Improvement of Nutritive Value of Cereal Straw by Solid State Fermentation Using *Pleurotus Ostreatus*

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Abstract

A series of experiments was conducted to study the effects of white rot fungal treatment of cereal straw on lignin degradation, dry matter digestibility by cellulase (Ce-DMD) and digestible organic matter intake (DOMI) in sheep. Four strains (Hi80-2, Hi76-3, IPB No. 53 and TMI30026) of *Pleurotus ostreatus* were used for the experiments. After 60 days of fermentation (Hi80-2) of rice straw, the content of acid-detergent lignin (ADL) decreased rapidly and Ce-DMD increased from 31 to 52 %. Supplementation of rice bran to the straw culture did not affect appreciably the ADL content and Ce-DMD. Treatment of rice straw, wheat straw and soybean straw with IPB No. 53 decreased the ADL content and increased Ce-DMD remarkably. After 135 days (Hi76-3 and TMI30026) and 140 days (IPB No. 53) of fermentation of rice straw, DOMI in sheep fed treated straw was higher than that in the animals fed untreated straw. Treatment with TMI30026 increased DOMI by 58 %. These results indicate that solid state fermentation with *Pleurotus ostreatus* improves the nutritive value of cereal straw.

Introduction

A large amount of cereal straw is produced every year, which could become a potential source of ruminant feed. The nutritive value of the straw, however, is very low due to the low digestibility and low voluntary intake. A number of attempts including treatments with sodium hydroxide and ammonia have been made to improve the nutritive value of cereal straw (Sundstøl and Owen, 1984 ; Okamoto and Abe, 1989 ; Okamoto and Abe 1990 ; Okamoto and Miyazaki, 1990). There are additional possibilities to improve the nutritive value of cereal straw by using microorganisms with lignin degradation ability (Zadrazil, 1984). Research in this field has been active within the member countries of the European Communities (EC) and a workshop sponsored by the Commission of EC was held (Zadrazil and Reiniger, 1988). Various biological methods for upgrading plant residues for use as feed have been reported ; however, most of the studies evaluated treated matter *in vitro* and only few studies dealt with *in vivo* treatment.

This paper reports the results of treatments of cereal straws with white rot fungi (*Pleurotus ostreatus*), including lignin degradation, dry matter degradability by cellulase, and digestibility and voluntary intake by sheep.

Materials and methods

The major strains of *Pleurotus ostreatus* used in the series of experiments consisted of Hi80-2, Hi76-3 which were supplied by the Hokkaido Forest Products Research Institute, IPB No. 53 by the Forestry and Forest Products Research Institute and TMI30026 by the

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Rice straw, wheat straw, and soybean straw were purchased from farms in the Sorachi District of Hokkaido. These straws were chopped to fragments about 3 cm long and soaked into water to adjust the moisture content to about 65 %. Then, about 286 g of moist straw (dry matter 100g) was packed either in a glass or a polypropylene bottle for the early experiments. In later experiments for digestion and intake trials, 1 kg of moist straw was packed in 2 l plastic bags. These straw packs were sterilized at 121℃ for 60 min. After cooling to room temperature, these straw substrates were inoculated with 4 g of homogenized fungus mass that had been grown in a shaking culture for 3 weeks. Inoculated straws were incubated at 25℃ for 15, 30, 45, 60, 90 and 120 days, then dried, and mixed. Aliquots were then taken for analysis. For digestion and intake trials, 300 packs of straw were incubated for 100, 135 and 140 days.

Four adult wethers weighing about 60 kg were kept in individual cages and fed *ad lib.* unraveled packs for 14 days. All the feces were collected in the last 7 days.

Carbohydrate content was determined by subtracting the crude protein and ether extract amounts from the amount of organic matter. Difference between the content of organic neutral detergent fider (NDF) and acid detergent fiber (ADF) was considered to correspond to the hemicellulose content, and that between the content of organic ADF and acid detergent lignin (ADL) to the cellulose content. Dry matter degradability by cellulase (Ce-DMD) was determined by the method described by Abe (1988) with slight modifications.

