CHEMICAL COMPOSITION AND IN-VITRO DIGESTIBILITY OF SUGARCANE BAGASSE AND RICE HUSK TREATED WITH THREE STRAINS OF WHITE ROT FUNGI AND EFFECTIVE MICROORGANISM

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Abstract: A study was conducted to evaluate the effect of biological treatments of sugarcane bagasse (SCB) and rice husk (RH) with three strains of white-rot fungi (WRF) (Pleurotusostreatus (Po), Pleurotusflorida (Pf) and *Trichodermaviride* (*Tv*) and effective microorganism (EM) on the chemical composition and in-vitro digestibility. The experiment consisted of 2x5 factorial arrangements, two levels of feed (SCB and RH) and five levels of biological treatments (Control, Po, Pf, Tv, and EM). Treatment of RH with EM, Tv, Po and Pf, significantly increased crude protein content from 7.90% in untreated to 7.92. 10.46, 10.61 and 11.35%, respectively. The corresponding increase in CP% of sugarcane from 2.61% was 3.41, 5.96, 5.89 and 5.95%. Treatments significantly (P<0.001) decreased neutral detergent fiber, acid detergent fiber, acid detergent lignin cellulose and hemicelluloses contents with the lowest value recorded for Tv. The IVOMD, IVDMD and metabolizable energy (ME) were significantly (P<0.001) increased. In conclusion, the study indicates that treatment of RH with Trichodermaviride and SCB with EM is more effective than others in improving the nutritive value of the roughages. We suggest evaluation of the treated roughages on animal performance.

Key words: *In-vitro* digestibility, white-rot fungi, effective microorganism, sugar cane bagasse, rice husk.

Introduction

Biological treatment employs microorganisms and their enzymatic machineries to break down lignin and alter lignocelulose structures. The use of white rot fungi (WRF) under solid-state fermentation (SSF) is the most promising biological treatment to mineralize lignin component to CO₂ and water in pure culture (Isroi et al., 2011). Several studies during the last decades indicated that colonization of lignocelluloses agro-industrial by-product with white-rot fungi had positive impact on in vitro degradability (Jalc, 2002; Tripathiet al., 2008). During SSF, the fiber fraction of feed such as NDF and ADF could be reduced, while the crude protein content increased. Studies also showed that dry matter is lost during SSF, but digestibility of ADF and NDF is improved. However, the improvement in nutritive value of lignocelluloses by WRF varies with feed type, fungal strains, temperature and fungal growing techniques (Tripathi and Yadav, 1992). Among the white-rot fungi, Trichoderma species such as Trichodermaviride and Trichodermareesie (Abdel-Azim et al., 2011), and Pleurotus species such as Pleurotusdjamor, Pleurotussajor-caju, Pleurotusostreatus and Pleurotus florid (Singh et al. 1990; Nasehi et al., 2013) have efficiently reduced indigestible cell wall component and increased dry matter digestibility (DMD) of lignocelluloses. Effective microbes (EM) is another biological treatment method that has been utilized to improve the nutritive value of lignocelluloses by-products.

Despite the promising potential of biological treatment with WRF and availability of large volume of lignocellulose by-products that can be utilized as animal feed in Ethiopia, research has not been conducted to evaluate the effects of biological treatment on the nutritive value of these potential feed resources under the production and environmental scenario of the country. Hence, there is no information to advice producers and users concerning appropriate and efficient use of these resources as animal feed. Therefore, the present study was conducted to evaluate the effects of three strains of WRF and effective microbes on chemical compositions, *in-vitro*, and *in-sacco* rumen degradability of sugarcane bagasse and rice husk.

Materials and Methods

Chemical analysis was conducted at Haramaya University Animal Nutrition Laboratory and In-vitro digestibility was done at Holeta Research Center. Haramaya University is located at 9° 26'N latitude, $42^{\circ}3'$ E longitudeand at about 1980 meters above sea level (m.a.s.l.). The mean annual rainfall of the study area is about 870 mm with a range of 560-1260 mm, and the mean maximum and minimum temperatures are 23.4°C and 8.25°C, respectively (Haramaya University Meteorological Station). Whereas, Holeta Research Center is located at 9° 3' N latitude, 38° 30' E longitude and at about 2400 m.a.s.l. with mean minimum and maximum temperatures of 6.1° C and 22.2° C, respectively.

