

Screening strains of genus *Pleurotus* for biomass production in solid state fermentation of agricultural residues

Diamantopoulou P.¹, Melanouri E.-M.^{1,*}, Papanikolaou S.²

¹Hellenic Agricultural Organization ELGO-DIMITER, Institute of Technology of Agricultural Products (ITAP), Laboratory of Edible Fungi, Sof. Venizelou 1, 14123, Lykovrysi Athens, Greece,

²Agricultural University of Athens, Department of Food Science & Technology, Laboratory of Food Microbiology & Biotechnology, Iera Odos 75, 11855 Athens, Greece

*Corresponding author: Diamantopoulou Panagiota, e-mail: sdiamantopoulou@gmail.com

Abstract

The ability of several strains belonging to *Pleurotus ostreatus* and *P. eryngii* mushroom genera to grow on various agricultural residues was examined and their mycelium growth rates and biomass production (estimated as glucosamine content) were compared. Four *P. ostreatus* (AMRL 137, 144, 147, 150) and four *P. eryngii* (AMRL 160, 163, 166, 173-6) strains were cultivated on wheat, barley and oat straw, poplar and beech-wood sawdust, cotton and coffee residues, corn-cobs, rice bark, olive cake supplemented with wheat bran on a final C/N 20-30. Colonization rate measurements of mycelium demonstrated faster colonization on wheat, beech, barley and oat, corn-cobs and rice with values of ~1.5 mm/day, yet the faster colonizers were *P. ostreatus* 144, 150 and *P. eryngii* 166, 173-6. Glucosamine content was similar for *P. ostreatus* and *P. eryngii* strains and the most pronouncing substrates for more biomass production were barley and oat straw, beech-wood sawdust, cotton and coffee residues, corn-cobs (max 450 mg/g d.w.). However, in most of the cases, glucosamine content was opposite to mycelial growth rate, as strains with high colonization rates produced the least biomass. These results are evaluated in the view of bio-converting agricultural wastes into mushrooms, an added value food with medicinal properties.

Keywords: mushroom cultivation, agro-residues fermentation, *Pleurotus*, biomass

1. Introduction

Mushroom fruiting bodies have been used as food and food-flavouring materials for centuries for their unique and subtle flavour. *Pleurotus* spp. (Oyster mushrooms) comprises the group of edible white-rot fungi with important medicinal properties and biotechnological and environmental applications (Chang and Miles 2004). *Pleurotus ostreatus* is an edible mushroom that possesses antioxidant potentials. This white-rot fungus can effectively biodegrade an extraordinary variety of lignocellulosic materials. It is easy to cultivate and can fruit on various agro-industrial wastes (Philippoussis 2009). Agro-industrial/lignocellulosic wastes on the other hand are formed in large amount every year. For this reason, finding alternatives for the reutilize of these wastes is an aim that has been taken into account by countries around the

world, considering not only environmental but also economical aspects. A quick and cheap method to determine the suitability of wastes as substrate for mushroom cultivation constitutes the 'glass-tube' method (Philippoussis et al. 2007). Therefore, experiments using several agro-wastes and *Pleurotus* strains were conducted in glass tubes and mycelial growth rate and biomass concentration (glucosamine content) were evaluated (Economou et al. 2017).

2. Materials and Methods

Strains of *P. ostreatus* (AMRL 137, 144, 147, 150) and *P. eryngii* (AMRL 160, 163, 166, 173-6) of the Edible Fungi Laboratory/ ITAP culture collection were used. All the substrates were prepared at a ratio of 80% residue to 20% supplements (wheat bran and CaCO₃). Residues (wheat, barley and oat straw, poplar and beech-wood sawdust, cotton and coffee residues, corn-cobs, rice bark, olive cake) were soaked in water for 2-4h and after drainage, supplements were added and mixed. The moisture content of the sterilized substrate was 68-72%. The glass – tubes were inoculated with the fungi and incubated at 26 ± 1 °C in the dark until the fungal colony occupied the whole tube. The growth rate (Kr, mm/d) of the fungal colony was estimated by measuring its visible penetration into the substrate daily in a set of four test tubes according to Philippoussis et al. (2003). The glucosamine content of the fungal cell wall was used to monitor mycelial biomass and it was determined according to (Economou et al. 2017).

3. Results – Discussion

The days needed for total mycelium growth of *Pleurotus* sp. from inoculation exhibited significant differences between substrates. Data revealed that all strains had a fast colonization in wheat straw (max Kr=1.66 mm/d), a result that was expected as wheat straw is the basic substrate for mushrooms cultivation. Beech and poplar residue, barley and oat, corn combs and rice (especially for *P. ostreatus* strains) were the substrates with the highest mycelium growth rate after wheat straw (Kr~1.5 mm/day). In general, *P. ostreatus* demonstrated faster growth rate and biomass production over *P. eryngii* strains. *P. ostreatus* 144, 140 and *P. eryngii* 166, 173-6 strains were the fastest colonizers and in most cases the

producers of the max amount of glucosamine (mean values 250-290 mg/g d.w.). Exceptionally, *P. ostreatus* 137 produced 450 mg/g d.w. on poplar sawdust. However, not always a correlation between biomass production and the various substrates was detected. For wheat straw and barley and oat there was a linear relationship between the glucosamine amount and the mycelium growth rate, whereas, cotton residue with one of the lowest colonization rates (mean Kr=1.01 mm/d) showed one of the largest biomass productions (mean

Gluc=280.25 mg/g d.w.). The obtained results are in accordance with our previous findings reporting that growth rate and biomass production were negatively related (Philippoussis et al. 2011, Philippoussis and Diamantopoulou 2012). However, the successful growth of all *Pleurotus* strains on all agrowastes tested supports the potential effective bio-conversion of several lignocellulosic wastes into mushroom carposomes, an added value food with medicinal properties.