Results and discussion

In the first experiment, rice straw substrates with and without rice bran were compared. Changes in the dry matter losses during the incubation with Hi80-2 were similar, while the cellulose content in the unsupplemented culture was higher. The hemicellulose content in the culture was lower than that in the supplemented culture during the incubation (Fig. 1). Ce-DMD for the unsupplemented culture was higher after 60 days of incubation (Fig. 2). Some other strains of *P. ostreatus* were compared in the experiment, and it was shown that the measurements varied considerably depending on the strains (data not shown). These observations suggest that the selection of strains with higher activities of lignolysis and lower cellulolytic activities is more important than supplementation, for the purpose of such an experiment. Incubation of straw culture for 120 days with Hi80-2 increased the Ce-DMD from 31 to 52%.

In the second experiment, rice straw, wheat straw and soybean straw were used as substrates without any supplementation. Dry matter losses ranged between 20 to 25 % after 90 days of incubation with IPB No. 53. Changes in the cellulose, hemicellulose and ADL contents during the incubation are shown in Fig. 3. The hemicellulose contents in the rice and wheat straws decreased rapidly in the early stage and became stabilized after 60 days of incubation. The ADL contents of the substrates decreased consistently after 15 days of incubation. The Ce-DMD of wheat straw culture increased rapidly from 20 to 52 % within 60 days, while that of rice and soybean straws increased after 45 days of incubation (Fig. 4). The rate of fungal growth appeared to be higher in the wheat culture than in the other cultures.

The chemical composition of the rice straws before and after incubation in the bags with Hi76-3, TMI30026 and IPB No. 53 is shown in Table 1. The hemicellulose content of the incubated straws decreased considerably, while the changes in the cellulose and crude protein contents were negligible. The ADL content increased after incubation except in the case of 135 days of incubation with TMI30026. Fungus growth in the plastic bags was relatively slower than that in the glass bottles, especially in the tightly packed bags. Incubation periods were not long enough to reduce the ADL content for all cases as seen in the Table.