The sugarcane bagasses (SCB) was obtained from Wonji Sugar Factory and rice husk (RH) from Bench Maji Zone. Three strains of WRF, namely Trichodermaviride, Pleurotusflorida and Pleurotusostreatus-were obtained from plant protection section of the school of plant sciences of Haramaya University. The samples were kept under 4°Cuntil used. Each fungi were grown on a Petri dish containing potato dextrose agar (PDA) as nutrient for three days and the activated microorganisms were sub cultured on Petri dish (9 cm) containing 25ml potato dextrose agar (PDA) for another seven days. The slant culture samples were used to inoculate the spawn flasks. At this stage, the fungal strains were cultured in a media containing: 4% molasses, 0.4% urea, 0.2% KH₂PO4 and 0.03% MgSO4 $(7H_2O)$ per one liter of water. Two hundred ml of this media was incubated in 500 ml sterilized conical flask contains 100g ground waste. The flasks were sterilized (121°C for 15 minutes), cooled and inoculated with the prepared inoculums and incubated at 25°C for seven days to prepare spawn. The spawn was used to inoculate polyethylene bag containing 500 g of sugar cane bagasse and rice husk moisten with 500 ml of fungal medium (w/v) and distributed in polyethylene bag in four replications. Each polyethylene bag was inoculated with 10 % (w/w) spawns of Trichodermaviride, Pleurotusflorida and Pleurotusostreatus (Jahromi et al., 2010). A control sample was treated with the above mentioned media without any fungal strain. The control and treated materials were adjusted with media to approximately 60% humidity and was incubated at room temperature for 21 days. Then after, the control and fungal treated feeds were dried in an oven to a constant weight in order to stop fungi growth.

Adequate quantities of an inert form of EM (EM-1) packed in plastic bottles was purchased from Weljijie PLC (Debrezeit). Molasses was added and mixed with EM at equal proportion in order to initiate the microbial (EM) activity such as multiplication and metabolism activities and diluted with a mixture of chlorine free water and molasses (18:1 ratio per liter) (*Higa and Wididana, 2007*). After stirring, the mixture was sprayed over the SCB and RH until they achieve moisture content of 60%. After preparation of the cultures, five hundred gram of treated sugarcane bagasse and rice husk were packed in airtight polyethylene bag each in four replications and incubated at room temperature for21 days. Then, the samples were removed from the bag, dried in an oven at 60°C for 48 hours and used for determination of chemical compositions, *in-sacco* and *in-vitro* degradability of the treated feeds.

A 2*5 factorial treatment arrangements with 4 replications were used to study the effect of feed type and treatment method. Samples of SCB and RH treated with three white rot fungi (*Trichodermaviride Pleurotusflorida*, *Pleurotusostreatus)* and essential microbes. Hence, the treatment combination consisted of eight treated and two untreated feed samples (Table 1).

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Treatment	Fungal type				Feed	Replic		
	UT	EM	Tv	Pf	Po	SCB	RH	
T1	+					+		4
T2		+				+		4
T3			+			+		4
T4				+		+		4
T5					+	+		4
T6	+						+	4
T7		+					+	4
T8			+				+	4
T9				+			+	4
T10					+		+	4
UT=Untreated:EM=Effective microbes: T							Tv=	

Table 1.Treatment combination and layout for chemical composition and *in vitro* digestibility of treated sugar cane bagasse and rice husk

Trichodermaviride;Pf=Pleurotusflorida;Po=Pleurotusostreatus; SCB = Sugarcane bagasse; RH = Rice husk.

One half of the sample was ground to pass 1mm sieve size Wiley mill and used for chemical analysis. The DM and ash contents of the feed samples were determined following *AOAC (2002)*. The NDF, ADF and ADL were determined based on the method described by *VanSoest and Robertson (1985)*. Hemicelluloses and cellulose were calculated as NDF-ADF and ADF-ADL, respectively. The N content of the samples was determined by the micro-Kjeldahl method and CP was calculated as N X 6.25. Metabolizable energy per kilogram was determined indirectly by conversion factors from its *in-vitro* organic matter digestibility (*MAFF, 1984*) as ME (Mcal/kg DM) = 0.16*IVOMD.

