

EVALUATION OF SUGAR BEET PULP TREATED WITH TRICODERMA REESI AND SACCHAROMYCES CERVICIA

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ABSTRACT

Sugar beet pulp (SBP) was treated with *Trichoderma verdi* and *Saccharomyces cervicia*, or with the mineral solution without the fungus or yeast (blank), by solid-state fermentation (SSF) technique. Crude protein (CP) and ash contents increased in treated sugar beet pulp without or with fungal treatment compared to the untreated material by (70.28 and 55.17%) and (72.99 and 105.315%), respectively. However, OM, CF, EE and NFE decreased in the SBP treated without or with fungal as compared to the untreated material. The value of NPN was highly elevated in the treated SBP compared to the untreated one. Insoluble and soluble protein in sodium chloride or pepsin as percentage of CP increased without or with fungal treatment.

No significant ($P > 0.05$) differences were detected at the different incubation times for in-situ DM, OM and CP disappearance or the fitted values between treated and untreated SBP. The intercept values at zero time (a), potentially degradable fraction (b), rate of degradation of b (c) for DM, OM and CP degradability were insignificantly ($P > 0.05$) different for both treated or untreated SBP.

In two digestibility and nitrogen balance trials the first group received a ration containing concentrate feed mixture (CFM), untreated SBP (USBP) and rice straw (RS) and the second group received a ration containing CFM, treated SBP (TSBP) and RS. Untreated or treated SBP were mashed and rice straw was chopped. A second digestibility trial was carried out to overcome the problems of the first. One group received CFM and TSBP in mash form with RS, while the second group received a ration containing CFM and TSBP mixed in pelleted form plus RS.

Feeding treated SBP significantly ($P < 0.05$) lowered feed consumption from CFM, treated SBP (TSBP) and rice straw (RS) than from untreated one. Digestion coefficients of DM, OM, CF and NFE in the first digestibility trials were significantly ($P < 0.05$) lower with treated SBP than that untreated, while EE digestibility was increased. In the second trial, digestibilities of DM, OM, CP, CF, EE and NFE were significantly ($P < 0.05$) increased with TSBP in the pelleted form as compared to the mash form. Comparing the two experimental stages, the results showed no significant ($P > 0.05$) differences between untreated SBP and TSBP in pelleted form. The values of TDN were significantly ($P < 0.05$) decreased with feeding treated SBP compared to untreated one, while treated SBP in pelleted form was significantly ($P < 0.05$) higher than that of mash form. Pelleted TSBP was significantly ($P < 0.05$) higher DCP values compared to mash form and untreated SBP.

The values of ruminal fluid pH were significantly ($P < 0.05$) lower when treated SBP was fed as compared to untreated one in the first digestibility trial, while in the second digestibility trial treated SBP in the pelleted form significantly ($P < 0.05$) decreased rumen pH values more than the mash form. No significant ($P > 0.05$) differences were found in both $\text{NH}_3\text{-N}$ and TVFA's concentrations, neither between treated and untreated SBP, nor between the pelleted and mash forms.

Blood plasma total protein, albumin and globulin were increased with the feeding of treated SBP as compared to the untreated one in the first digestibility trial, whereas in the second digestibility trial these values increased with the pelleted form of SBP more than those of the mash form. No significant differences were found for urea-N, AST and ALT with feeding treated or untreated SBP or between pelleted and mash forms.

It could be concluded from this study that, feeding untreated sugar beet pulp in rations for ruminants were the best in terms of total digestible nutrients, feed intake and rumen environmental conditions of the present study.

Key Words: Sugarbeet pulp, digestibility, fungal treatment, *Trichoderma Verdi*, *Saccharomyces cervicia*

INTRODUCTION

The interest in biological treatment with fungi as a method to improve utilization of crop residues has spread widely in recent years. There are two directions of biological treatments using aerobic fermentation to improve the nutritional quality of agriculture by-products. The first direction is the cellulolytic fungi which have the ability to produce extracellular cellulase enzymes that can hydrolyze cellulose to fermentable sugars. The second direction is to make use of fungi with high growth rates and high efficiency in converting the substrates into a biomass with high protein content.

Numerous fungal strains were used by many authors for the hydrolysis of cellulosic material such as *Aspergillus niger*, *A. terreus*, *Fusarium moniliforme* and *Chaetomium cellulolyticum* (Kim et al. 1985), *C. globosum* (Fadel 1983), *Myrothecium verrucosum* and *Trichoderma viride* (Larwenece and Abada 1987), *T. harzianum* (Fadel 1983 and 1989, Fadel and Foda 1993).