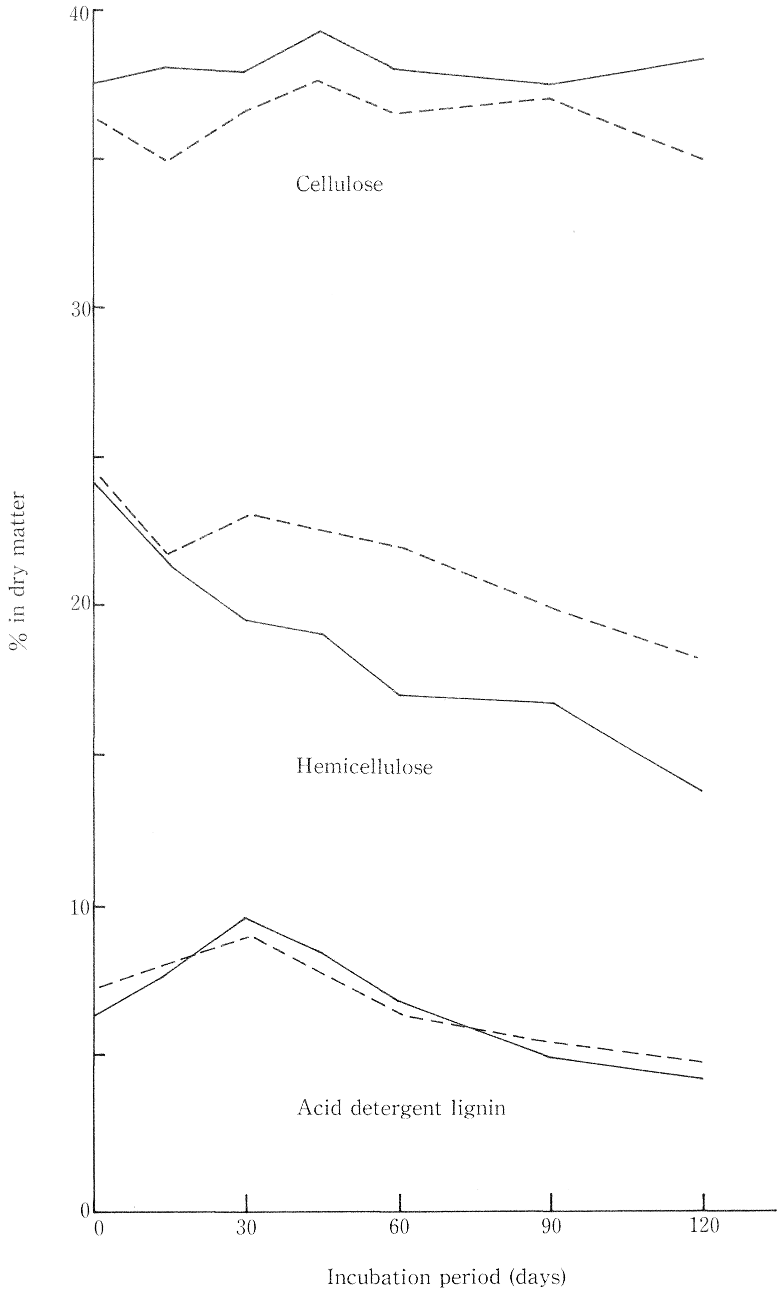

Fig. 1 Changes in cellulose, hemicellulose and ADL content of rice straw substrates with (——) and without (---) rice bran during the incubation with *P. ostreatus* (Hi 80-2).

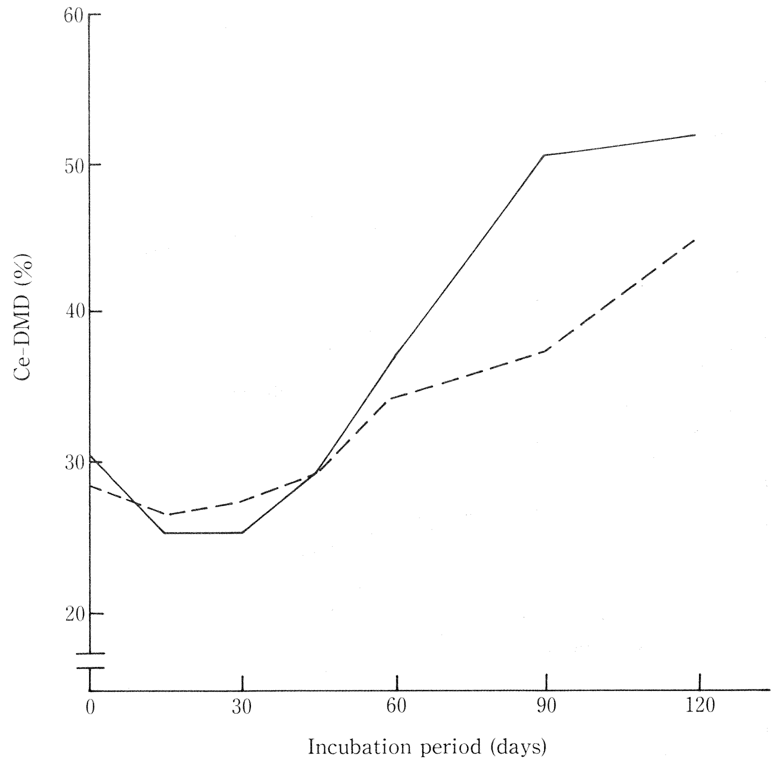

Fig. 2 Changes in dry matter degradability by cellulase (Ce-DMD) of rice straw substrates with (——) and without (---) rice bran during the incubation with *P. oetreatus* (Hi 80-2).

Table 1 Chemical composition of rice straw substrates incubated in plastic bags with *Pleurotus ostreatus* (% DM)

Strains	Hi76-3		TMI30026		IPB No.53	IPB No.53
Incubation (days)	0	135	0	135	100	140
Organic matter	83.9	80.3	85.3	81.2	84.7	83.5
Crude protein	5.3	6.3	5.6	7.9	6.9	7.1
Ether extracts	1.7	1.3	1.0	0.5	0.5	0.4
Carbohydrate	76.9	72.7	78.7	72.7	77.3	76.0
Cellulose	35.6	36.0	35.6	32.3	34.2	35.6
Hemicellulose	25.8	19.4	28.1	15.9	22.7	19.8
Acid detergent lignin	4.7	5.4	5.8	5.1	10.9	7.9

The incubation lowered the *in vivo* apparent digestibilities in wethers, except in the case of carbohydrates after 135 days of incubation with TMI30026 (Table 2). Voluntary dry matter consumption by the wethers increased from about 12-13 g/kg BW to about 20 g/kg BW after 135-140 days of incubation. Intakes of digestible dry matter, organic matter and

