Full Length Research Paper

# The effect of the interaction of various oil types with different culture media on biomass production of *Psathyrella atroumbonata* Pegler

Nwanze PI<sup>1\*</sup>, Ameh JB<sup>2</sup> and Umoh VJ<sup>3</sup>

<sup>1</sup>Department of Biological Sciences, College of Natural and Applied Sciences, Igbinedion University, Okada, P.M.B. 0006, Edo State, Nigeria.

<sup>2</sup>Department of Microbiology, Ahmadu Bello University, Zaria. Kaduna State, Nigeria.

<sup>3</sup>Department of Microbiology, Ahmadu Bello University, Zaria. Kaduna State, Nigeria.

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*Psathyrella atroumbonata*, an indigenous mushroom species, was cultured on six different media that were inoculated separately with three different grain spawns and amended with six different oils at five different rates. The results revealed that the interaction of the various oils with the different culture media produced a highly significant effect (p<0.01) on the dry weights and stipe and pileus diameters of *P. atroumbonata*. The widest mean stipe diameters were produced by the interaction of coconut x animal bedding and rice as well as butterfat x sawdust media. Both the heaviest mean dry weight and widest pileus diameters were induced by coconut x animal bedding and rice medium.

Key words: Psathyrella atroumbonata, supplemented medium, oil type, biomass production.

# INTRODUCTION

Mushrooms have innumerable practical uses apart from their edibility and commercial value (Sunagawa and Magae, 2005; García-Pascual et al., 2005). They may be used as biocontrol agents for nematodes, insects and other fungi and they have a varied range of applications in bioremediation of soil, bioconversion of wastewater and medicine (Sudirman et al., 1994; Luo et al., 2004; Ragunathan and Swaminathan, 2004; Cliquet and Jackson, 2005; D'Annibale et al., 2005). In addition, they are nutritionally commendable and the enzymes extracted from them have great potential industrial applications (Cohen et al., 2004; Chang and Miles, 2004; Kimura et al., 2005; Mata et al., 2005). More important, however, is agricultural waste conversion, which produces fungal biomass, contributes to products of commercial interest and minimizes pollution (Nwanze and Adamu, 2004; Nwanze et al., 2005a).

Environmental factors such as temperature, light, humidity and pH, as well as, mechanical injury and chemical treatment have been reported to affect carpophore production (Hibbett et al., 1994; Cordeiro et al., 2004; Chang and Miles, 2004). In addition, media components also play an integral part in mushroom culture (Kim et al., 2005). Media components such as rice bran, brown rice, cassava peels, glucose and some of its derivatives induce fruiting or carpogenesis (Lim et al., 2004; Hanai et al., 2005; Magae et al., 2005).

Genetic crossing may be used to increase carpophore size. However, spawn grains such as wheat, millet and corn and various lipid sources such as groundnut, coconut, palm kernel, palm and cotton oils may also be used to achieve similar results (Nwanze et al., 2004a).

Previous work by Nwanze et al. (2004a, 2004b, 2005b, 2005c) examined the effect of spawn grains, culture medium, oil type and rate as well as some of their

<sup>\*</sup>Corresponding author. E-mail: stonenwanze@yahoo.com. Tel: 2348053067394.

Medium	Components	Method of preparation
Sawdust	62.5 g sawdust 62.5 g wood chips 125.0 g brown rice	All the components were thoroughly mixed, moistened and sterilized for 15 min at 121°C
Animal bedding and rice	125.0 g wood chips 125.0 g brown rice	Same as above
Lime 1	195.0 g sawdust 50.0 g rice bran 2.5 g CaSO₄ 2.5 g CaCO₃	Same as above
Lime 2	235.0 g sawdust 10.0 g rice bran 2.5 g corn meal 2.5 g CaCO <sub>3</sub>	Same as above
Lime 3	182.5 g sawdust 62.5 g corn cobs 5.0 g CaCO₃	Same as above
Formulated	175.0 g sawdust 70.0 g rice bran 2.5 g CaCO₃ 2.5 g oatmeal	Same as above

**Table 1.** Different carpophore production medium.

interactions on carpophore weights and dimensions. The present work, however, seeks to examine the effect of the interaction of oil type and growth medium on the culture of *P. atroumbonata*. It is of paramount importance because there is a need to examine the interactions of parameters rather than optimizing individual parameters (Deshpande et al., 2004).