The technique of solid state fermentation (SSF) has been applied to improve the nutritional quality of agriculture by-products. This fermentation technique was based on cultivation of *Trichoderma* spp. on natural substrates such as sorghum grains (Chamswarng, 1992), sugar cane bagasse (Hamissa, et al., 1987), wheat straw (Singh et al., 1990), rice straw (Subhash, et al., 1991) and ground oil palm seeds (Kanjamaneesathiam et al., 2003).

This study was conducted to evaluate the effects of fungal treatment on feed intake, chemical composition, ruminal degradation, and nutritive value of sugar beet pulp.

MATERIALS AND METHODS

The present study was conducted at the experimental animal house belonging to Animal Production Research Institute, Agriculture Research Center, Ministry of Agriculture, Egypt, in collaboration with the Animal Production Department, Faculty of Agriculture, El-Mansoura University. Sugar beet pulp (SBP) "Int. Feed No. 4-00-669" was bought from factories produce sugar from sugar beets in Kafer El-Shaikh Governorate and was treated with *Trichoderma verdi* and *Saccharomyces cervicia* to increase CP content.

Treatment of sugar beet pulp with fungi and yeast

Treatment was carried out at room temperature in shaded area as follow: Sugar beet pulp (SBP) was spread as a layer of 25 cm depth on cement-plated ground. The spawn fungal mycelium, which was incubated previously on SBP, was mixed with the same residue (5kg/100kg w/w on DM basis).

Nutrient solution containing 10 kg ammonium sulfate, 7.5 kg urea 46% N, 3.3 kg magnesium sulfate, 6.6 kg super one phosphate and 6.6 kg molasses was dissolved in 20 litter water then added to one ton SBP. Moisture was adjusted to 60-65% with water during the treatment period. On the 5th day, 0.09% dried yeast (*Saccharomyces cervicia*) was dissolved in two litters of water and sprayed on the crop residue material. Treated material was mixed once every 48 hrs during the treatment period. Samples of treated material were taken daily and dried in the oven at 60 °C for 36 hrs. The blank samples were taken without added fungi or yeast but with added minerals and treated similarly.

Dried samples were ground to pass 1 mm screen and stored for chemical analysis (DM, CP, CF, EE & ash % and true protein nitrogen "TPN" according to official procedures (A.O.A.C. 1995). Non- protein nitrogen (NPN) was calculated by subtracting the TPN value from the total nitrogen (TN) concentration. Insoluble protein in pepsin (ISPP) and insoluble protein in sodium chloride (ISP) were determined according to Goering and Van Soest (1970) and Waldo and Goering (1979), respectively.

Soluble protein in either pepsin or sodium chloride was calculated as the differences between CP and insoluble fractions. After the seventh day of the treatment, treated materials were air-dried and used in feeding the animals for the digestibility studies. The chemical compositions of tested material before and after treatment are shown in Tables (1, 2 and 3).

Stages of the experiment

The study was carried out in three stages as follows:

First stage - In-Situ trials

The nylon bag technique was applied to compare the fungi-treated and untreated sugar beet pulp in 6 fistulaeted animals and to determine the disappearance of dry matter, organic matter and nitrogen contents in the rumen according to the method of (Mehrez and Ørskov, 1977 and Ørskov *et al.*, 1980). Nylon cloth (100% polyester) with a mean pore size of 120 µm was used for making the in-situ bags (5 x 8 cm) having a surface area of 80 cm² which were sown using nylon thread. The dry weighed bags containing approximately 2 gm of dried tested materials were introduced into the rumen through the fistula for the determination of DM and OM degradability and were incubated for 6, 12, 18, 24, 36 and 48 hrs. Another set of bags each containing 2 gm were incubated for protein degradability at the same time. After removing the bags from rumen they were washed gently with flowing stream of tap water until the rinsing fluid was clear in a desiccators then weighed. Dry matter, organic matter and nitrogen were determined according to the official methods (A.O.A.C., 1995). The data were fitted to the model of Ørskov and McDonald (1979).

Second stage - first digestibility trial

In the second stage, two digestibility and nitrogen balance trials were conducted to study effects of treating sugar beet pulp with fungi (TSBP) as compared to untreated pulp (USBP). Six adult Ossimi rams with an average live body weight 65 kg were randomly distributed to two groups. The first group received a ration containing concentrate feed mixture (CFM), untreated SBP (USBP) and rice straw (RS) and the second group received a ration containing CFM, treated SBP (TSBP) and RS as shown in Table 7. The animals were adapted for 3 weeks in semi-open pens. The rams were individually placed into metabolic cages for 14 days, where seven days were for adaptation, while the following seven days were for faeces and urine collection. Concentrate feed mixture was offered to animals as 1% of live body weight once at 07.00 hr, while untreated or treated SBP were mashed and offered ad-libitum with chopped rice straw.

