Original scientific paper

# Leftover bread as a substrate for *Pleurotus ostreatus* cultivation

Andrej **GREGORI**[1*], Andrej **KRAŠEVEC**[2], Bojan **PAHOR**[3], Dušan **KLINAR**[1]

**ABSTRACT:**
Many different leftover materials have been used as substrates for mushroom cultivation in the past decades, solving problems with their deposition, their negative impact on the environment, reducing price of the substrates and some of them even raising quality and quantity of harvested fruiting bodies. Leftover bread as being one of those materials has, as far as we are aware, never been tested before as an ingredient of mushroom cultivation substrate. 12.000 to 15.000 tons of leftover bread is dumped each year in Slovenia alone, representing a burden for bakery industry and a negative effect on the environment. In this research leftover bread showed to be an appropriate supplement for wood based substrate used for *P. ostreatus* fruiting bodies cultivation, resulting in 63 % biological efficiency. In comparison to wheat bran and crushed corn seed containing substrates *P. ostreatus* fruiting bodies yields were higher on substrates composed of leftover bread, sawdust and $CaCO_3$. Using leftover bread for mushroom cultivation could be an appropriate solution for its problematic deposition, resulting in solving financial problems of its producers, negative impact on the environment and in substrate price reduction.

**KEY WORDS:**
Leftover bread, *Pleurotus ostreatus*, Mushroom cultivation.

Received: 5. 5. 2008.
Accepted: 16. 5. 2008.

[1] Andrej Gregori
SRC Bistra Ptuj,
Slovenski trg 6, SI - 2250 Ptuj,
Slovenia
E-mail: andrejg@bistra.si

[*] corresponding author

[2] Andrej Kraševec
Institute of Natural Sciences,
Ulica bratov Učakar 108, SI - 1000 Ljubljana, Slovenia

[3] Bojan Pahor
Perutnina Ptuj d.d.,
Potrčeva cesta 10, SI - 2250 Ptuj,
Slovenia

[1] Dušan Klinar
SRC Bistra Ptuj,
Slovenski trg 6, SI - 2250 Ptuj,
Slovenia

## INTRODUCTION

*Pleurotus* species accounting for 14.2 % of the total edible mushrooms world production in 1997 [1] are very popular because of simple cultivation, high nutritional value and high yield potential [2]. They can be cultivated on wide spectrum of lignin and cellulose containing organic matter and therefore also play an important role in managing further use of these organic matter [3]. Substrates that can be used for *Pleurotus* sp. cultivation are: cotton stalks and sorghum stover [4], banana straw [5], spent brewery grains [6], cottonseed hulls [7], cardboard [8], rice straw [9], different grass types [10], weed plants [11] and many others. In Slovenia in average 12 % of bread is returned from the supermarkets back to the producers. This kind of bread is considered opposable, therefore can be used for animal feed or bread crumbs only under strict conditions. Due to that 12.000 to 15.000 tons of leftover bread has to be dumped each year in Slovenia only, representing a financial burden to the industry and also considerable negative impact on the environment [12].

In our research appropriateness of leftover bread as a component of substrate for *Pleurotus* ostreatus fruiting bodies production was tested.

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## MATERIALS AND METHODS

### *Fungal inoculum* preparation

*P. ostreatus* strain Pl.o4 was obtained from fungal culture collection at Biolab Corp., Podkoren 72, 4280 Kranjska gora, Slovenia. Culture was transferred to Potato Dextrose Agar (PDA) and maintained at 24 °C in a dark place. PDA overgrown with mycelium was cut with a sharpened steel pipe to acquire 9 mm diameter discs overgrown with mycelia, which were used for substrate mixtures inoculation.

### Substrate preparation

Leftover bread (LB) was dried and crushed (Universalmixer UM 2927, CTC Clatronic, Germany) and mixed with other substrate components (Table 1). 720 mL glass jars were filled with 300 grams of substrate mixture, covered with punctured lids (9 mm hole closed with cotton stopper), and sterilized at 121 °C for three hours. Five replicates were conducted for each substrate mixture.

### Substrate inoculation

After cooling, a 9 mm diameter disc was used for inoculation of each filled glass jar and centered to the leveled substrate surface. Inoculated substrate mixtures were incubated in a dark place at 24 ±1°C.

### Mycelium growth measurements

Mycelium growth on substrate mixtures filled into glass tubes was measured and the average growth calculated from the fastest and slowest mycelium growth front point and averaged for all five replicates.

**Table 1.**
Substrate mixtures composition.

| | wheat bran (%) | crushed corn seeds (%) | leftover bread (%) | $CaCO_3$ (%) | sawdust (%) |
|---|---|---|---|---|---|
| WB1 | 10 | | | 2 | 88 |
| WB2 | 20 | | | 2 | 78 |
| WB3 | 30 | | | 2 | 68 |
| CC1 | | 10 | | 2 | 88 |
| CC2 | | 20 | | 2 | 78 |
| CC3 | | 30 | | 2 | 68 |
| LB1 | | | 10 | 2 | 88 |
| LB2 | | | 20 | 2 | 78 |
| LB3 | | | 30 | 2 | 68 |
| LB4 | | | 40 | 2 | 58 |
| LB5 | | | 50 | 2 | 48 |

### Mushroom cultivation

When the mycelium has overgrown the substrate mixture and reached the bottom of glass jars, the jars were transferred to the mushroom cultivation room having 16 ± 1 °C, 90 ± 5 % relative humidity, 10 hours light cycle and approximately 1,300 mg/kg $CO_2$. Sporocarps were harvested before caps started to invert. Acquired fruiting bodies were cleaned of excess substrate and their weight determined. Biological efficiency (BE), defined as fresh fruiting bodies weight divided by initial dry substrate weight multiplied by 100, for each substrate was calculated. The average BE for each substrate mixture was determined. Experiments were performed at the mycological laboratory of Biolab Corp. and Institute of Natural Sciences, Podkoren, Slovenia.

