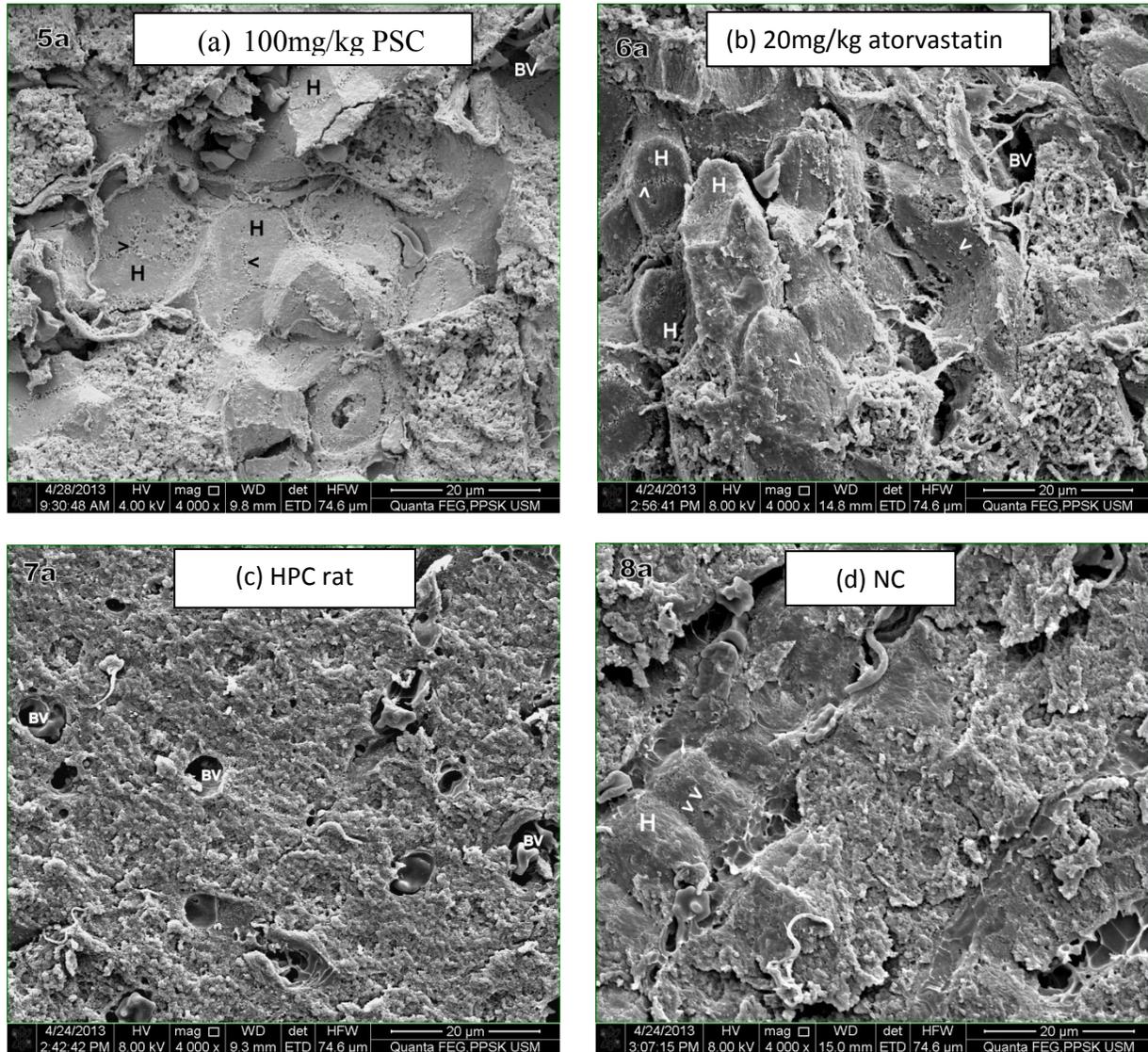


Fig 8 (a): Micrograph of rat liver after 100 mg/kg PSC treatment; **(b):** atorvastatin treatment; **(c):** HPC control; **(d):** normal control. (H: hepatocytes; arrowhead: bile canaliculi; BV: blood vessel) (4 000 x magnification)