Third stage - second digestibility trials

According to results obtained from the second experimental stage this study was modified to overcome the problems experienced during that second stage. Another six animals were divided into two groups, three animals each. The first experimental group received a ration containing CFM and TSBP in mash form with RS, while the second group received a ration containing CFM and TSBP mixed in pelleted form plus RS. Both the concentrate mixture forms and chopped rice straw were offered ad-libitum. Refusals were collected daily to determine the actual amount of feed intake.

Sampling and chemical analysis

The digestion experiment

Samples from feeds, refusals, feces and urine were analyzed for its proximate constituents according to official procedures (A.O.A.C. 1995). The NFE was calculated by difference.

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Rumen Parameters

Rumen liquor samples were taken from each animal individually at the end of the digestibility trials in the second and third stages. These samples were taken using stomach tube before feeding (Zero time) and at three and six hours after feeding. Samples were then filtered through four layers of muslin cloth. The pH values in the rumen liquor were measured by a HANA pH meter. Ammonia-N ($\text{NH}_3\text{-N}$) was determined according to Conway (1978). Total volatile fatty acids (TVFA's) were determined by the steam distillation method as described by Warner (1964).

Blood Parameters

Blood samples were withdrawn from the jugular vein in the second and third stages from each animal individually at the end of the digestibility trials. These samples were taken after 4 hrs post feeding in heparinized tubes. Blood plasma was immediately separated by centrifugation and stored at -20°C until subjected for analysis (Siest *et al.*, 1981). The analyses included total proteins, albumin, creatinine, alanine transaminase (ALT) and aspartic transaminase (AST) which were determined using commercial kits (Sentine CH. 20155 Milan, Italy[®]). Globulin was calculated by difference between total protein and albumin concentrations.

Statistical analysis

Statistical analysis was carried out using SAS (1999). Degradability of DM, OM and CP of each incubation interval, were analyzed as one-way analysis of variance according to the following model:

$$Y = \mu + x_i + e_{ij}$$

where Y = observation, μ = mean, x_i the effect of treatment for $i=1$, untreated and 2, treated, and e_{ij} = experimental error

Digestibility, rumen and blood data in each stage were also analyzed as one-way analysis of variance according to the following model:

$$Y = \mu + x_i + e_{ij}$$

where Y = observation, μ = mean, x_i = the effect of treatment for $i = 1$, untreated and 2, treated, and e_{ij} = experimental error

Then, digestibility, rumen and blood data of the two stages together were analyzed as a two-way analysis of variance according to the following model:

$$Y = \mu + x_i + x_j + x_{ij} + e_{ijk}$$

where Y = observation, μ = mean, x_i = the effect of treatment for $i = 1$, untreated (USBP), 2, treated (TSBP), 3, treated in mash form (MSBP) and 4, tread in pelleted form (PSBP), x_j = the effect of stage for $i = 1$, the 1st stage and 2, the 2nd stage, x_{ij} = the interaction and e_{ijk} = experimental error.

Duncan's Multiple Range Test (Duncan, 1955) was used to separate the means when the main effect was significant.

RESULTS AND DISCUSSIONS

Chemical composition

Data in Table (1) show increased CP and ash contents in treated sugar beet pulp without or with fungal treatment compared to untreated one. The contents of CP and ash of SBP increased by (70.28 and 55.17%) when treated with the mineral solution alone and by (72.99 and 105.32%) when treated with added fungi. However, OM, CF, EE and NFE decreased after treatment without or with added fungi as compared to the untreated SBP. The NRC (1989) reported the standard composition of dried SBP to be 91% DM, 95.6% OM, 9.7% CP, 0.6% EE, 19.8% CF, 65.5% NFE and 4.4% ash. In

Table 1. Chemical composition (% , dry matter basis) of sugar beet pulp as affected by experimental treatments.

Item	Before treatments	After treatments	
		without fungi ¹	with fungi
DM	90.30	46.92	46.92
Nutrients content of DM, %			
OM	96.23	94.15	92.26
CP	11.07	18.85	19.15
CF	22.57	21.89	19.14
EE	3.32	2.18	1.66
NFE	59.27	51.23	52.31
Ash	3.77	5.85	7.74

¹ Treated without fungi = treated with the same mineral solution which was added as nutrients for fungal growth.

other studies (Haaksma, 1982) the CP content of SBP ranged from 9.88 to 11.9%, and CF content was about 20% on DM basis. Kelly (1983) found that dried beet pulp was a useful ruminant feed containing 12.5 MJ metabolizable energy and 79 g digestible CP per Kg DM. Bhattacharya *et al.* (1975) reported that, beet pulp could be used as a satisfactory source of energy in rations for growing and fattening ruminants.