The samples used for *in vitro* digestibility test was ground to pass through 1mm sieve size Wiley mill, labeled and transported to animal nutrition laboratory of Holeta Agricultural Research Center. The *In vitro* digestibility was determined according to *Tilley and Terry* (1963) two stage techniques for *in vitro* digestion of forage crops, as modified by *Van Soest and Robertson* (1985), where a second stage (HCL-pepsindigestion) was substituted by neutral detergent extraction to simulate true digestibility.

The results of feed sample chemical assay and *in vitro* digestibility of feeds were analyzed by using SAS software version 9.1 (SAS, 2008). When there was significant difference between means, the mean separation was made by

adjusting with Tukey honestly significant difference test. The model employed for the analysis is described below: Model: $yijk=\mu + ai + bj + a*bij + \epsilon ijk$ Where: Yijk= the dependent variables, μ =overall mean; ai= theith feed type; bj= thejth treatment method, a*b= The ijth interaction (between feed type and treatment method) ϵijk =random error.

Results

The chemical composition was significantly affected (p<0.01) by the interaction of feed and treatment types (Table 2). Biological treatments significantly(p<0.01) decreased organic matter (OM), neutral detergent fiber (NDF), acid detergent fiber (ADF) and acid detergent lignin (ADL) of the RH and SCB as compared to untreated samples. However, the treatments were significantly (p<0.01) improved crude protein (CP) content as compared to untreated samples. The crude protein content of sugarcane bagasse and rice husk was significantly (p<0.01) improved in all treatments as compared to the untreated sample, except for rice husk treated with EM. The treatments raised CP content from 7.90% in untreated to 10.5%, 10.5% and 11.35% for RH treated with T ν , Po and Pf, respectively while the CP content of sugar cane bagasse increased from 2.61% of untreated to 3.5%, 5.96%, 5.89% and 5.95% for sugarcane treated with EM, T ν , Po and Pf, respectively. The interaction show that increase in CP content of RH was higher when treated with Pf than the EM and the other WRF, while the improvement in CP content of SCB treated with all WRF is similar and higher than for EM treated SCB.

Factors	Levels	DM	ОМ	ASH	СР	NDF	ADF	ADL
	RH	91.94 ^b	80.24 ^b	19.76 ^a	9.47 ^a	58.34 ^b	38.44 ^b	13.59 ^b
Feed	SCB	94.22 ^a	91.76 ^a	8.24 ^b	4.76 ^b	81.74 ^a	59.89 ^a	14.35 ^a
	SEM	0.12	0.03	0.028	0.044	0.07	0.10	0.05
	P-value	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001
	U	93.97 ^a	87.12 ^a	12.88 ^c	5.26 ^c	76.35 ^a	53.65 ^a	14.60 ^a
	EM	93.92 ^a	86.94 ^a	13.06 ^c	5.21 ^c	71.54 ^b	48.47 ^b	14.31 ^a
	Pf	92.38 ^b	85.10 ^c	14.90 ^a	8.65 ^a	67.21 ^c	48.65 ^b	14.14 ^b
Treatments	Ро	92.54 ^b	85.34 ^b	14.66 ^b	8.25 ^b	67.38 ^c	48.25 ^b	13.68 ^c
	Tv	92.54 ^b	85.50 ^b	14.50 ^b	8.20 ^b	67.72 ^c	46.79 ^c	13.13 ^d
	SEM	0.22	0.048	0.047	0.077	0.12	0.17	0.09
	P-value	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001
	RH,U	93.16 ^b	80.89 ^e	19.11 ^c	7.90 ^c	65.82 ^d	44.39 ^d	14.08 ^{bc}
	RH,EM	92.53°	80.98 ^e	19.02 ^c	7.92 ^c	59.62 ^e	38.61 ^e	14.37 ^b
	RH,Pf	90.88 ^d	79.50 ^g	20.50 ^a	11.34 ^a	55.30 ^f	37.11 ^f	13.61 ^{cd}
	RH,Po	90.98 ^d	79.90 ^f	20.10 ^b	10.61 ^b	55.30 ^f	36.99 ^f	13.24 ^{de}
	RH,Tv	91.12 ^d	79.93 ^f	20.07 ^b	10.45 ^b	55.68 ^f	35.12 ^g	12.66 ^e
Feed * Treatments	SCB,U	94.79 ^a	93.35 ^a	6.65 ^g	2.61 ^g	86.88 ^a	62.92 ^a	15.12 ^a
	SCB,EM	92.31 ^c	92.90 ^b	7.10 ^f	3.41 ^f	83.46 ^b	58.33°	14.25 ^b
	SCB,Pf	93.88 ^b	90.69 ^d	9.31 ^d	5.95 ^e	79.12 ^c	60.20 ^b	14.67 ^{ab}
	SCB,Po	94.11 ^b	90.78 ^{cd}	9.22 ^{de}	5.89 ^e	79.46 ^c	59.52 ^{bc}	14.13 ^{bc}
	SCB,Tv	94.03 ^b	91.07 ^c	8.93 ^e	5.96 ^e	79.76 ^c	58.46 ^c	13.59 ^{cd}
	SEM	0.30	0.068	0.068	0.11	0.17	0.23	0.13
	P.value	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001