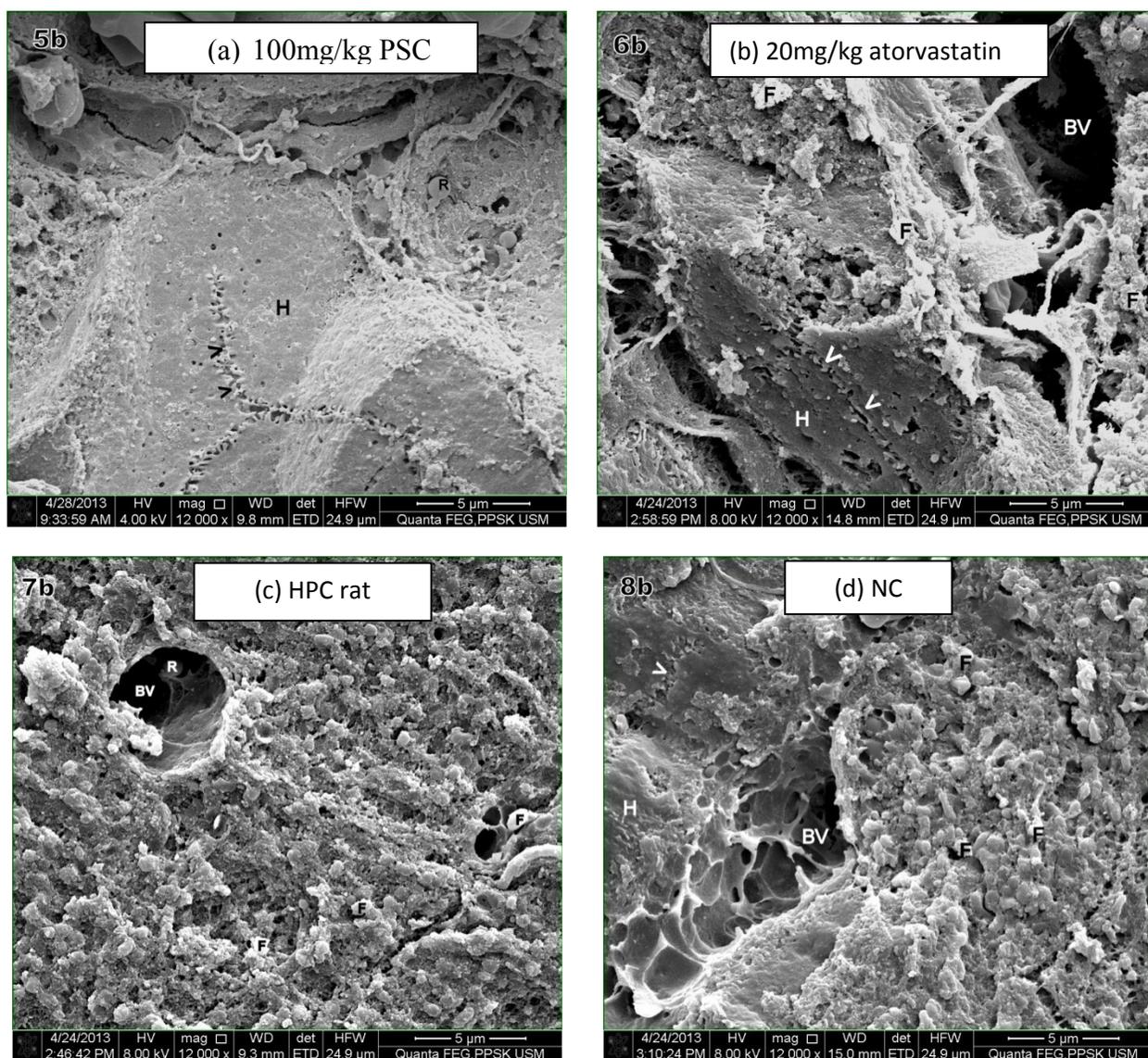


Fig 9 (a): Micrograph of rat liver after 100 mg/kg PSC treatment; **(b):** atorvastatin treatment; **(c):** HPC control; **(d):** normal control. (H: hepatocytes; arrowhead: bile canaliculi; BV: blood vessel; R: red blood cell) (12 000 x magnification)

