

## The Effect of Spent Mushroom Substrate on Blood Metabolites and Weight Gain in Kurdish Male Lambs

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### ABSTRACT

The objective of this study was use different levels of spent mushroom substrate as a suitable substitute for wheat straw in the ration of male lambs. In this study 20 male lambs with the age of 90 days and initial average weight of  $33 \pm 1.7$  kg were used. The animals were divided separately into single boxes with four treatments (control treatment, spent mushroom substrate 15%, spent mushroom substrate 25% and spent mushroom substrate 35%) and five replications. The experiment period was 114 days being 14 days adaptation and 90 days for breeding. On the days 36 and 94, blood samples were taken from the jugular vein. In order to carry out the trial, 20 male lambs received the four experimental diets in completely randomized design. The statistical analyses were carried out by using the GLM procedure of SAS 9.1. Means among treatments were compared by Tukey test. The results of the study showed that there was no significant differences between the serum biochemical and hematological contents of the lambs in the four treatments ( $p > 0.05$ ). A significant difference was observed between the spent mushroom substrate 25% and other treatments in average daily gain and final weight of the Kurdish lambs ( $p < 0.05$ ). It was concluded that spent mushroom substrate consumption has no harmful effect on the blood parameters of Kurdish male lambs. It is recommended that 25% of wheat straw could be replaced by spent mushroom substrate.

**Key words:** Alternative food, Nutrition, Sheep performance.

### INTRODUCTION

In 2012 the total production of mushroom in Iran was 57,933 tons from which 983 tons went to waste. The waste is mainly used for soil amelioration [2] and soil conditioner [26], but Collecting and removing the remains after each production cycle, regardless of expenses, can cause environmental problems because the nitrate existing in the waste leaches into the ground water and causes water pollution [16]. A portion of the remains containing compounds of cellulose such as straw may be used as food for ruminants [10, 27] because the waste contains a remarkable level of silicon and iron as well as a fairly high level of calcium, potassium, phosphorus, trace elements and nitrogen [17, 6]. Park et al [25], reported that a growing mushroom bed contains wheat straw, wheat bran, rice straw, beet pulp, saw dust, bean curd, and as such can be considered a food resource beneficial to ruminants. In the process of making compost, a relatively broad spectrum of hydrolyzing enzymes is produced by thermophile, a microorganism that in turn destroys the carbohydrates-structural in the straw [5]. Therefore, the remaining straw from the mushroom harvest is different from the initial straw in connection with organic compounds and minerals. The biggest change in the compost straw is that while its crude protein increases a decrease in its organic material is observable [27]. Most of the researchers inform us of the effects of fiber from SMS on ruminant diets [1, 10, 15, 23], but little information has been reported about the use of SMS on hematological and biochemical blood properties in lambs. The objective of the study is to probe the use of SMS on the diet and weight of Kurdish male lamb as well as its effect on hematological and biochemical blood properties.

## MATERIAS AND METHODS

**Animals and sample preparation**

Twenty Kurdish male lambs with the age between two and three months and average weight of ( $33 \pm 1.7$  Kg), distributed in four treatments (control, 15% of spent mushroom substrate (SMS), 25% of SMS and 35% of SMS with five replications were utilized in this study, performed in Mashhad, Iran. The lambs in each treatment were located in a confined area. Food and water were provided ad libitum during 114-d study period (14-d adaptation and 100-d of evaluation). On the days 36 and 94, blood samples were taken from the jugular vein.

**The ingredients of feed intake and the Chemical Compounds of feed**

The mushroom compost ingredients were as follows:

Wheat straw 58%, hen manure 34%, urea 1%, beet molasses 1.5%, gypsum 4% and mushroom spawn (*Pleurotus osteratus*) 1.5%.

When mushroom preserving period was over, the wastes were taken out from the production unit and the top layer of soil was removed and dried under the sun. The samples from the compost and initial wheat straw were taken. The chemical compounds of the samples were presented in table 1. The formula and chemical composition of the experimental feed can be noticed in the table 2.

