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Effect of (*Pleurotus florida*) Fungi on Chemical Composition and Rumen Degradability of Wheat and Barley Straw

Research Article

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ABSTRACT

This study was conducted to investigate the effect of biological treatment with *Pleurotus florida* fungi on chemical composition and rumen dry matter (DM) and organic matter (OM) degradability of wheat and barley straw. Wheat and barley straw were collected from Golestan province of Iran and after pasteurization mixed with fungi spawn (than 5% by weight). After 21 days samples were dried in oven (60 ˚C) in order to stop fungi growth. Dry matter (DM) and organic matter (OM) degradation determined using nylon bag technique with two fistulated Dalagh rams and samples were incubated at zero to 96 hours in rumen. Fungi cultivation significantly decreased the amount of dry matter (DM), Neutral detergent fiber (NDF) and acid detergent fiber (ADF) in wheat and barley straw ($P<0.01$). Ash and crude protein (CP) content significantly increased with processing by fungi in treatments ($P<0.01$). Ether extract (EE) and acid detergent lignin (ADL) content did not differ by processing with fungi. The soluble fraction (a) and potential degradability (a+b) of dry matter (DM) increased ($P<0.05$) by processing with fungi in both straw. The insoluble but potentially degradable fraction (b) of dry matter (DM) degradability increased ($P<0.05$) by processing with fungi in wheat straw, but did not change in Barley straw. The a and a + b fractions of organic matter (OM) degradability increased ($P<0.01$) by processing in both straw. The b fraction of organic matter (OM) degradability increased ($P<0.01$) by processing with fungi in wheat straw, but did not observed difference in Barley straw. Therefore, it has been concluded that the treating the wheat and barley straw with fungi improved the nutritive value in the present study.

KEY WORDS fungi, nutritive value, nylon bag, straw.

INTRODUCTION

There are a lot of agricultural residues annually produced in countries all over the world including Iran. The large portion of these residues is important feed stuff for ruminants and can be used as a potentially important source of carbohydrates and energy. However, the utilization of these materials as a feed source for ruminants is limited for their complex biological structure and low protein content (Rodrigues *et al*. 2008; Yalchi and Hajieghrari, 2011). The low ability of lignocelluloses to hydrolyze (more for crystalline structure of cellulose fibrils and presence of lignin) reduces the digestibility and restricts efficient utilization of the feed produced by ruminal microorganisms. Although, microorganisms within the rumen are able to exude enzymes that have potential to directly hydrolyze cellulose and hemicelluloses in the rumen (Yalchi and Hajieghrari, 2011). The complex network formed by cellulose, hemicelluloses and lignin reduces their digestibility because of lacking ligninolytic activity (Falcon *et al*. 1995; Otjen *et al*. 1987; Zadrazil *et al*. 1985). Therefore, they are not very efficient in order to break down the lignocellulosic bond of

straw. The various methods that could increase its nutritive value through physical and chemical processing have been studied (Matsuzaki *et al*. 1994; Rahal *et al*. 1997). Although these methods have advantages, they are costly, low in effectiveness, not environmentally friendly and also require application of technology (Leng, 1991; Sharma *et al*. 1993). These factors limit their application, particularly at small farm levels. Recently, biological delignification of straw by solid-state fermentation (SSF) has been considered because of its capacity to remove lignin preferentially (Moyson and Verachtert, 1991).

Fungal treatment could be an approach to convert low quality crop residues into a higher quality of ruminant feed (Arora *et al*. 1994; Zadrazil *et al*. 1996). Attempts had been made to identify species of white-rot fungi for their ability to grow on straws that improved their nutritive value (Yamakawa *et al*. 1992).

During the SSF of wheat straw by fungi, its OM and detergent fiber content could be reduced and the lignin selectively removed from the lignocellulosic complex (Singh *et al*. 1990; Kundu, 1994). However, such changes were dependent on the strain of fungi and the cultural conditions (Tripathi and Yadav, 1992). Among the edible white-rot fungi, the *Pleurotus* (*P*) species have been shown to be more efficient (Zadrazil *et al*. 1996). The potential of some species of *Pleurotus* fungi such as *Pleurotus ostreatus* and *Pleurotus eryngii* to reduce indigestible cell wall components and increase dry mater digestibility (DMD) of straw has been reported (Agosin *et al*. 1986; Singh *et al*. 1990). Despite much research has been done about effect of fungal treatment on nutritive value of agro-by products, there is little information about effect of some species. Therefore, this study conducted in order to investigate the effect of biological treatment with *Pleurotus florida* fungi on chemical composition and rumen DM and OM degradability of wheat and barley straw.