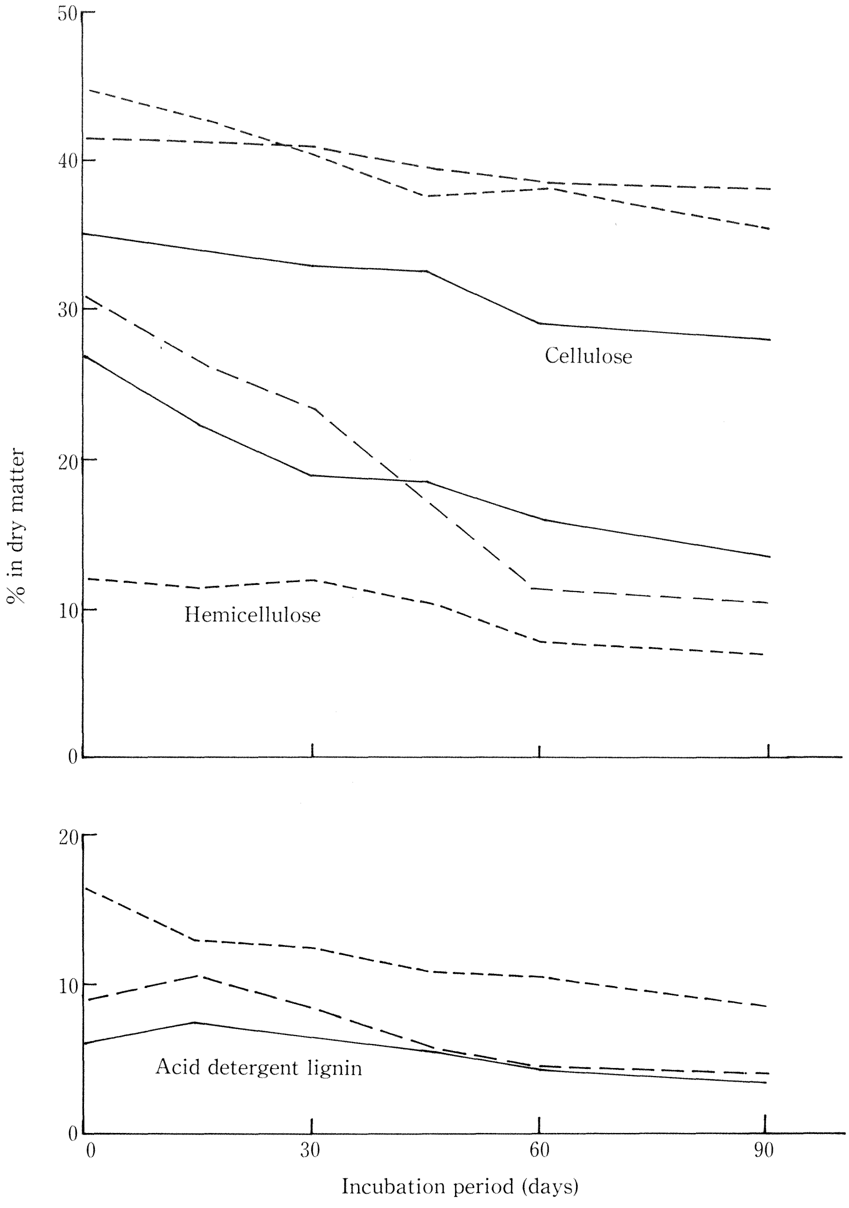

Fig. 3 Changes in cellulose, hemicellulose and ADL content of rice straw (——), wheat straw (– – –) and soybean straw (-----) during the incubation with *P. ostreatus* (IPB No. 53).