**Table1: Chemical composition of initial wheat straw and spent mushroom substrate (SMS)**

Content	Initial Wheat Straw	Spent Mushroom Substrate (SMS)
Dry Matter (%)	96.40	83.90
Ash (%)	10	37.04
Crude Protein (%)	4.20	12.80
Calcium (%)	0.40	5
Phosphorus (%)	0.04	1.30
Sodium (%)	1.49	2.40
Crude Fiber (%)	44	6.30
Neutral detergent fiber (%)	85	28.20
Acid detergent fiber (%)	50	9.70
Energy (kcal/kg)	1864	2069

**Hematological and Biochemical Analysis**

Blood samples were analyzed for hematocrit (HCT), hemoglobin (Hb), mean corpuscular volume (MCV) and mean corpuscular hemoglobin (MCH) using an equipment Mindray model BC-5800 (Mindray Co., China). This same instrument was used to evaluate the contents of neutrophils (NE), lymphocytes (LY), monocytes (MO), eosinophils (EO) and basophils (BA).

The contents of blood urea nitrogen (BUN), aspartate aminotransferase (AST), aminotransferase (ALT), cholesterol (Ch), high density lipoprotein (HDL), low density lipoprotein (LDL), triglyceride (TG), calcium (Ca), phosphorus (P) and iron (Ir) were measured by Motic Spectrophotometer (Motic Co., China).

**Table 2: Chemical compounds (%) and ingredients of the experimental feeds**

Ingredients	Control	SMS 15%	SMS 25%	SMS 35%
Alfalfa	45	45	45	45
Wheat straw	13.90	11.80	10.50	9.10
Barley	27	27	27	27
Soybean meal	7	7	7	7
Wheat bran	6	6	6	6
Calcium carbonate	0.50	0.50	0.50	0.50
Salt	1	1	1	1
Vitamin premix	0.50	0.50	0.50	0.50
Spent mushroom substrate (SMS)	0	2.08	3.40	4.80
Crude Protein	14.50	14.55	14.78	14.60
Total digestible nutrients (g/day)	848	788	780	772
FAT	2.70	2.40	2.28	2.16
Neutral detergent fiber	38.50	38.40	39.80	41.2
Acid detergent fiber	25.20	26.70	27.6	28.6
Calcium	1.10	1.10	1	0.9
Phosphorus	0.40	0.35	0.25	0.16

**Statistical Analysis**

The trial was analyzed considering a completely randomized design by the GLM procedure of SAS 9.1. Means among treatment were compared by Tukey test.

## RESULTS AND DISCUSSION

The results indicated that there was no significant ( $p>0.05$ ) differences in the serum biochemical and hematological properties of the lambs among the four treatments (Tables 3 and 4), despite it was observed significant differences ( $p<0.05$ ) among the treatments for traits average daily gain and final weight of the Kurdish lambs (Table 5). For many centuries mushrooms have mainly been consumed for the purpose of health and safety [9, 20, 22]. Researchers have come to the conclusion that mushroom consumption has numerous useful effects on animals and humans besides immunomodulatory, hypocholesterolemic and anti-tumor influences [12, 19, and 29].

**Table 3: The serum biochemical properties, serum lipid properties and serum mineral content on blood of Kurdish lambs fed with different levels of spent mushroom substrate added in the feed**

Parameter	Control	SMS 15%	SMS 25%	SMS 35%	SEM	P Value
BUN (mg/dl)	43.87	35.77	42.50	32.75	4.97	0.36
AST (mg/dl)	127.12	141.77	125.35	115.58	10.84	0.42
ALT (mg/dl)	38.97	26.68	57.05	58.59	18.53	0.58
TG (mg/dl)	30	35	47.21	35	7.99	0.49
LDL (mg/dl)	43.33	26.55	38.22	23.07	13.22	0.67
HDL (mg/dl)	12	18.68	22.73	14.65	5.37	0.53
Calcium (mg/dl)	10.12	8.44	6.16	9.49	1.35	0.23
Phosphorus (mg/dl)	6.60	5.96	6.30	7.18	0.34	0.13
Iron (mg/dl)	97	115.75	171.25	93	31.41	0.31

SMS: Spent Mushroom Substrate, BUN: Blood Urea Nitrogen, AST: Aspartate aminotransferase, ALT: Alanine aminotransferase, TG: Triglyceride, LDL: Low Density Lipoprotein, HDL: High Density Lipoprotein, SEM: Standard Error of the Mean.