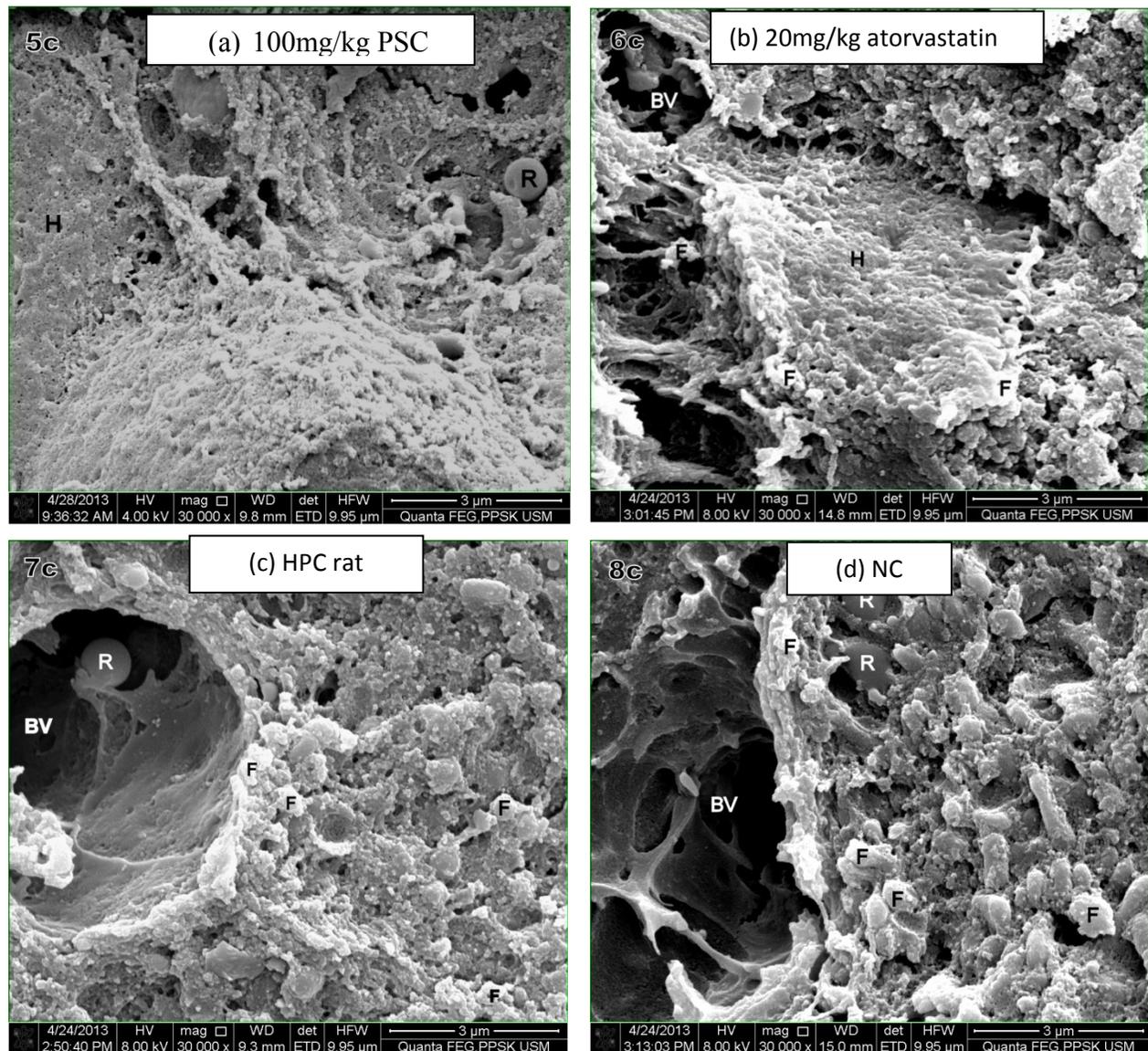


Fig 10 (a): Micrograph of rat liver after 100 mg/kg PSC treatment; **(b):** atorvastatin treatment; **(c):** HPC control; **(d):** normal control. (H: hepatocytes; arrowhead: bile canaliculi; BV: blood vessel; R: red blood cell) (30 000 x magnification)