Results in Table 2 show the nitrogen fractionations and protein solubility of SBP before and after treatment.

Despite the slight increase of true protein nitrogen by 1.18 and 5.88% in treated SBP without or with fungi, respectively, the values of NPN was greatly increased in treated SBP as compared to the untreated material. Abo-Donia *et al.* (2004) also found that TN and NPN were increased after treatment with fungi of both peanut hulls (PNH) and sugarcane bagasse (SCB) than before treatment, while TPN was decreased with the

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Table 2. Content of nitrogen fraction and protein solubility (%on dry matter basis) before and after fungal treatment of sugar beet pulp.

Item ¹	Before treatments	After treatments	
		without fungi	with fungi
<u>Nitrogen fractions (%)</u>			
Total nitrogen	1.77	3.02	3.06
True PN	1.70	1.72	1.80
TPN/TN	95.98	57.03	58.75
NPN	0.07	1.30	1.26
TPN of TN, %	4.02	42.97	41.25
<u>Solubility of protein,%</u>			
CP content, %	11.07	18.85	19.15
<u>1. In sodium chloride</u>			
ISP	10.96	18.66	18.95
SP	0.11	0.19	0.20
ISP/CP	0.99	0.99	0.99
<u>2. In pepsin</u>			
ISPP	4.12	1.89	1.38
SPP	6.95	16.97	17.77
SPP/CP	0.37	0.10	0.07

¹TN= Total nitrogen and TPN= True PN.

ISP= Insoluble protein in sodium chloride and SP= soluble protein in sodium chloride.

ISPP= Insoluble protein pepsin and SPP= soluble protein in pepsin.

fungal treatment of crop residue than untreated one. In this regard, the finding of **EI-Ashry *et al.* (2002)** and **Rane *et al.* (2004)** supported the present findings. The TPN as a parentage of TN decreased after treatment without or with fungal. This result could be attributed to the supplemented nitrogen in the mineral solution (urea and ammonium sulphate). Insoluble and soluble protein as percentages of CP were increased without or with the fungal treatment. These results are in a good agreement with **Lynch *et al.* (1977)**. Protein insoluble in sodium chloride decreased after fungal treatment. The decrease of protein insoluble in sodium chloride might be due to the higher content of true protein in SBP, while the higher content of NPN in SCB after treatment could be a reflection to the of increasing soluble nitrogen that was reflected on insoluble protein in sodium chloride. At the same time, fungal treatment of SBP increased insoluble and soluble protein in pepsin than untreated one. **Gupta and Singh (1991)** found that insoluble N increased from an initial value of 0.979% on day 0 to 1.409% on day 13 in fungal-treated paddy straw.

Chemical composition (%) of rice straw (RS), concentrate feed mixture (CFM), untreated or treated sugar beet pulp (U-SBP and T-SBP) which were used in the experiments are shown in Table (3). The summative analysis of the RS and CFM used to formulate the experimental rations were within the normal published ranges according to **Abou-Raya, (1967)**. Meanwhile, SBP composition was comparable to the results of **Haaksma, (1982)** and **Alibes *et al.* (1982)**.

Table 3. Chemical composition of the different ingredients %on dry matter basis.

Item ¹	DM	OM	CP	CF	EE	NFE	Ash
CFM1	89.65	87.81	14.04	15.29	2.73	55.75	12.19
CFM2	90.21	90.79	18.28	18.62	2.01	51.89	9.21
Rice straw	90.85	83.97	3.04	37.35	1.71	41.87	16.03
U-SBP	90.30	96.60	10.00	20.38	3.00	63.22	3.40
T-SBP	90.40	93.00	17.31	17.30	1.50	56.89	7.00

¹CFM1= used in the 1st experiment and composed of 25% cottonseed meal, 26% yellow corn, 39% wheat bran, 6% molasses, 3%limestone and 1% common salt.

CFM2= Used in the 2nd experiment and containing of (75% TSBP and 25% CFM1 as mash or pelted form)

In-situ studies

At the different incubation times no significant differences were detected for DM, OM and CP disappearance between treated or untreated SBP (Table 4).

Table 4 Effect of experimental treatments on in-situ disappearance of DM, OM and CP of SBP.

Item	Incubation time (hr)					
	6	12	18	24	36	48
<u>Dry matter</u>						
Untreated SBP	10.08	15.81	19.23	23.12	28.71	32.87
Treated SBP	10.02	15.77	19.16	23.10	28.70	32.85
±SE	0.29	0.75	0.53	0.20	0.53	0.52
<u>Organic matter</u>						
Untreated SBP	10.82	17.20	21.41	25.38	32.19	34.66
Treated SBP	10.74	17.14	21.33	25.35	32.18	34.63
±SE	0.07	0.33	0.29	0.41	0.29	0.54
<u>Crude protein</u>						
Untreated SBP	7.07	12.63	17.06	20.88	24.38	27.90
Treated SBP	7.04	12.25	17.00	20.86	24.37	27.88
±SE	0.31	0.58	0.54	0.48	0.64	0.65

Table 5. Fitted disappearance values ($P = a + b(1 - e^{-ct})$) of DM, OM and CP of untreated or treated SBP.