The results of this study showed that the consumption of SMS had no significant ( $p>0.05$ ) impact on blood urea nitrogen (BUN). The results of our study are compatible with those of [24], and [7], but different from the studies of Oh et al. [23] and Park et al [25]. Measuring concentrations of alanine aminotransferase (ALT) and aspartate aminotransferase (AST) indicated the functional and healthy ability of the animal's liver. Their increased activity is visible in liver injury. According to our research no significant ( $p>0.05$ ) toxicity can be seen in the lambs even if SMS is added up to 35% in the diet. SMS, as can be seen in table 3, made no significant ( $p>0.05$ ) difference in the levels of AST, ALT and triglyceride (TG), according to studies of Park et al [24]; Park et al [25] and Oh et al [23]. The addition of mushroom in the rat diet had an impact on serum triglyceride and cholesterol concentration. It also hindered the storing of cholesterol in liver [3, 11, and 21]. Results were similar to these finds. Mushroom affects the concentration of cholesterol and triglyceride in the serum and prevents cholesterol accumulation in liver [28]. The study concluded that SMS had no significant ( $p>0.05$ ) effect on the concentration of low density lipoprotein (LDL), high density lipoprotein (HDL) and triglyceride in Kurdish lambs serum. Although low density lipoprotein (LDL) levels in ruminants fed with SMS had no significant effect [23, 25], a remarkable decrease of LDL was visible [24]. In relation to high density lipoprotein (HDL) the case is opposite. Mushrooms have been recognized as a source of iron which is essential for the synthesis of Hb and oxygenation of RBCs [25]. The results of our study showed that SMS had no significant ( $p>0.05$ ) effect on the levels of iron, hematocrit (HCT) and hemoglobin (Hb) in Kurdish lambs. Calcium (Ca) and phosphorus (P) levels in elks and goats diets with SMS with no significantly increase, while iron, hematocrit (HCT) and hemoglobin(Hb) levels were significantly increased [24, 25]. The present study as shown in table 4 indicates that SMS consumption has no significant ( $p>0.05$ ) effect on mean corpuscular volume (MCV) and mean corpuscular hemoglobin (MCH) and coincides with the results obtained by [24, 25].

**Table 4: Hematological properties of Kurdish lambs fed with different levels of spent mushroom substrate added in the feed**

Parameter	Control	SMS 15%	SMS 25%	SMS 35%	SEM	P Value
HCT (%)	34.50	34	32.75	34.50	0.94	0.53
Hb (g/dl)	11.47	11.30	10.87	11.45	0.32	0.54
MCV (fl)	45.97	45.30	43.62	45.95	1.26	0.53
MCH (pg)	15.27	15.02	15.12	15.55	0.65	0.94
LY (%)	62	63	62.50	67	3.43	0.72
NE (%)	36.25	35.25	34.50	31.75	3.03	0.75
MO (%)	0.75	1	0.50	0.50	0.37	0.75
EO (%)	0	0	2.25	0.25	0.63	0.07
BA (%)	1	0.75	0.25	0.50	0.22	0.16

SMS: Spent Mushroom Substrate, HCT: Hematocrit, Hb: Hemoglobin, MCV: Mean corpuscular volume, MCH: Mean corpuscular hemoglobin, LY: Lymphocyte, NE: Neutrophil, MO: Monocyte, EO: Eosinophil, BA: Basophil, SEM: Standard Error of the Mean.

The present study as shown in table 4 indicates that SMS consumption has no significant ( $p>0.05$ ) effect on mean corpuscular volume (MCV) and mean corpuscular hemoglobin (MCH) and coincides with the results obtained by [24, 25]. Lymphocytes, small white blood cells, play a major role in the defending system of the body against infectious diseases. The results show that the consumption of SMS has no significant ( $p>0.05$ ) effect on lymphocytes, which is incompatible, with what park et al [25]. Apparently there is no sufficient information

concerning SMS in blood parameters of lambs. On the other hand, the experiments done on other animals may not be comparable to the case of lambs.