CONCLUSION

This preliminary study demonstrated that feeding 100 mg/kg of PSC significantly reduced plasma ALP concentration. However, this finding was not significant as HPC group also showed significant reduction in ALP enzymes. Atorvastatin drug was proven to have clinically significant adverse effects as an elevated ALT and AST enzymes were prominent in 20 mg/kg atorvastatin-treatment group as compared to PSC-treatment groups. However, there was no

significant finding on the micrograph study on the liver of HPC and PSC-treatment rat. The only different in this histological study is the surface of hepatocyte in PSC- and atorvastatin treatment rat was smooth as compared to HPC rat. For future work, it is suggested to used normal light microscopic staining (Haematoxylin and Eosin, H&E and Oil-Red staining) or TEM to study for any fatty infiltration and fatty changes in HPC and treatment rats.

ACKNOWLEDGEMENTS

The study was funded by Universiti Sains Malaysia (1001/PPSK/813057). Special thanks to all staffs who assist this preliminary investigation (Roslina, Shazwan) including Mr Fakurudin for his technical assistance and support when viewed samples using SEM.

REFERENCES

- Adams, L. A., Angulo, P. and Lindor, K. D. (2005) Nonalcoholic fatty liver disease. *Canadian Medical Association Journal*, **172**(7), 899-905.
- Agrawal, R., Chopra, A., Lavekar, G., Padhi, M., Srikanth, N. and Ota, S. (2010) Effect of oyster mushroom on glycemia, lipid profile and quality of life in type 2 diabetic patients. *Australian J Med Herbalism* **22**, 50-54.
- Alam, N., Amin, R., Khan, A., Ara, I., Shim, M. J. and Lee, M. W. (2009) Comparative Effects of Oyster Mushrooms on Lipid Profile, Liver, Kidney Function in Hypercholesterolemic Rats. *The Korean Society of Mycology* **1**, 37-42.
- Alam, N., Amin, R., Khan, A., Ara, I., Shim, M. J. and Lee, M. W. (2009) Comparative effects of oyster mushrooms on lipid profile, liver and kidney function in hypercholesterolemic rats. *Mycobiology* **37**(1), 37-42.
- Alam, N., Yoon, K. N., Lee, J. S., Cho, H. J., Shim, M. J. and Lee, T. S. (2011) Dietary effect of *Pleurotus eryngii* on biochemical function and histology in hypercholesterolemic rats. *Saudi Journal of Biological Sciences* **18**(4), 403-409.
- Alam, N., Yoon, K. N. and Lee, T. S. (2011) Antihyperlipidemic activities of *Pleurotus ferulae* on biochemical and histological function in hypercholesterolemic rats. *Journal of research in medical sciences: the official journal of Isfahan University of Medical Sciences* **16**(6), 776.
- Alam, N., Yoon, K. N., Lee, J. S., Lee, M. W. and Lee, T. S. (2011) Evaluation of Biochemical and Histological Effectiveness of *Pleurotus Citrinopileatus* on Plasma, Feces and Liver in Hypercholesterolemic Rats. *Advances in Environmental Biology* **5**(6), 1095-1103.
- Alarcon, J., Aguila, S., Arancibia-Avila, P., Fuentes, O., Zamorano-Ponce, E. and Hernandez, M. (2003) Production and purification of statins from *Pleurotus ostreatus* (Basidiomycetes) strains. *Zeitschrift Fur Naturforschung C*, **58**(1/2), 62-64.
- Alberts, A., Chen, J., Kuron, G., Hunt, V., Huff, J. and Hoffman, C. (1980) Mevinolin: a highly potent competitive inhibitor of hydroxymethylglutaryl-coenzyme A reductase and a cholesterol-lowering agent. *Proceedings of the National Academy of Sciences* **77**(7), 3957-3961.
- Altunkaynak, B. Z. and Ozbek, E. (2009) Overweight and structural alterations of the liver in female rats fed a high-fat diet: a stereological and histological study. *Turk J Gastroenterol* **20**(2), 93-103.
- Austin, M. A., Hutter, C. M., Zimmern, R. L. and Humphries, S. E. (2004) Familial hypercholesterolemia and coronary heart disease: a HuGE association review. *American journal of epidemiology* **160**(5), 421-429.