MATERIALS AND METHODS

Treatment of straws and chemical analysis

Wheat and barley straw were collected from local farms in Gorgan, Golestan province, Iran. *Pleurotus florida* fungi were used for treating wheat and barley straw. Wheat and barley straw treatment was carried out in 1000 mL bottles. The straw of each 50 g was placed in individual bottle and water added to give moisture content of about 85%. The bottles autoclaved at 121 ˚C for 20 min. Each bottle was inoculated with 5% (w/w) spawns of *Pleurotus florida* fungi (Jahromi *et al*. 2010).

Each treatment was four replicate. The bottles incubated in an incubator where temperature was automatically adjusted to 25 ˚C and relative humidity was kept at 78 ± 5%.

After 21 days, samples were dried in oven (60 ˚C) in order to stop fungi growth and then the chemical composition of rumen DM and OM degradability were determined. DM determined by drying the samples at 105 ˚C overnight and Ash by igniting the samples in muffle furnace at 550 ˚C for 8 h. EE was determined by soxhlet extraction method (AOAC, 2002).

NDF, ADF and ADL content was measured by Fiber-Tec system (Vansoest *et al*. 1991). Nitrogen (N) content was measured by the Kjeldahl method and CP was calculated as N × 6.25 (AOAC, 2002).

In situ degradation procedures

Two ruminally cannulated Dalagh rams (about 45 kg BW) were used to determine *in situ* degradation characteristics. Rams were housed in individual tie stalls bedded with sawdust. Rams fed diets containing wheat straw (70%) and concentrate mixture (30%) at the maintenance levels (NRC, 1985). The concentrate mixture contained barley grain and wheat bran. Dacron bags (40-45 micron pore size) were filled with 3 g dried and ground samples then incubated in the rumen of rams for the periods of 0, 4, 8, 12, 24, 48, 72 and 96 h. Two bags incubated per each presented time for each sample in each sheep. Bags were removed, washed and dried according to the procedure of Orskov *et al*. (1980). Rumen degradation kinetics of DM and OM was fitted by the nonlinear model proposed by Orskov and McDonald (1979) using Fitcurve software version 6 (Chen, 1995).

$$P= a + b (1-e^{-ct})$$

Where:
P: percentage of degradability for response variables at t.
t: time relative to incubation (h).
a: soluble fraction (%).
b: insoluble but potentially degradable fraction (%).
c: rate constant for degradation (h^{-1}).
e: 2.7182 (Natural logarithm base).

Following determination of these parameters, the effective degradability of DM and OM in the samples was calculated using and equation described by Orskov and McDonald (1979):

$$ED= a + (bc) / (c+k)$$

Where:
ED: effective degradability for response variables (%).
a: soluble fraction (%).
b: insoluble but potentially degradable fraction (%).
c: rate constant for degradation (h^{-1}).

k: rate constant of passage (h^{-1}).

Statistical analysis

For all data, a completely randomized design with a 2 × 2 factorial arrangement was used. The experimental factors were straw type at 2 levels (wheat and barley straw) and fungus application at 2 levels (Control and *Pleurotus florida* fungi). Each parameter was measured with four replicates.

The data obtained were analyzed for parametric statistics, including analysis of variance using the general linear model procedure (GLM) and the differences among treatments' means were compared by Tukey's test. SAS (2002) was used for statistical analyss.

RESULTS AND DISCUSSION

Chemical composition

The nutrient composition of experimental treatments is presented in Table 1. Treating with fungi decreased (P<0.01) the amount of DM, NDF and ADF in treatments. Whereas, Ash and CP content increased (P<0.01) with processing with fungi in both straw. EE and ADL content of treatment did not differed by processing with fungi.

Table 1 Effect of fungal treatment on chemical composition (%) of experimental treatments

Items	UWS	TWS	UBS	TBS	P-value	SEM
DM	92.60^{b}	84.53^{c}	93.49^{a}	86.53^{d}	0.005	0.43
Ash	6.41^{c}	9.27^{a}	7.20^{c}	8.26^{b}	0.0008	0.20
EE	0.87^{a}	0.79^{a}	1.16^{a}	1.15^{a}	0.8	0.13
CP	3.71^{b}	7.38^{a}	3.14^{b}	8.23^{a}	0.002	0.65
NDF	77.49^{a}	69.43^{b}	77.17^{a}	68.09^{b}	0.009	0.85
ADF	56.48^{a}	48.24^{b}	54.92^{a}	44.57^{b}	0.0008	0.71
ADL	10^{a}	8.96^{a}	7.37^{a}	6.93^{a}	0.7	1.02

UWS: untreated wheat straw; TWS: treated wheat straw; UBS: untreated barely straw and TBS: treated barely straw.
DM: dry matter; EE: ether extract; CP: crude protein; NDF: neutral detergent fiber; ADF: acid detergent fiber and ADL: acid detergent lignin.
The means within the same row with at least one common letter, do not have significant difference (P>0.01) and (P>0.05).
SEM: standard error of means.