The amount of crude protein (CP), calcium (Ca), phosphorus (P), sodium (Na) crude fiber (CF), neutral detergent fiber (NDF), acid detergent fiber (ADF) and ash in SMS have remarkably changed in proportion to initial wheat straw (table 1). The ash concentration in the initial wheat straw was 10% while it gave rise to 37.04% in SMS. This change can have two reasons. The first reason is that when straw is used as an energizing source in the mushroom culture, in portion of its organic materials is analyzed and utilized by mushroom [4]. The second reason is that hence the mushroom cultivation contains some soils; the remaining compost is not completely free from soil although the soil is mechanically removed. Other researchers have reported that the ash in SMS was measured 38% to 53% [1, 27]. The low level of NDF and ADF is related to the severe decrease of organic substances in SMS, which in turn has been reported by other researches [1, 15]. The existence of high crude protein in SMS in comparison to initial wheat straw is because of added nitrogen materials to the process of composting period as well as the remaining parts of mushroom containing high amounts of nitrogen [1].

**Table5: Effect of treatments on the performance of Kurdish lamb**

Parameter	Control	SMS 15%	SMS 25%	SMS 35%	SEM
Initial weight (Kg)	32.27	32.52	32.98	33.91	1.70
Final weight (Kg)	50.50 <sup>a</sup>	50.66 <sup>a</sup>	55.01 <sup>b</sup>	49.43 <sup>a</sup>	0.91
Total body weight gain (Kg)	18.22 <sup>a</sup>	18.13 <sup>a</sup>	22.02 <sup>b</sup>	15.52 <sup>c</sup>	0.28
Average daily gain (g)	173.57 <sup>a</sup>	172.73 <sup>a</sup>	209.76 <sup>b</sup>	147.85 <sup>c</sup>	2.68
DMI (g/d/animal)	1198 <sup>a</sup>	1173 <sup>a</sup>	1098 <sup>ab</sup>	893 <sup>b</sup>	175

SMS: Spent Mushroom Substrate, DMI: Dry Matter Intake, SEM: Standard Error of the Mean

The above mentioned study shows that SMS 25% has a significant difference ( $p < 0.05$ ) with other treatments in average daily gain and final weight. The high concentration of Ash can lead to the limitation of minerals in the diet which in turn would have a negative effect on feeding [8, 13, 14, 15] that is why average daily gain in SMS 35% significantly reduces. The results concerning the weight parameters of this study approve the conclusions obtained by other researchers [1, 10, 15, and 18].

## CONCLUSION

This study concluded that SMS consumption had no harmful effect on the blood parameters of Kurdish male lambs. It is recommended that 25% of wheat straw be replaced by SMS.