Item	Time					
	6	12	18	24	36	48
Dry matter						
Untreated SBP	10.29	15.29	19.53	23.13	28.76	32.81
Treated SBP	10.23	15.24	19.49	23.09	28.74	32.81
±SE	0.46	0.42	0.40	0.41	0.47	0.53
Organic matter						
Untreated SBP	10.82	16.91	21.79	25.71	31.38	35.04
Treated SBP	10.75	16.84	21.74	25.67	31.36	35.02
±SE	0.14	0.19	0.26	0.32	0.42	0.47
Crude protein						
Untreated SBP	7.07	12.74	17.09	20.43	24.96	27.63
Treated SBP	6.92	12.59	16.97	20.33	24.93	27.66
±SE	0.32	0.43	0.50	0.54	0.60	0.67

Mehrez and Ørskov (1977) pointed out that the most important factors affecting accuracy of disappearance results were homogeneity of the tested sample and sample size in relation to bag size which were standing in the present study. Any differences in degradability should reflect the nature of the tested material and its cell wall components.

The data were fitted to the exponential equation ($P = a + b(1 - e^{-ct})$) for more accurate comparison (Table 5). Fitted value of DM, OM and CP at different times were not significantly ($P < 0.05$) different for treated or untreated SBP. These results are similar to those obtained by Mehrez and Maklad (1999) with clover straw based diet. Feed particles must be digested before passage from the rumen (Allen and Mertens, 1988).

The intercept values at zero time (a), potentially degradable fraction (b), rate of degradation of b (c) for DM, OM and CP degradability are presented in Table (6). They were not significantly different ($P > 0.05$) for both treated and untreated SBP. Richardson *et al.* (2003) reported that sugar beet pulp did not contain a rapidly degradable N fraction (a) and had a slower rate of degradation (c) of the potentially degradable (b) fraction than winter barley. Although the degradation characteristics of DM, OM and CP of both treated or untreated SBP were not recognized using the in situ measurement, yet there were significant effects of the biological treatment in vivo (Table 6). Such difference might be due to varying retention time in the rumen. Therefore, markers should be used to determine the “K” value. Another possibility is that treatment effects were taking place posterior to the rumen. This subject needs further detailed studies.

The digestion trials

Table 7 shows the chemical composition of the consumed rations during the first experimental stage where contents of OM, CP, EE and NFE with treated SBP were decreased, but contents of CF and ash increased, however. These results are mainly due to the increased consumption of rice straw and the decreased consumption of CFM and TSBP. For this reason, the second experimental stage was carried out to solve this problem. Contents of OM, CP, EE and NFE were increased during the second stage, while CF and ash were decreased. These were achieved through increasing feed consumption from SBP and CFM offered in a pelleted form and consequently decreasing the intake of rice straw.

Feeding treated SBP significantly ($P < 0.05$) lowered feed consumption from CFM, treated SBP (TSBP) and rice straw (RS) than from untreated one (Table 8). **Thompson and Kennington (2001)** reported that alkaloids are nitrogenous compounds produced by some fungus and are implicated in decreasing palatability and intake because of its bitterness.

Table 6. Degradation characteristics of the treated and untreated sugar beet pulp.

Item	a	b	c	a + b
<u>Dry matter</u>				
Untreated SBP	4.39	38.80	0.028	43.19
Treated SBP	43.1	38.89	0.030	43.20
±SE	0.55	1.35	0.001	0.87
<u>Organic matter</u>				
Untreated SBP	3.25	38.43	0.036	41.68
Treated SBP	3.15	38.53	0.037	41.68
±SE	0.25	0.64	0.001	0.60
<u>Crude protein</u>				
Untreated SBP	-0.32	31.82	0.044	31.50
Treated SBP	-0.45	32.19	0.044	31.73
±SE	0.32	0.63	0.002	0.94

a = the zero time intercept, **b** = potentially degradable fraction and **c** = rate of degradation of **b**

In the second stage, pelleting the treated SBP significantly ($P < 0.05$) increased CFM and TSBP consumption than the un-pelleted form. Feed consumption of treated SBP mixed with CFM in pelleted form was the best in sheep diet, where it was significantly ($P < 0.05$) higher compared to mash form. Reduction in feed intake might be due to increase $\text{NH}_3\text{-N}$ concentration in the blood (**Hadjipanayiotou 1984 and Khorshed 2000**), which might cause unfavorable condition, for the activity of rumen microorganisms, then reflected on rate of rumen fermentation and subsequently reduced voluntary feed intake (Balch and Campling 1962).