***In situ* degradation**

The DM degradability of experimental treatments is presented in Table 2. Fungal treatment increased (P<0.01) DM degradation at 96 h in both straw. The data of OM degradability is presented in Table 3. Fungal treatment increased (P<0.01) OM degradation at 96 h in both straw. The degradability parameters of the DM obtained from the fitted values are presented in Table 4.

The a and a + b fractions of DM degradability increased (P<0.05) with processing by fungi in both straw. The b fraction of DM degradability increased with processing by fungi in wheat straw (P<0.05), but did not significant change in barley straw. DM effective degradability of the experimental treatments at 2 and 8 percent out flow rate increased (P<0.01) with fungal treatment in both straw.

Table 2 Ruminal dry matter degradation (%) of treatments at different incubation times

Incubation time (h)	UWS	TWS	UBS	TBS	P-value	SEM
0	11.45^{d}	21^{b}	14.20^{c}	23.70^{a}	0.0001	0.82
4	15.67^{d}	27^{b}	17.47^{c}	30.85^{a}	0.0001	0.77
8	19.70^{d}	28.92^{b}	22.32^{c}	33.65^{a}	0.0001	0.80
12	22.30^{d}	31.87^{b}	25.75^{c}	36.10^{a}	0.0001	1.22
24	29.77^{c}	33.12^{bc}	35.05^{b}	39.87^{a}	0.0003	1.58
48	35.15^{c}	35.52^{c}	40.42^{b}	50.07^{a}	0.0001	1.48
72	37.32^{c}	41.02^{b}	41.90^{b}	55.20^{a}	0.0001	1.09
96	43.12^{c}	47.62^{b}	49^{b}	59.27^{a}	0.0001	0.71

UWS: untreated wheat straw; TWS: treated wheat straw; UBS: untreated barely straw and TBS: treated barely straw.
The means within the same row with at least one common letter, do not have significant difference (P>0.01).
SEM: standard error of means.

Table 3 Ruminal organic matter degradation (%) of treatments at different incubation times

Incubation time (h)	UWS	TWS	UBS	TBS	P-value	SEM
0	10.79^{d}	19.53^{b}	15.54^{c}	24.96^{a}	0.0001	0.72
4	16.01^{d}	24.93^{b}	19.94^{c}	31.18^{a}	0.0001	0.80
8	23.92^{c}	27.94^{b}	26.61^{b}	34^{a}	0.0001	0.77
12	25.73^{c}	30.70^{b}	30.89^{b}	36.49^{a}	0.0001	0.77
24	38.32^{b}	34.73^{c}	46.03^{a}	40.32^{b}	0.0001	1.03
48	41.31^{b}	41.05^{b}	49.23^{a}	51.63^{a}	0.001	1.57
72	45.25^{c}	48.07^{c}	51.28^{b}	57.33^{a}	0.001	1.34
96	48.67^{c}	55.13^{b}	55.87^{b}	62.51^{a}	0.0001	0.74

UWS: untreated wheat straw; TWS: treated wheat straw; UBS: untreated barely straw and TBS: treated barely straw.
The means within the same row with at least one common letter, do not have significant difference (P>0.01).
SEM: standard error of means.

Table 4 Ruminal dry matter degradation parameters and effective degradability of treatments

Items	UWS	TWS	UBS	TBS	P-value	SEM
a (%)	12.12^{b}	24.95^{a}	13.90^{b}	26.15^{a}	0.6	0.58
b (%)	31.17^{b}	35.87^{a}	33.82^{ab}	36.85^{a}	0.3	0.87
a+b (%)	43.32^{c}	60.82^{a}	47.72^{b}	63.02^{a}	0.2	0.93
c (h^{-1})	0.03^{a}	0.01^{b}	0.03^{a}	0.02^{ab}	0.3	0.004
ED= 0.02	30.97^{c}	36.32^{b}	35.42^{b}	45.62^{a}	0.0007	0.53
ED= 0.05	24.10^{d}	30.60^{b}	27.90^{c}	37.57^{a}	0.02	0.86
ED= 0.08	20.95^{d}	28.70^{b}	24.30^{c}	34.22^{a}	0.0001	0.80

UWS: untreated wheat straw; TWS: treated wheat straw; UBS: untreated barely straw and TBS: treated barely straw.
a: soluble fraction; b: insoluble but potentially degradable fraction; a + b: potential degradability; c: rate of degradation of fraction b (h-1), ED: effective degradability in out flow rates (0.02, 0.05 and 0.08) / h.
The means within the same row with at least one common letter, do not have significant difference (P>0.01) and (P>0.05).
SEM: standard error of means.