### MATERIALS AND METHODS

# The effect of various grains, culture media, oil types and rates on carpophore production of *P. atroumbonata*

Various non-composted media including sawdust (Carey, 1974), animal bedding and rice (Roxon and Jong, 1974), formulated (Nwanze, 1996) and lime were used for these studies. To distinguish among three lime media, they were arbitrarily named as lime 1 (Cangy, 1994), lime 2 (Oei, 1991) and lime 3 (Oei, 1991) (Table 1). These six different media were supplemented with different rates (0.007, 0.014, 0.021 and 0.028 ml/g) of different lipid sources viz. groundnut, coconut, palm kernel, butterfat, palm and cotton oils respectively, in order to study the effect of lipids on carpophore production. Two hundred and fifty gram of dry substrate from each of the above six different supplemented and nonsupplemented media were placed in separate polypropylene heat resistant bags (Kadiri, 1999a,b). After thoroughly wetting the substrates, the bags were autoclaved for 15 min at 121°C and allowed to cool (Bhandari et al., 1991). The substrates were then separately inoculated with 10 g (4% on dry weight basis) of three different types of spawn separately (wheat, corn and millet) (Bahukandi and Munjai, 1990). All the bags were incubated in total darkness at 30 ± 2°C for three weeks after which the bags were aerated and exposed to light (Kadiri, 1999a; Caten and Newton, 2000) (Table 1).

# Experimental design

The experiment was conducted in a split-split plot design replicated thrice, with medium as the main plot, oil type and rate as the subplot and grain as the sub-subplot treatment (Jefferson et al., 2001; Garling and Boehm, 2001). The fruiting bodies from different flushes (1-3) in the different experiments were collected and the pileus and stipe diameters as well as the stipe lengths measured (Largent, 1986; Bhandari et al., 1991). In addition, fresh and dry weights were also taken (Šrobárová and Pavlová, 2001; Malone, 2002).

### Statistics

In order to test the main and interactive effects of spawn grain, medium, oil type and rate of amendment, pileus and stipe diameter, stipe length and wet and dry weights of fruiting bodies were recorded and the data subjected to factorial analysis of variance (Kluth et al., 2001). When significant differences were determined for the main effects or their interactions ( $p \le 0.05$ ), comparisons among means were made using Duncan's multiple range test (Snedecor and Cochran, 1987). The values 0.01, 0.1 and 1.0 were added to dry weights, stipe and pileus diameters; and wet weight and stipe length values, respectively prior to analysis (Cowger et al., 2000). The present results were analyzed by analysis of variance as a 6X6 factorial, with 3 replicates, using Genstat.

Table 2. Spawn preparation using different grain and conditions.	
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Spawn	Components	Method of preparation		
Wheat	1.0 kg wheat grains 12.0 g CaSO $_4$ 2H $_2$ O 3.0 g CaCO $_3$ 1.5 litre distilled water	1.0 kg of wheat grains was boiled in 1.5 litre of water for 15 min and left to cool for an additional 15 min. The water was poured off and 900.0 g of the cooked grains was mixed with 12.0 g gypsum and 3.0 g $CaCO_3$ . The grains were then filled into bottles and sterilized for 20 min at 121°C. After cooling, the bottles were inoculated with pieces of agar medium colonized with mycelium and incubated for 2 weeks in total darkness.		
Corn	Same as above except for use of corn as grain	Same as above.		
Millet	Same as above except for the use of millet as grain	Same as above except that the grains were boiled for 5 min.		

Table 3. Stipe diameter, dry weight and pileus diameter of P. atroumbonata as affected by the interaction of oil type and media.