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Table 7. Chemical composition of the consumed rations %on dry matter basis.

Item	Experimental--1		Experimental--2	
	USBP	TSBP	MSBP	PSBP
Consumed ingredients (% dry matter basis)				
RS	14.37	41.53	69.50	34.37
CFM	42.9	34.80	7.51	16.40
SBP	42.73	23.67	22.99	49.23
Chemical composition of the tested rations				
DM	90.1	90.33	90.66	90.43
OM	90.02	86.10	84.91	87.85
CP	11.92	11.34	7.85	13.05
EE	3	2.23	1.92	1.96
CF	22.9	27.6	34.33	26.49
NFE	52.21	44.93	40.81	46.34
Ash	9.98	13.90	15.09	12.15

MSBP= (75%TSBP+25% CFM) in mashed form

PSBP= (75%TSBP+25% CFM) in pelleted form

There are many factors that influence the functional specific gravity of a particle. Particle size is a major factor. As particle size of forage decreases, its specific gravity increases. Part of the function of rumination is to reduce the particle size so that it has an appropriate size and specific gravity to pass out of the rumen. This is recognized on how long hay escapes the rumen and does not remain floating at the top (Weiss, 2000).

Digestibility coefficients and nutritive value

Digestion coefficients of DM, OM, CF and NFE in the second stage were significantly ($P<0.05$) lower with treated SBP than the untreated, while EE digestibility was increased (Table 8). Van Beek (1991) showed that the biological treatments act, as enzyme system, on 3 different levels. Firstly, it neutralizes the anti nutritive factors such as pentosans and beta-glucans. These anti nutritive factors increase the viscosity and lower the digestibility, resulting in a lower absorption of the feed nutrients. Secondly, it is able to breakdown the cellulose by the cellulase complex, As well as the breakdown of some other cell-wall components. Thirdly, it gives supports to the animal's digestive enzymes resulting in an overall better digestion. However, sometimes it is difficult to prove its role in improving animal performance.

In the third experimental stage, digestibilities of DM, OM, CP, CF, EE and NFE were significantly ($P<0.05$) increased with TSBP in the pelleted as compared to that offered in the mash form. Comparing the two experimental stages the results showed no significant ($P>0.05$) differences between untreated SBP and TSBP in pelleted form. Adams (1989) mentioned that there was a clear wide range of possibilities for the use

of biological treatment in animal nutrition. A wide variety of enzymes is available and this can be applied to appropriate feed materials to stimulate their suitability for use in animal feeding.

Table 8. Feed intake, nutrients digestibility and nutritive values during the two experimental stages for testing treated or untreated SBP in sheep diets.

Item	Experiment - 1 (second stage)		Experimental - 2 (third stage)		±SE
	USBP	TSBP	TSBP1	TSBP2	
<u>Feed intake, g/h/d</u>					
CFM	582.73 ^{Aa} ±0.00	286.95 ^{Bb} ±0.00	34.49 ^{Db} ±14.07	159.88 ^{Ca} ±14.07	±9.95
Sugar beet pulp	584.54 ^{Aa} ±33.75	184.42 ^{Bb} ±33.75	106.43 ^{Bb} ±42.59	483.94 ^{Aa} ±42.59	±38.43
Rice straw	197.75 ^{Bb} ±17.52	325.24 ^{Aa} ±17.52	323.43 ^A ±8.06	339.48 ^A ±8.06	±13.64
Total DMI	1365.02 ^{Aa} ±34.94	778.61 ^{Cb} ±34.94	414.34 ^{Db} ±56.69	983.29 ^{Ba} ±56.69	±47.09
<u>Nutrients digestibilities, %</u>					
DM	65.20 ^{Aa} ±0.34	59.87 ^{Cb} ±0.34	55.41 ^{Db} ±0.45	61.47 ^{Ba} ±0.45	±0.40
OM	67.43 ^{Aa} ±0.33	63.58 ^{Bb} ±0.33	60.02 ^{Cb} ±0.55	66.74 ^{Aa} ±0.55	±0.46
CP	64.24 ^A ±0.56	63.77 ^A ±0.56	50.11 ^{Bb} ±1.27	66.22 ^{Aa} ±1.27	±0.98
CF	61.68 ^{Aa} ±0.39	57.11 ^{Bb} ±0.39	55.78 ^{Bb} ±0.60	60.56 ^{Aa} ±0.60	±0.51
EE	63.05 ^{Bb} ±0.40	66.09 ^{Aa} ±0.40	58.43 ^{Db} ±0.23	60.88 ^{Ca} ±0.23	±0.33
NFE	70.93 ^{Aa} ±0.31	67.38 ^{Bb} ±0.31	65.54 ^{Cb} ±0.45	70.68 ^{Aa} ±0.45	±0.38
<u>Nutritive value, %</u>					
TDN	63.06 ^{Aa} ±0.42	56.59 ^{Cb} ±0.42	52.36 ^{Db} ±0.63	60.13 ^{Ba} ±0.63	±53
DCP	7.66 ^B ±0.23	7.24 ^B ±0.23	3.95 ^{Cb} ±0.35	8.65 ^{Aa} ±0.35	±0.30