Table 2. Chemical composition (%DM) of sugarcane bagasse and rice husk as affected by treatment with white-rot fungi and effective microorganisms

^{a-g}LSMeans with different superscripts within the same column and factor are significantly different at P<0.01; SEM= Standard Error Mean;RH=Rice husk; SCB= Sugarcane bagasse; U=Untreated; EM= effective microorganism; Po=Pleurotusostreatus; Pf.=Pleurotusflorida; Tv.=Trichodermaviride; RHU= untreated Rice husk; RHPo.= Rice husk treated with Pleurotusostreatus; RHPf.= Rice husk treated with Pleurotusflorida; RHTv.=Rice husk treated with Trichodermaviride; RHEM =Rice husk treated witheffective microorganism; SCBU= untreated Sugarcane bagasse; SCBPo.= Sugarcane bagasse treated with Pleurotusostreatus; SCBPf.= Sugarcane bagasse treated with Pleurotusflorida; SCBTv.= Sugarcane bagasse treated with Trichodermaviride; SCBEM = Sugarcane bagasse treated with effective microorganism; DM=Dry matter; OM=Organic matter; CP=Crude protein; NDF=Neutral detergent fiber; ADF= Acid detergent fiber; ADL=Acid detergent lignin.

In-vitro digestibility, cellulose, hemicelluloses, and Metabolizable energy content of treatments are significantly (p<0.01) affected by interaction of feed and biological treatment types (Table 3). Biological treatments significantly (p<0.01) reduced cellulose and hemicelluloses content of RH and SCB, while *in-vitro* organic matter digestibility, dry matter digestibility and Metabolizable energy content were significantly (p<0.01) increased as compared to untreated RH and SCB. Large decrease in cellulose and hemicelluloses content of RH and SCB occurred when treated with Tv and EM, respectively. Similarly, large improvement in IVOMD of RH and SCB were recorded when treated with Tv and EM, respectively. Improved IVOMD resulted in leads increased Metabolizable energy from 4.5MJ/kg in untreated to 7.5MJ/kg in Tv. treated RH and from 4MJ/kg in untreated to 6MJ/kg in EM treated SCB.