Table 1. The glucosamine content (U/g d.w.) and mycelium growth rate (Kr, mm/d) of *Pleurotus ostreatus* and *Pleurotus eryngii* strains grown on glass-tubes with wheat straw (W) and cotton residue (C) at 26± 1 °C

Mushroom strains	Substrate	Glucosamine (U/g d.w.)	Kr (mm/d)
<i>P. ostreatus</i> 137	W	335.02	1.66
	C	202.39	1.19
<i>P. ostreatus</i> 144	W	350.58	1.50
	C	247.70	1.13
<i>P. ostreatus</i> 147	W	340.32	1.58
	C	240.32	1.16
<i>P. ostreatus</i> 150	W	310.26	1.38
	C	236.74	1.15
<i>P. eryngii</i> 160	W	230.51	1.16
	C	236.74	1.00
<i>P. eryngii</i> 163	W	192.98	1.19
	C	198.77	0.85
<i>P. eryngii</i> 166	W	247.25	1.37
	C	422.59	0.85
<i>P. eryngii</i> 173-6	W	176.60	1.31
	C	220.01	0.87

Acknowledgements

This research has been co-financed by the European Regional Development Fund of the European Union and Greek national funds through the Operational Program Competitiveness, Entrepreneurship and Innovation, under the call RESEARCH – CREATE – INNOVATE (project code: T1EDK-05027), scientifically coordinated by the Hellenic Agricultural Organization – DEMETER (Institute of Technology of Agricultural Products/ Laboratory of Edible Fungi).

References

- Chang S.T. and Miles P.G. (2004) Mushrooms: Cultivation, nutritional value, medicinal effect and environmental impact (2nd edition), S.T. Chang and P.G. Miles, (Eds.), CRC Press LLC, Boca Raton, Florida.
- Economou C.N., Diamantopoulou P.A. and Philippoussis A.N. (2017). Valorization of spent oyster mushroom substrate and laccase recovery through successive solid-state cultivation of *Pleurotus*, *Ganoderma* and *Lentinula* strains. *Applied Microbiology and biotechnology* **101** (12), 5213-5222.
- Philippoussis A. (2009) Production of mushrooms using agro-industrial residues as substrates. In: Sing Nigam P, Pandey A (eds) *Biotechnology for agro-industrial residues processing*. Springer, Berlin, pp 163–196.
- Philippoussis A. and Diamantopoulou P. (2012) Exploitation of the biotechnological potential of agro-industrial by-products through mushroom cultivation. In M. Petre & M. Berovic (eds), *Mushroom Biotechnology and Bioengineering*, University of Pitesti, Bucuresti: CD Publishing House, ISBN 978-606-528-146-2, pp. 161–184

- Philippoussis A., Diamantopoulou P. and Zervakis G. (2003) Correlation of the properties of several lignocellulosic substrates to the crop performance of the shiitake mushroom *Lentinula edodes*. *World Journal of Microbiology and Biotechnology* **19**,551–557.
- Philippoussis A., Diamantopoulou P. and Israilides C. (2007). Productivity of agricultural residues used for the cultivation of the medicinal fungus *Lentinula edodes*. *International Biodeterioratio and Biodegradation* **59**, 216–219.
- Philippoussis A, Diamantopoulou P, Papadopoulou K, Laktar H, Roussos S, Parissopoulos G. and Papanikolaou S. (2011). Biomass, laccase and endoglucanase production by *Lentinula edodes* during solid state fermentation of reed grass, bean stalks and wheat straw residues. *World Journal of Microbiology and Biotechnology* **27**, 285–297.



Ευρωπαϊκή Ένωση
Ευρωπαϊκό Κοινωνικό Ταμείο



ΕΛΛΗΝΙΚΗ ΔΗΜΟΚΡΑΤΙΑ
ΥΠΟΥΡΓΕΙΟ
ΟΙΚΟΝΟΜΙΑΣ & ΑΝΑΠΤΥΞΗΣ
ΕΙΔΙΚΗ ΓΡΑΜΜΑΤΕΙΑ ΕΠΙΧΕΙΡΗΣΙΑΚΩΝ
ΕΠΙΧΕΙΡΗΣΙΑΚΩΝ ΠΡΟΓΡΑΜΜΑΤΩΝ



Με τη συγχρηματοδότηση της Ελλάδας και της Ευρωπαϊκής Ένωσης