a-b Means in the same row having different superscripts differ (P<0.01) in stage

A-D Means in the same row having different superscripts differ (P<0.01) in different stages.

The values of TDN were significantly (P<0.05) decreased with feeding treated SBP compared to the untreated one, while treated SBP in the pelleted form was significantly (P<0.05) higher in TDN than that in the mash form. Pelleted TSBP had

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significantly ($P < 0.05$) higher DCP values as compared to the mash form and the untreated SBP. Mehrez and Maklad (1999) showed that the TDN%, ME (Mj/Kg DM) and DCP% significantly ($P < 0.01$) decreased with adding enzyme to clover straw based diet.

Rumen fermentation parameters

The rumen pH were significantly ($P < 0.05$) lower in animals fed the treated SBP as compared to the untreated ones during the 1st digestibility trial, whereas in the 2nd digestibility trial feeding treated SBP in the pelleted form significantly ($P < 0.05$) decreased pH values than that fed in the mash form (Table 9). No significant ($P > 0.05$) differences were found for both $\text{NH}_3\text{-N}$ and TVFA's neither between treated and untreated nor between pelleted and mash forms. El-Sayed *et al.* (1997) found that the trend of pH values might reflect increase in TVFA's production in the rumen through the first 3 hours post feeding. This may be due to the fermentation process of non structural carbohydrates (NSC) leading to decrease in pH. Mehrez (1992) reported that the optimal ruminal NH_3 concentration for maximal rate of rumen fermentation is associated with dietary source and level of energy to be fermented in the rumen.

Table 9. Rumen parameters during the two experimental stages for testing treated and untreated SBP in sheep diets.

Item	Experiment-1 (stage 2)		Experiment-2 (stage 3)		±SE
	USBP	TSBP	TSBP1	TSBP2	
pH	6.98 ^{Ba} ±0.15	6.51 ^{Cb}	7.78 ^{Aa} ±0.12	7.70 ^{Bb}	±0.14
$\text{NH}_3\text{-N}$ (mg/dl)	14.47 ±0.27	14.81	12.50 ±0.18	14.10	±0.23
TVFA's (meq/dl)	6.40 ±1.53	6.29	6.29 ±0.83	6.98	±1.23

Blood parameters

Data in Table 10 show that total protein, albumin and globulin were increased in the plasma of sheep fed the treated SBP as compared to their mates fed the untreated SBP in the 1st digestibility trial. In the 2nd digestibility trial, on the other hand, these values increased with the pelleted form of SBP than those of the mash form. Pambu-Gollah *et al.* (2000) concluded that the plasma concentration of all the blood metabolites studied were sensitive to seasonal changes in nutrient supply and that they could be of use as a management tool in free-range farming systems in which conventional methods of nutritional assessment are difficult to apply. No significant differences were found for urea-N, AST and ALT with feeding treated or untreated SBP or between pelleted and mash forms. The presented data was in the normal range as

described by Mohamed and Selim (1999) for GOT (60-150 IU/l) and GPT (15-27 IU/l) for healthy sheep.

Table 10. Some blood parameters at two experimental stages for tested treated or untreated SBP in sheep diet.

Item	Experiment-1 (Stage 2)		Experiment-2 (Stage 3)		±SE
	USBP	TSBP	TSBP1	TSBP2	
Total protein (%)	6.75 ^{Db} ±0.10	7.38 ^{Ba}	7.09 ^{Cb} ±0.07	8.04 ^{Aa}	±0.09
Albumin (%)	3.97 ^{CBb} ±0.08	4.30 ^{Aa}	3.95 ^{Cb} ±0.04	4.17 ^{ABa}	±0.06
Globulin (%)	2.79 ^{Cb} ±0.07	3.08 ^{Ba}	3.13 ^{Bb} ±0.06	3.87 ^{Aa}	±0.07
Urea nitrogen (mg/dl)	31.27 ±0.31	30.65	30.88 ±0.32	31.03	±0.31
GOT (iu/l)	19.00 ±4.00	23.00	23.00 ±2.83	27.00	±3.46
GPT (iu/l)	24.67 ±1.65	27.00	24.67 ±2.17	31.00	±1.93