Table 3. Cellulose, hemicelluloses, *invitro* digestibility and Metabolizable Energy content of sugar cane bagasse and rice husk treated with white rot fungi and essential microbes

Factors	Levels	Cellulose	Hemi-Cellulose	IVDMD	IVOMD	ME MJ/Kg DM	
	RH	24.85 ^b	19.90 ^b	36.01 ^a	37.59 ^a	6.01 ^a	
Food	SCB	45.53 ^a	21.85 ^a	29.27 ^b	30.59 ^b	4.89 ^b	
reeu	SEM	0.12	0.08	0.12	0.11	0.02	
	p-value	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	
	U	39.05 ^a	22.69 ^a	27.53 ^d	28.73 ^d	4.59 ^a	
	EM	34.16 ^{cb}	23.07 ^a	35.35 ^b	36.88 ^b	5.9 ^b	
Treatments	Pf	34.51 ^{cb}	18.56 ^c	31.65 ^c	33.17 ^c	5.3 ^c	
	Ро	34.57 ^b	19.12 ^c	31.8 ^c	33.11 ^c	5.29 ^c	
	Tv	33.66c	20.93 ^b	36.88 ^a	38.57 ^a	6.17 ^a	
	SEM	0.2	0.13	0.2	0.19	0.03	
	p-value	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	
	RH,U	30.31 ^c	21.43 ^c	26.89 ^{ef}	28.09 ^{ef}	4.49 ^{ef}	
	RH, EM	24.24 ^d	21.01 ^c	35.74 [°]	37.14 ^c	5.94 ^c	
	RH ,Pf	23.5 ^{de}	18.19 ^e	37.42 ^b	39.35 ^b	6.3 ^b	
	RH, Po	23.75 ^{de}	18.32 ^{cd}	35.09 ^c	36.39 ^c	5.82 ^c	
	RH ,Tv	22.46 ^e	20.56 ^c	44.94 ^a	47.02 ^a	7.52 ^a	
Easd*Tractments	SCB,U	47.80^{a}	25.13 ^a	28.17 ^{de}	29.36 ^{de}	4.7 ^{de}	
reeu* meannents	SCB,EM	44.08^{b}	24.96 ^a	34.97 ^c	36.64 ^c	5.86 ^c	
	SCB, Pf	45.52 ^b	18.93 ^e	25.88 ^f	26.99 ^f	$4.32^{\rm f}$	
	SCB, Po	45.39 ^b	19.94 ^e	28.53 ^d	29.84 ^d	4.77 ^d	
	SCB,Tv	44.87 ^b	21.30 ^c	28.83 ^d	30.13 ^d	4.82 ^d	
	SEM	0.28	0.19	0.28	0.27	0.04	
	p-value	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	

^{a-g}LSMeans with different superscripts within in the same column and factor are significantly different at P<0.05. SEM= Standard Error Mean, RH=Rice husk, SCB= Sugarcane bagasse, U=Untreated EM= effective microorganism, Po=Pleurotusostreatus, Pf.=Pleurotusflorida, Tv.=Trichodermaviride, RHU= untreated Rice husk; RHPo.= Rice husktreated withPleurotusostreatus; RHPf.= Rice husk treated withPleurotusflorida; RHTv.=Rice husk treated withTrichodermaviride; RHEM =Rice husk treated witheffective microorganism; SCBU= untreated sugarcane bagasse; SCBPo.= Sugarcane bagassetreated withPleurotusostreatus; SCBPf.= Sugarcane bagassetreated withPleurotusflorida; SCBTv.= Sugarcane bagassetreated withTrichodermaviride; SCBEM = Sugarcane bagassetreated witheffective microorganism; IVOMD=In-vitro organic matter digestibility, IVDMD= In-vitro dry matter digestibility, ME= Metabolizable energy.

Discussion

The biological treatment on average reduced ADF, NDF and ADL from 55.5 to 47%, 79 to 67 % and 14.5 to 13%, respectively, which was significant among treatments with the lowest cell wall components recorded for feed treated with Trichodermaviride. Similar to the result of the present study, Hassan et al. (2015) noted that treatment of rice husk with P. ostreatus decreased the cell wall components as compared to untreated. Salman et al. (2011) reported that treatment of sugarcane bagasse with fungi, yeast and bacteria decreased the cell wall components. Abdel-Azim et al. (2011) stated that treatment of rice straw and corn stalks by Trichodermaviride decreased NDF and ADF. Similarly, Baraghit et al. (2009) reported that biological treatments with different fungal and bacteria strains decreased cell wall constituents of different crop residues. The decrease in cell wall components might be due to the breakdown of lignocelluloses bonds resulting into hydrolysis of cellulose by fungi and bacteria (El-Ashrvet al., 2002; El-Shafie et al., 2007; Fayed et al., 2009 and Mahrous et al., 2010). Fazaeli et al. (2004) noted that fungi treatment solubilize and utilize the cell wall components as carbon source and thus change the ratio of insoluble to soluble carbohydrates in the by-products. The result of treatment inoculated with EM agree with the finding of Yonatan (2010) who reported that treatment of coffee husk with EM decreased the cell wall components as compared to the untreated husk. Similarly, Mullgeta (2015)noted inoculating crop residues with EM reduced cell wall components as compared to untreated crop residues.