- Beltowski, J., Wojcicka, G. and Jamroz-Wisniewska, A. (2009) Adverse effects of statins - mechanisms and consequences. *Curr Drug Saf* **4**(3), 209-228.
- Bobek, P., Hromadová, M. and Ozdin, L. (1995) Oyster mushroom (*Pleurotus ostreatus*) reduces the activity of 3-hydroxy-3-methylglutaryl CoA reductase in rat liver microsomes. *Experientia* **51**(6), 589-591.
- Bobek, P., Ozdín, L. and Galbavý, Š. (1998). Dose-and Time-Dependent Hypocholesterolemic Effect of Oyster Mushroom (*Pleurotus ostreatus*) in Rats. *Nutrition* **14**(3), 282-286.
- Bobek, P., Ozdin, L. and Kuniak, L. (1994) Mechanism of hypocholesterolemic effect of oyster mushroom (*Pleurotus ostreatus*) in rats: reduction of cholesterol absorption and increase of plasma cholesterol removal. *Zeitschrift Für Ernährungswissenschaft* **33**(1), 44-50.
- Calderon, R. M., Cubeddu, L. X., Goldberg, R. B. and Schiff, E. R. (2010) Statins in the treatment of dyslipidemia in the presence of elevated liver aminotransferase levels: a therapeutic dilemma. *Mayo Clin Proc* **85**(4), 349-356.
- Calderon, R. M., Cubeddu, L. X., Goldberg, R. B. and Schiff, E. R. (2010) *Statins in the treatment of dyslipidemia in the presence of elevated liver aminotransferase levels: a therapeutic dilemma*. Paper presented at the Mayo Clinic Proceedings.
- Chang, S. and Miles, P. (1989) The nutritional attributes and medicinal value of edible mushrooms (pp. 27-40): CRC Press, Boca Raton, FL.
- Chang, S. T. (1992) Mushroom biology-A new discipline. *The Mycologist* **6**, 64-65.
- Chorváthová, V., Bobek, P., Ginter, E. and Klvanová, J. (1993) Effect of the oyster fungus on glycaemia and cholesterolaemia in rats with insulin-dependent diabetes. *Physiological research/Academia Scientiarum Bohemoslovaca* **42**(3), 175
- Jayakumar, T., Ramesh, E. and Geraldine, P. (2006) Antioxidant activity of the oyster mushroom *Pleurotus ostreatus* induced liver injury in rats. *Food and Chemical Toxicology* **44**(12), 1989-1996.
- Khan, M. A., Rahman, M. M., Tania, M., Uddin, M. N. and Ahmed, S. (2011) *Pleurotus sajor-caju* and *Pleurotus florida* Mushrooms Improve Some Extent of the Antioxidant Systems in the Liver of Hypercholesterolemic Rats. *The Open Nutraceuticals J* **4**, 20-24.
- Khan, M. A. and Tania, M. (2012) Nutritional and medicinal importance of pleurotus mushrooms: an overview. *Food Reviews International* **28**(3), 313-329.
- Magosso, E., Ansari, M. A., Gopalan, Y., Abu Bakar, M., Karim Khan, N. and Wong, J. (2010). Prevalence of non-alcoholic fatty liver in a hypercholesterolemic population of northwestern peninsular Malaysia. *Southeast Asian Journal of Tropical Medicine and Public Health* **41**(4), 936.
- Manzi, P., Marconi, S., Aguzzi, A. and Pizzoferrato, L. (2004) Commercial mushrooms: nutritional quality and effect of cooking. *Food Chemistry* **84**, 201-206.
- Mishra, S. and Singh, R. B. (2010) Effect of Mushroom on the Lipid Profile, Lipid Peroxidation and Liver Functions of Aging Swiss Albino Rats. *The Open Nutraceuticals Journal* **3**, 248-253.
- Mohd Adzim Khalili, R., Norhayati, A. H., Rokiah, M. Y., Asmah, R., Siti Muskinah, M. and Abdul Manaf, A. (2009) Hypocholesterolemic effect of red pitaya (*Hylocereus sp.*) on hypercholesterolemia induced rats. *International Food Research Journal* **16**, 431-440.
- Sumy, A. K., Jahan, N. and Sultana, N. (2010) Study on the Hepatoprotective effect of oyster mushroom (*Pleurotus florida*) against paracetamol induced liver damage in Wistar albino rats. *Journal of Bangladesh Society of Physiologist* **5**(2), 46-52.