Ethanol production from switchgrass: Can mushrooms reduce cost?

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One of the challenges in switchgrass ethanol production is breaking down lignin in the plant cell wall. Lignin is one of the plant cell components that provides vegetation support and structure and encapsulates the carbohydrates or sugars. To access the plant sugars for ethanol conversion, lignin has to be broken down through a pretreatment process, which is costly. Oyster mushrooms produce lignin degrading enzymes that can selectively remove lignin from switchgrass (Figure 1). The goal of this project was to determine if oyster mushrooms could be used as a fungal pretreatment during switchgrass bale storage.

Figure 1. Oyster mushrooms after maturation (left) and managed switchgrass production field (right).

The idea is that oyster mushroom spores could be introduced into switchgrass bales prior to bales being placed into storage. During storage, the mushrooms would degrade lignin without consuming the fermentable sugars required for ethanol production, which would reduce ethanol production costs significantly. Small-scale laboratory tests suggest that this concept is feasible, as the oyster mushrooms reduced lignin content after storage for two months. A scaled-up experiment was developed using small (24" x 15" x 12") Kanlow switchgrass bales. Twenty-seven bales received one of three mushroom spore application rates: 1) no mushroom spores - control, 2) low spore loading (~0.75 lbs/bale) or, 3) high spore loading (~1.5 lbs/bale). All bales were placed in storage at the same time. Nine bales, three of each loading rate, were removed from storage after 27 days, another nine bales were removed after 54 days, and the last bales were pulled from storage after 81 days. When the bales were removed from storage, they were sampled for lignin degradation and sugar content evaluation.

Maintaining ideal oyster mushroom spore growth conditions was a challenge. The ideal conditions required maintaining the environmental temperatures between 75 - 85° Fahrenheit and a bale moisture content at 50% or greater. Prior to putting the bales into storage, the bales were weighed, soaked in water overnight, and then inoculated with mushroom spores at four separate locations within each bale (Figure 2). At each inoculation location, a thermocouple was inserted to monitor internal bale temperature and a drip hose was inserted to add water if the internal moisture content dropped below 50%. After inoculation, the bales were reassembled and compressed using ratchet straps.

The reassembled bales were hung from load cells on a custom built storage rack. The load cells were used to monitor bale weight changes which were used to calculate bale moisture content. The load cells and thermocouple information was used to control an automated watering system to maintain a bale moisture content between 50 and 60%.


Figure 2. Bales were soaked in water overnight in swimming pools (left), inoculated with fungal spores between bale flakes in four locations (middle), and then placed in a storage rack where their moisture content was monitored (right).

Lignin, glucan and xylan was degraded in all of the bales, which indicates that there was significant biological activity in both the control and mushroom-treated bales. However, more lignin and xylan was degraded in the mushroom-treated bales than in the control bales. Both the loadings resulted in similar lignin and xylan degradation. Further, there was no difference in glucan degradation among the control and mushroom-treated bales. These results indicate that addition of mushroom spores to the bales had the desired effect of decreasing lignin without reducing glucan any more than the bales without the mushroom addition. This should improve sugar yield from the grasses ,after degradation by cellulase enzymes, for fermentation to ethanol and other products. The results also show the need for a heat treatment step prior to mushroom addition to kill existing microorganisms present in the bales.

Figure 3. Fungal growth (white) being overtaken by bacterial/microbial growth (black).

For more information about this project, contact Dr. Mark Wilkins using the information below.

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