The CP content of feed samples on average increased from 4.61 to 8.42% with highest value recorded in feed treated with white rot fungi species. This result is in agreement with *Shoukry et al.* (1985) who noted that treatment of sugarcane bagasse with different microorganisms leads to increased CP and ash content as compared to the untreated treatment. *Nasehi et al.*(2013) noted that fermentation of barley and wheat straw with *Pleurotusflorida* decreased the cell wall components and increased the CP content. *Yonatan* (2010) and Mulgeta (2015) reported that EM treatment improve the CP content of roughage as compared to untreated. The increased crude protein content when feed is treated with biological media is attributed to the growth and production of mycelium (*Ragunathan et al.*, 1996). Mycelium relatively contain high protein, hence it is expected that the treated by-products containing fungal mycelium to have a higher

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concentration of CP. There is secretion of certain proteineous extra cellular enzymes into the waste during breakdown and their subsequent metabolism (*Kadiri, 1999; Akinfemiet al., 2009*), which also adds to the protein content of the treated feed. Moreover, the increased CP could be due to the capture of excess nitrogen by fermentation (*Sallam et al., 2007*). Increased CP content in general suggests that the treated feed could be a good source of protein for livestock.

Significantly higher improvement in IVOMD, IVDMD and ME were recorded for feed treated with Trichodermaviride followed bv EM. *Pleurotusostreatus* and Pf. The higher digestibility of lignocelluloses by-products treated with WRF and EM could mainly be attributed to lower cell wall components and higher CP content of biological treated by-products due to the action of microorganisms during fermentation. Surinder and Suman(1986) reported that *Pleurotusostreatus* and *S. pubverulentum* used as biological treatment of paddy straw increased IVDMD. Shah and Rehman (1988) noticed increased IVDMD when cotton seed hulls were fermented by *Bacillus polymexa* and Trichodermaviride as compared to unfermented. Bassuny et al. (2003) found that IVDMD and IVOMD ofrice and bean straw treated with biological treatment were significantly improved compared to the control. Yonatan (2010) noted that treatment of coffee husk with EM improve the IVOMD. Similarly, *Mulgeta* (2015) reported that treatments of different crop residues resulted in to improvement of IVOMD as compared to untreated roughages.

Conclusion

The results of the experiment indicate that biological treatments can improve the nutritive value of rice husk and sugarcane bagasse through decreasing cell wall content, improving percent crude protein and *in-vitro* dry matter digestibility. The results of fermentation characteristics of treatments suggested that the best biological treatment is obtained from feed treated with EM. The best result of IVOMD, IVDMD and ME were achieved for rice husk treated with Tv. and sugarcane bagasse treated with EM. The overall result implies that biologically treated sugarcane bagasse and rice husk can be incorporated into other ruminants' diet for better productivity. We also suggest *in-vivo* metabolism and feeding trial study on Tv. and EM treated rice husk and sugarcane bagasse for more complete information.