		Media								
Oil	Sawdust	Animal bedding and rice	Lime 1	Lime 2	Lime 3	Formulated				
Stipe diameter (cm)										
Coconut	0.34cde	0.38ab	0.33def	0.29hij	0.30ghi	0.32efg				
Cotton	0.35cd	0.24m	0.25lm	0.30ghi	0.31fgh	0.27jkl				
Groundnut	0.29hij	0.26klm	0.30ghi	0.27jkl	0.34cde	0.30ghi				
Butterfat	0.39a	0.28ijk	0.36bc	0.26klm	0.31fgh	0.34cde				
Palm kernel	0.26klm	0.27jkl	0.28ijk	0.28ijk	0.26klm	0.19n				
Palm	0.28ijk	0.25lm	0.35cd	0.33def	0.29hij	0.32efg				
SE± 0.007										
Dry weight (g)										
Coconut	0.09cde	0.18a	0.11bc	0.09cde	0.09cde	0.10bcd				
Cotton	0.12b	0.07efg	0.05gh	0.08def	0.10bcd	0.07efg				
Groundnut	0.07efg	0.07efg	0.07efg	0.04h	0.09cde	0.07efg				
Butterfat	0.10bcd	0.06fgh	0.07efg	0.11bc	0.07efg	0.11bc				
Palm kernel	0.06fgh	0.05gh	0.06fgh	0.11bc	0.08def	0.06fgh				
Palm	0.06fgh	0.06fgh	0.08def	0.07efg	0.07efg	0.08def				
		SE± 0.0	)07							
		Pileus diame	eter (cm)							
Coconut	2.30bcd	2.74a	2.20c-g	1.59jkl	2.25b-f	2.26b-e				
Cotton	2.07d-h	1.45k-o	1.26no	2.01fghi	2.24c-f	1.66jk				
Groundnut	1.64jkl	1.30mno	1.80ij	1.54klm	2.33bc	2.03efghi				
Butterfat	2.49b	1.59jkl	2.01fghi	1.33mno	2.15c-h	2.32bc				
Palm kernel	1.49k-o	1.40I-o	1.50k-n	1.98ghi	1.91hi	0.98p				
Palm	1.63jkl	1.250	1.79ij	2.29bcd	1.97ghi	1.97ghi				
SE± 0.076										

Means followed by the same letter(s) within the same row or column in a treatment group are not significantly different statistically at 5% level of probability using Duncan's multiple range test.

#### Spawn preparation

Three different types of grain, including corn, wheat and millet were used to produce spawn in order to determine which spawn produces the most excellent crop yield. The spawns were prepared as described by Fritsche (1978) (Table 2) and kept inside a water bath at  $37^{\circ}$ C and  $70^{\circ}$  relative humidity for two weeks in order for the spawn to run (Belewu and Adeniyi, 2001; Gordon et al., 2002) (Table 2).

## **RESULTS AND DISCUSSION**

Mean stipe diameter, dry weight and pileus diameter of *P. atroumbonata* as affected by the interaction of oil type and media is shown in Table 3. The interactions of butterfat and sawdust medium as well as coconut oil with animal bedding and rice medium produced the widest stipe diameters, while the least was produced by the interaction between palm kernel and formulated. The interaction of coconut oil with animal bedding and rice mean dry weight of *P. atroumbonata* that was significantly heavier than those produced by the interaction of coconut oil with animal bedding and rice medium gave rise to a mean dry weight of *P. atroumbonata* that was significantly heavier than those produced by the interaction of the other oils and media. The interaction of coconut oil with animal bedding and rice medium also produced the widest pileus diameter.

The above results can be explained by the sterol content of the oils as well as the media composition of sawdust and animal bedding and rice media. The animal bedding and rice media includes brown rice (Schisler and Volkoff, 1977; Nwanze et al., 2005b). In addition to increasing the surface area for fungal colonization, the high protein, carbohydrate, fatty acid and amino acid content of the brown rice also stimulated fruit production (Tinoco et al., 2001; Lim et al., 2004). The results obtained with lime 1 and 2 and formulated media also support the finding that rice bran increases biomass production (Hanai et al., 2005). In general, however, the results exemplify the importance of media components in mushroom cultivation (Cordeiro et al., 2004; Estévez et al., 2005). In conclusion, the interaction of different media with the various oils had a significant effect on the weight and dimensions of P. atroumbonata. Coconut oil seems to have a profound effect on this particular species. The species should therefore be considered for economic exploitation.

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