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تقييم تفل بنجر السكر المعامل بفطر التريكوديرما ريسى والخميرة
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تمت معاملة تفل بنجر السكر بفطر التريكوديرما فيردى ، وخميرة أو بدونها (المعيار) . أوضحت النتائج زيادة محتوى البروتين الخام في تفل البنجر المعامل بالفطر أو المعامل بالمحلول الملحي بدون إضافة الفطر مقارنة بتفل البنجر غير المعامل حيث كانت النسب (70.28 و 55.17%) و (%) (105.32 , 72.99) في حين حدث انخفاض في محتوى التفل من كل من المادة العضوية والألياف الخام ومستخلص الأثير والمستخلص الخالي من الأزوت. محتوى الأزوت غير البروتيني ارتفع بشدة في التفل المعامل بالفطر أو بدونه مقارنة بغير المعامل. البروتين الذائب وغير الذائب في محلول كلوريد الصوديوم أو الببسين ازداد مع معاملة تفل البنجر بالفطر أو المعاملة بدون الفطر (المعيار).

لم تظهر اختلافات معنوية ($P > 0.05$) في معدل اختفاء المادة الجافة أو العضوية أو البروتين الخام عند تحضين العينات المعاملة وغير المعاملة في كرش الحيوانات وكذلك المصححة. الجزء المتكسر في الكرش ومعدل التكسر للمادة الغذائية في الكرش للمادة الجافة أو العضوية أو البروتين الخام لم يحدث له تغير معنوي بمعاملة تفل البنجر بالفطر مقارنة بعدم المعاملة. من خلال تجربتي الهضم الأولتين حيث غذيت الأولى على عليقه لا تحتوي تفل معامل في حين غذيت الأخرى على عليقه تحتوي التفل المعامل وكان العلف المركز في صورة أصابع وتفل البنجر المعامل وغير المعامل مطحونا بينما في تجربتي الهضم التاليتين تم خلط تفل البنجر مع العلف المركز في صورة أصابع بينما تم الخلط ولم يتم التصبيغ في المجموعة الثانية. أوضحت نتائج التجربة الأولى انخفاض المأكول معنوية في المجموعة التي غذيت على عليقه تحتوي تفل البنجر المعامل بالفطر مقارنة بالمجموعة التي غذيت تفل بنجر غير معامل. كما أدت التغذية على العليقه التي تحتوي تفل البنجر المعامل إلى انخفاض معامل هضم كل من المادة الجافة والعضوية والألياف الخام والمستخلص الخالي من الأزوت في حين ارتفع معامل هضم مستخلص الأثير. في التجربة التالية وجد ارتفاع في قيمة معاملات الهضم لكل من المادة الجافة والعضوية والألياف الخام والمستخلص الخالي من الأزوت والبروتين الخام ومستخلص الأثير وذلك عند التغذية على علف مركز مع تفل معامل في صورة مكعبات مقارنة بالمجموعة الثانية التي غذيت على نفس المركز في صورة مجروشة. المقارنة بين التجريبتين الأولى والثانية أوضحت عدم وجود اختلافات معنوية بين التغذية على علف مركز مع تفل بنجر غير معامل وتفل بنجر مع علف مركز في صورة مكعبات. معاملة تفل البنجر بالفطر خفضت من القيم الغذائية للعلائق التي تحتوي عليه. بينما ارتفعت قيمة البروتين المهضوم عند تكعيب العلف المحتوى على تفل معامل بالفطر.

من نتائج التجربة الأولى وجد أن التغذية على العليقه المحتوية على تفل البنجر المعامل خفضت من حموضة الكرش معنويا مقارنة بالتي تحتوي تفل غير معامل كذلك عند خلطه مع العلف المركز في صورة أصابع في التجربة الثانية في حين لم يحدث تغير لقيم نتروجين الأمونيا والأحماض الكلية الطيارة سواء في العليقه المعاملة أو غير المعاملة أو المكعبة أو المجروشة في التجريبتين. نتائج تحاليل الدم أوضحت زيادة البروتين الكلى والألبومين والجلوبيولين عند التغذية على علائق تحتوي تفل البنجر المعامل كذلك زادت قيم هذه القياسات في التجربة الثانية مع العلف المكعب مقارنة بالمطحون. إنزيمات وظائف الكبد لم يحدث بها تغير في التجريبتين على السواء.

من نتائج هذه التجربة يمكننا القول إن استخدام تفل بنجر السكر غير المعامل في علائق المجترات كان أفضل من المعاملة حيويا بالفطر والخميرة