## RESULTS AND DISCUSSION

Highest mycelium growth rate after 22 days of incubation was noticed on the substrates with 50 % of LB as an additive, while 10 % crushed corn seed (CC) addition to the substrate resulted in the lowest mycelia growth rate. With increasing proportions of additive to the substrate, mycelium growth rate was increasing (Fig. 1).

Highest BE of harvested *P. ostreatus* fruiting bodies (63 %) was achieved on substrates containing 50 % LB as a supplement. BE on substrates containing wheat bran (WB) was the lowest at all three rates tested for this additive (Fig. 1).

LB showed to be an appropriate substitute for WB or CC when used as a wood based substrate component for *P. ostreatus* cultivation. LB showed to be more appropriate than other two additives tested. Higher proportions of LB up to 50 % induce mycelium growth rate and harvested fruiting bodies BE. Research work will be further focused on other edible and medicinal mushroom species (*Grifola sp., Ganoderma*

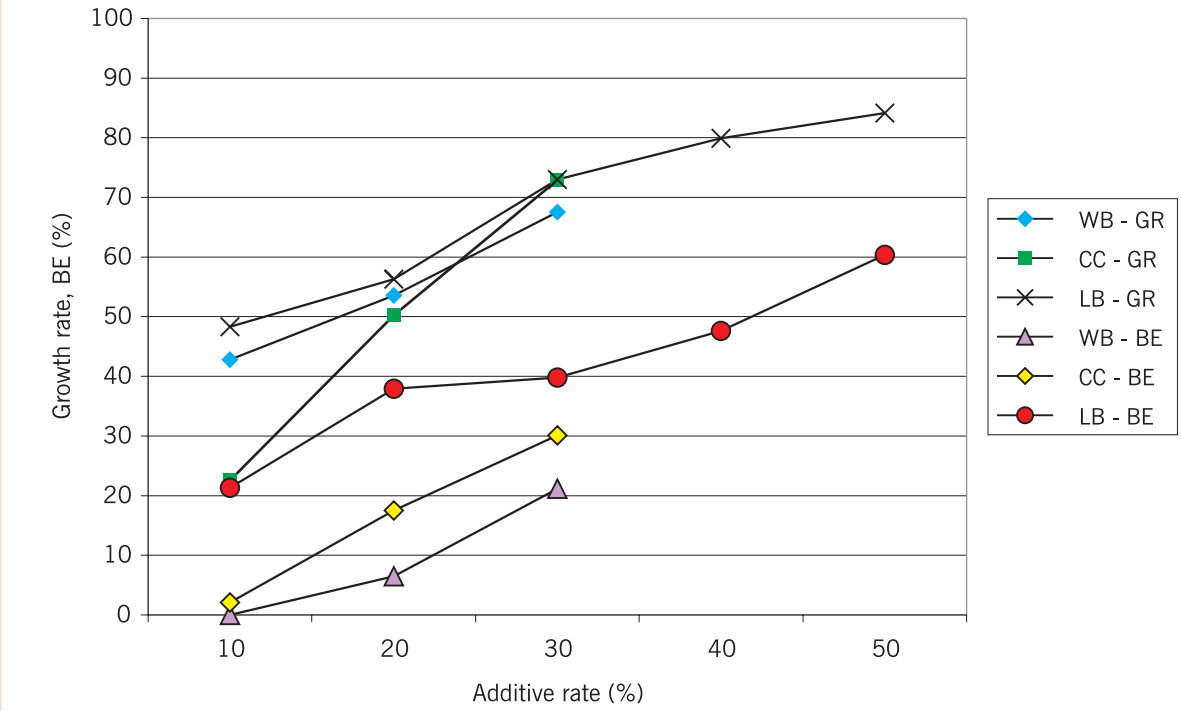


**Figure 1.**
Mycelium growth rate (GR) after 22 days of incubation and fruiting bodies BE on substrates composed of beech sawdust and different additive rates.

*sp., Flammulina sp., Lentinula sp.* and others) and will determine LB substrate appropriateness for cultivation of these species.

If LB substrate will be proved as suitable forcultivation of these species, technology could be used also in large scale, solving problems with LB deposition and mushroom cultivation for human food.

According to the results of our research LB could be successfully used as a substrate additive for *P. ostreatus* cultivation. Using LB for mushroom cultivation substrate could be an appropriate solution for problematic deposition of LB, resulting in solving financial problems of LB producers, negative impact on the environment and substrate price reduction.

Results of research could be important especially for Slovenia because problems with LB are more distinctive in Slovenia than other countries [12].

## Acknowledgements

This research work was financed by the Slovenian Research Agency (applicative research project No. L2-7598), Perutnina Ptuj d.d., the Municipality of Ptuj and Institute of Natural Sciences. We would like to thank all of them for their support.

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