Also fungal treatment increased (P<0.05) the amount of DM effective degradability at 5% out flow rate. The a and a + b fractions of OM degradability significantly increased (P<0.01) by processing in both straw. The b fraction of OM degradability significantly increased with processing by fungi in wheat straw (P<0.01), but did not significant difference in barley straw.

OM effective degradability of experimental (in all out flow rates) treatments significantly increased (P<0.01) by fungal treatment in both straw (Table 5).

Table 5 Ruminal organic matter degradation parameters and effective degradability of treatments

Items	UWS	TWS	UBS	TBS	P-value	SEM
a (%)	10.46^{d}	22.20^{b}	14.29^{c}	27.07^{a}	0.0001	0.38
b (%)	36.66^{b}	44.34^{a}	44.77^{a}	43.01^{a}	0.0005	1.42
a + b (%)	47.12^{c}	66.55^{a}	59.07^{b}	70.09^{a}	0.001	1.47
c (h^{-1})	0.05^{a}	0.01^{c}	0.04^{b}	0.01^{c}	0.0001	0.001
ED= 0.02	36.62^{d}	39.65^{c}	44.80^{b}	47.15^{a}	0.0001	0.33
ED= 0.05	28.80^{d}	31.35^{c}	34.90^{b}	38.25^{a}	0.0001	0.30
ED= 0.08	24.55^{d}	28.40^{c}	29.90^{b}	34.85^{a}	0.0001	0.29

UWS: untreated wheat straw; TWS: treated wheat straw; UBS: untreated barely straw and TBS: treated barely straw.
a: soluble fraction; b: insoluble but potentially degradable fraction; a + b: potential degradability; c: rate of degradation of fraction b (h-1), ED: effective degradability in out flow rates (0.02, 0.05 and 0.08) / h.
The means within the same row with at least one common letter, do not have significant difference (P>0.01) and (P>0.05).
SEM: standard error of means.

Chemical composition

The protein content of the mycelium was reported relatively high (Ragunathan *et al*. 1996), so it was expected that the treated straw, that contained fungal mycelium to have a higher concentration of CP. An increase of CP content in wheat straw incubated with *Pleurotus species* had also been reported (Ardon *et al*. 1996; Zadrazil *et al*. 1996; Fazaeli *et al*. 2004). The increase in the CP contents may be due to secretion of certain extra cellular enzymes which are proteineous in nature into the waste during their breakdown and its subsequent metabolism (Kadiri, 1999; Akinfemi *et al*. 2009). CP increase could also be due to the capture of excess nitrogen by fermentation (Sallam *et al*. 2007), suggesting that the treated substrates are good source of protein for livestock. This agrees with the findings of Zadrazil (1993), Belewu and Okhawere (1998) and Iyayi and Aderolu (2004). The NDF content of wheat and barley straw decreased by fungal treatment. This might be due to the natural habitats of the white-rote fungi that largely depend on organic carbon for their energy requirement form of structural material such as lignocellulosic (Jennings and Lysek, 1996). The losses of NDF from the straw suggested that these fungi could solubilize and utilize the cell wall as carbon source and thus changed the ratio of insoluble to soluble carbohydrates in the straw (Fazaeli *et al*. 2004). The decrease in NDF contents of the treated straw has been supported by other reports (Yalchi and Hajieghrari, 2011).

In situ degradability

Fungal treatment increased DM and OM degradability at 96 h in both straws. The reason for such improvement in the degradability might be due to the breaking down of cell wall bonds during the digestion of straw with the fungi (Call and Mücke, 1997; Fazaeli *et al*. 2004). Rumen degradability of both straws significantly increased by fungal treatment. These higher values of degradability in fungal treated straws explain the fact that the lower cell wall components and higher soluble fraction in both straws (Valmaseda *et al*. 1991) and (Valmaseda *et al*. 1991; Gutierrez *et al*. 1996).

CONCLUSION

In conclusion, treatment of wheat and barley straw with *Pleurotus florida* fungi, resulted in a reduction of the cell wall components and increased CP content and its degradability. The results obtained in this study suggest that the treatment of wheat and barley straw by application of fungi will help in conversion of agricultural wastes to higher quality ruminant feed thereby enhancing their digestibility by ruminants.

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