Hemijski sastav i *in-vitro* svarljivost šećerne trske i ljuske pirinča tretirane sa tri soja gljivica bele truleži i delotvornim mikroorganizmima

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Rezime

Sprovedena je studija za procenu uticaja biološkog tretmana šećerne trske (SCB) i pirinčane ljuske (RH) sa tri soja gljivice bele truleži (VRF) (*Pleurotusostreatus* (Po), *Pleurotusflorida* (Pf) i *Trichodermaviride* (Tv)) i efikasnim mikroorganizmima (EM) na hemijski sastav i in-vitro svarljivost. Eksperiment se sastojao od 2x5 faktorijalnog ogleda, dva nivoa hraniva (SCB i RH) i pet nivoa bioloških tretmana (kontrola, Po, Pf, Tv i EM). Tretman RH sa EM, Tv, Po i Pf, značajno povećava sadržaj sirovog proteina od 7,90% u netretiranom do 7,92; 10,46; 10,61 i 11,35%, respektivno. Odgovarajući porast sadržaja sirovog proteina šećerne trske od 2,61% bio je 3,41; 5,96; 5,89 i 5,95%. Tretmani su značajno (P <0,001) smanjili NDF, ADF, ADLC i hemicelulozu sa najnižom vrednošću evidentiranom za tretman Tv. IVOMD, IVDMD i metabolička energija (ME) su bile značajno (P <0,001) povećane. U zaključku, studija ukazuje da je tretman RH sa *Trichodermaviride* i SCB sa EM efikasniji od drugih u poboljšanju nutritivne vrednosti krmiva. Predlažemo procenu tretiranih krmiva prema proizvodnim performansama realizovanim u stočarskoj proizvodnji.

Ključne reči: *In-vitro* svarljivost, gljivice bele truleži, efikasan mikroorganizam, šećerna trska, pirinač

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References

ABDEL-AZIM S.N., AHMED M.A., ABO-DONIA F., SOLIMAN H. (2011): Evaluation of fungal treatment of some agricultural residues. Egyptian Journal of Sheep Goat Science, 6, 1-13.

AKINFEMI A., ADU O.A., DOHERTY F. (2009): Conversion of sorghum stover into animal feed with white-rot fungi: *Pleuro-tusostreatus* and *Pleurotuspulmonarius*. African Journal of Biotechnology, 9 (11), 1706-1712.

AOAC.(2002): Official Methods of Analysis 16th Ed. Association of Official Analytical Chemists, Arlington, VA.

BARAGHIT G.A., AHMED B.M., EL-MAHY M.A. (2009): Digestibility, nutritive value and rumen fermentation of rice straw and sugar cane bagasse treated with a commercial bacterial culture. Egyptian Journal of Nutrition and Feeds, 12 (3), 511-522.

FAYED AFAF M., EL-ASHRY M.A., AZIZ HEND A. (2009): Effect of feeding olive tree pruning by-products on sheep performance in Sinai. World Journal Agricultural Sciences, 5 (4): 436-445.

FAZAELI H., MAHMODZADEH H., JELAN Z.A., ROUZBENHAN Y., LIANG J.B., AZIZI A. (2004): Utilization of fungal treated wheat straw in the diet of late lactating cow. Asian-Australian Journal of Animal Science, 17, 467-472.

HASSAN A.A., ALSAMAREE W.H., ABBAS E.R, FENJAN K. (2015): Effect of some chemical and biological treatment of rice hulls (subose) on chemical composition and in vitro digestibility. Journal of International Academic research for multidisciplinary, 2, 2320-5083.

HIGA T., WIDIDANA G.N. (2007): The Concept and theory of Effective Microorganism.

ISROI MILLAT R., SYANSIAH S., NIKLASSON C., CAHYANTO M.N., LUNDQUIST K., TAHERZADEH M.J.(2011): Biological Pretreatment of Lingocelluloses with white rot fungi and its application. Bioresources, 6 (4), 5224-5259.

JAHROMI M.F., LIANG J.B., ROSFARIZAN M., GOH Y.M., SHOKRYZADEH P., HO Y.W. (2010): Effects of *Aspergillusniger* (K8) on nutritive value of rice straw. African Journal of Biotechnology, 9, 7043-7047.

JALC D. (2002): Straw enrichment for fodder production by fungi. In: The Mycota XI Agricultural Applications (Ed. F. Kempken). Springer-Verlag, Berlin, Heidelberg. pp. 19-38.

JAFARI M.A., NIKKHAH A., SADEGHI A.A., CHAMANI M.(2007): The effect of *Pleurotusspp*. fungi on chemical composition and *in vitro* digestibility of rice straw. Pakistan. Journal of BiologicalScience, 10: 2460-2464.