## REFERENCES

- [1] Bakshi, M.P.S. and P.N. Langar. **1991**. *Indian J. Anim. Sci.* 61:6, 653-654.
- [2] Brunetti, G., P. Soler-Rovira, F. Matarrese and N. Senesi. **2009**. *J. Agric. Food Chem.* 57:10859-10865.
- [3] Bobek, P., O. Ozdin and M. Mikus. **1995**. *Physiol. Res.* 44:287-291.
- [4] Bonnen, A.M., L.H. Anton and A.B. Orth. **1994**. Lignin-degrading enzymes of the commercial button
- [5] Burton, S.G, J.R. Duncan, P.T. Kaye, and P.D. Rose. **1993**. *Biotechnol. Bioengin.* (USA). 42(8): 938-944.
- [6] Burton, K. S., J. B. V. Hammond and T. Minamide. **1994**. *Current Microbiol.* 28:275-278.
- [7] Chumpawadee, S., Sommart, K., Vongpralub, T. and Pattarajinda, V. **2006**. *J. Sci. Technol.* 28 (1): 59-70.
- [8] Crosbie, G. B. and J. B. Rowe. **1988**. The effect of lignin silica content of oat hulls on their in vitro digestibility. Proceeding of the Australian Society of Animal Production. P: 17.
- [9] Diyabalange, T., V. Mulabagal, G. Mills, D.L. DeWitt and M. g. Nair. **2008**. *Food Chem.*, 108:97-102.
- [10] Fazaeli, H.; Masoodi, A. R. T. **2006**. *Asian-Aust. J. Anim. Sci.*, 19 (6): 845-851.
- [11] Fukushima, M., T. Ohashi, Y. Fujiwara, K. Sonoyama and M. Nakano. **2001**. *Exp. Biol. Med.* (Maywood) 226:758-765.
- [12] Fukushima, M., Y. Nakano, Y. Morii, T. Ohashi, Y. Fujieara, K. Sonoyama. **2000**. *J. Nutr.* 130:2151-2156.
- [13] Gerrits, J. P. G. **1994**. *Compost. Sci. Util* (2):24-30.
- [14] Giovannozzi-Sermanii, G., G. Bertoni and A. Porri. **1989**. Biotransformation of straw to commodity chemicals and animal feeds. In: Enzyme Systems for Lignocellulose Degradation (Ed. W. Coughlan). Elsevier science, Amsterdam. P:371-382.
- [15] Kakkar, V. K., H. S. Garcha, S. Dhanda and G. S. Makkar. **1990**. *Indian J. Anim. Nutr.* 7(4):267-272.
- [16] Kley J, G. and T. F. Wetzler. **1981**. *Can. J. Microbiol.* 27:748-753.
- [17] Langar, P. N., J. P. Sehgal and H. S. Garcha. **1980**. *Indian J. Anim. Sci.* 50:942-946.
- [18] Langar, P.N., J.P. sehgal, V.K. Rana, M.M. Singh and H.S. Garcha. **1982**. *Indian J. Anim Sci.* 52:8, 634-637.
- [19] Lull, C., H.J. Wichers and H.F.J. Savelkoul. **2005**. *Mediators Inflamm*:63-80.
- [20] Mizuno, M., Morimoto, K.I. Minato and H. Tsuchida. **1998**. *Biosci. biotechnol. Biochem.* 62:434-437.

- [21] mushroom, *Agaricus bisporus*. *Applied and environmental microbiology (USA)*. 60: 960-965.
- [22] Nakajima, A., Ishida, T., Koga, M., Takeuchi, T., Mazda, O and M. Takeuchi. **2002**. *Int. Immunopharmacol*, 2:1205-1211.
- [23] Oh, Y.K., Lee, W.M., Choi, Ch. W., Kim, K.H., Hong, S. K., Lee, S. Ch., Seol, Y. J., Kwak, W.S., Nag-Jin Choi. **2010**. *Asian-Aust. J. Anim. Sci.* 12: 1608 – 1613.
- [24] Park, J.H, Yoon, S.H, Kim, S.W, Shin, D., Jin, S.K.,Yang,B.S and Y.M.Cho. **2012a**. *Asian Journal of Animal and Veterinary Advances*,11:1139-1147.
- [25] Park, J.H, Kim, S.W, Do, Y.J, Kim, H, Ko, Y.G, Yang, B.S, Shin, D and Young Moo Cho. **2012b**. *Asian-Aust. J. Anim. Sci*, 3: 320-324.
- [26] Tajbakhsh, J., M. A. Abdoli, E. Mohammadi Goltapeh, I. Alahdadi and M. J. Malakouti. **2008**. *Environmentalist* 28:476-482.
- [27] Takenaga, M., Aso, H.S and Y.Yamanaka. **1993**. *J. Japanese Society Grassland Sci*, 39:1, 22-27.
- [28] Shu Hui Hu , Zeng Chin Liang , Yi Chen Chia , Juang Lin Lien , Ker Shaw Chen , Min Yen Lee , and Jinn Chyi Wang. **2006**. *J. Agric. Food Chem.* 54:2103–2110.
- [29] Yoshioka, Y., R Tabeta, H saito, N. Uehara and F.Fukuoka.**1985**. *Carbohydr. Res.* 140:93-100.