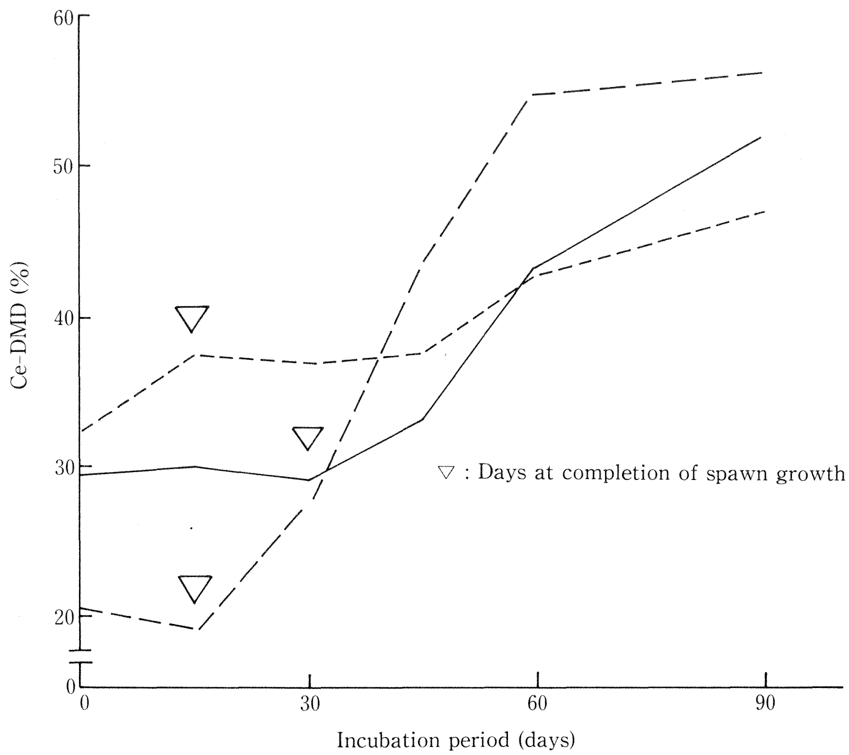

Fig. 4 Changes in dry matter degradability by cellulase (Ce-DMD) of rice straw (——), wheat straw (– – –) and soybean straw (-----) during the incubation with *P. ostreatus* (IPB No. 53).

Table 2 Apparent digestibility of rice straw substrates by sheep before and after incubation with *Pleurotus ostreatus* (%)

Strains	Hi76-3		TMI30026		IPB No.53	IPB No.53
Incubation (days)	0	135	0	135	100	140
Dry matter	52.0	42.7	44.8	43.4	37.0	41.4
Organic matter	57.1	49.9	48.5	48.4	39.9	45.6
Crude protein	30.9	14.2	27.9	8.2	16.0	22.2
Ether extracts	73.1	33.8	57.2	9.4	30.7	44.6
Carbohydrate	58.5	53.3	49.8	53.0	42.0	47.9

Table 3 Intake of rice straw substrate by sheep before and after incubation with *Pleurotus ostreatus* (g/kgBW)

Strains	Hi76-3		TMI30026		IPB No.53	IPB No.53
Incubation (days)	0	135	0	135	100	140
Dry matter	13.3	20.8	12.0	21.0	12.4	19.8
Digestible dry matter	6.9	8.9	5.6	9.1	4.6	8.2
Digestible organic matter	6.3	7.8	5.0	8.2	4.1	7.4
Total digestible nutrients	6.6	8.4	5.1	8.3	4.2	7.7

total digestible nutrients increased after 135-140 days of incubation in conjunction with the increase in dry matter intake (Table 3).

It has been reported that the *in vitro* digestibility increased with the incubation of cereal straws with selected fungi (Zadrazil, 1977 ; Streeter *et al*., 1982 ; Kamra and Zadrazil, 1988), while no positive effects have been reported in *in vivo* trials with straw substrates after harvest of mushroom (Sommer *et al*., 1978 ; Reihild and Schoner, 1984). In the present experiment, the incubation with Hi76-3 for 135 days increased the Ce-DMD from 25.5 to 37.5 %, while the *in vivo* dry matter digestibility decreased from 52.0 to 42.7 %. The reason for the discrepancy between the *in vitro* and *in vivo* results is not clear. Voluntary intake increased remarkably unlike the digestibility. Fungal incubation decreased the hemicellulose content. Okamoto and Miyazaki (1990) pointed out that hemicellulose reduction by ammoniation of rice straw increased the intake due to the development of a more brittle structure, and enhanced the particle size reduction in the rumen. Fungal treatment may exert a similar effect on the brittleness of the straw structure.

Further investigations into such topics as the duration of incubation required for the decrease of ADL content, incubation with more active fungi in lignolysis, and incubation in a larger package should be carried out.

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Discussion

Kume, T. (Japan) : Is there any special reason for selecting the strain of *Pleurotus ostreatus* for the fermentation among the many kinds of white rot fungi? If the mushroom is harvested earlier as edible mushroom, is there any adverse effect on the feed quality?

Answer : We selected *P. ostreatus* because there is a long history of cultivation of this mushroom and the technology is well developed. Moreover since the growth rate is very high, it is easy to incubate and inoculate the mushroom. The nutritive value of the feed decreases slightly after harvest of the edible mushroom. However there is no alternative, mainly for economic reasons, since the edible mushroom fetches a high price, hence enabling to decrease the cost of production.