KADIRI M. (1999): Changes in intracellular and extracellular enzymes activities of *Lentinussubnudus* during sporophore development. Bio scientific Research Communications, 11(2), 127-130.

KEWARAMANI N., KAMARA D.N., LALL D., PHATHAK N.N. (1988): Bioconversion of sugarcane bagasse with white-rot fungi. Bioltechnology letters, 10 (5), 369-372.

MAHROUS A.A., KHORSHED M.M., HAFEZ Y.H. (2010): Effect of biological treatment on improvement of sugar cane bagasse, nutritive value and its effect on productive performance of lactating Buffaloes. Egyptian Journal of Nutrition and Feeds, 13(2): 245-257.

MULGETA A. (2015): Evaluation of effective microbes (EM) treatment on chemical composition of crop residues and performance of crossbred dairy cows. MSc. Thesis, College of Agriculture and Natural Resources. Haramaya University.

NASEHI M., TORBATINEJAD N.M., ZEREHDARAN S., SAFAEI A.R. (2013): Effect of (*Pleurotusflorida*) Fungi on Chemical Composition and Rumen Degradability of Wheat and Barley Straw. Iranian Journal of Applied Animal Science, 4(2), 257-261

RAGUNATHAN R., GURUSAMY R., PALANISWAMY M., SWAMINATHAN M. (1996): Cultivation of *Pleurotus spp*. on various agro-residues. Food Chemistry, 55, 139-144.

SAS INSTITUTE. (2008): SAS/STAT Software, Release 9.1. SAS Institute, Inc., Cary, NC.

SALLAM S.M.A., NASSER M.E.A., EL-WAZIRY A.M., BUENO F.C.S., ABDALLAH A.L. (2007): Use of yam *in vitro* rumen gas production technique to evaluate some ruminant feedstuffs. Journal Applied Science Research, 3(1), 34-41.

SALMAN F. M., SALAMA R., KHATTAB A .E., SOLIMAN S .M., EL-NOMEARY Y .A. (2011). Chemical, biological and biochemical treatments to improve the nutritive values of sugarcane bagasse (SCB): 1- Chemical composition, scanning electron microscopy, *in vitro* evaluation, nutrients digestibility and nitrogen utilization of untreated or treated SCB. Journal of Life Science, 8, 361-363.

SHAH F. H., REHMAN Z. (1988): Nutritive value of cotton seed hulls after

biological treatment. Pakistan Journal of Sciences and Industry Research, 31, 425.

SINGH G. B., GUPTA B. N., SINGH K.(1990): Effect of microbial treatment of paddy straw on chemical composition and nutrient utilization in crossbred goats. Indian Journal of Animal Nutrition, 7 (4), 251 - 256.

SURINDER S. K., SUMAN K. D.(1986): Biological conversion of paddy straw into feed. Journal of Biological wastes, 22, 11.

TILLEY J.M.A., TERRY R A.(1963): A two-stage technique for the *in vitro* digestion of forage crops. Journal of British Grassland Society,18 (2),104-111.

TRIPATHI J.P., YADAV J.S. (1992): Optimization of SSF of wheat straw into animal feed by *Pleurotus ostreatus*: A pilot effort. Animal Feed Science and Technology, 37, 59–72.

TRIPATHI M.K., MISHRA A.S., MISRA A.K., VAITHIYANATHAN S., PRASAD R., JAKHMOLA R.C. (2008): Selection of white-rot basidiomycetes for

bioconversion of mustard (Brassica compestris) straw under solid-state fermentation into energy substrate for rumen micro-organism. Letters in Applied Microbiology, 46, 364–370.

VAN SOEST P.J., ROBERTSON J.B. (1985): Analysis of forages and fibrous foods. AS 613 Manual Department of Animal Science, Cornell University, Ithaca, NY.

YONATAN K. Y. (2010): Chemical composition and *in vitro* digestibility of coffee pulp and coffee husk ensiled with grass (*hyperchenniahirta*) hay and effective microorganism (EM) M.Sc.Thesis, College of Agriculture Jimma University, Jimma.

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