

## Optimization of Oyster Mushroom (*Pleurotus Ostreatus*) Mycelium Production Using Amaranth-Based Media for Cell-Cultured Meat Hybrids

Kayise Maseko<sup>1</sup>, Thierry Regnier<sup>1</sup>, Belinda Meiring<sup>1</sup>, Paul Bartels<sup>2</sup>

<sup>1</sup>Tshwane University of Technology, Pretoria, South Africa. <sup>2</sup>Mogale Meat Company, Pretoria, South Africa

### Abstract

Over the last few decades, extensive research has evaluated various aspects of the cultivation and metabolism of different mushroom species, whereby the most prevalent method is through the cultivation in enriched media. Filamentous fungi are the source of mycoprotein, initially identified in the 1960s, cultivated as mycelium for vegan and cell-cultured meat products. The objective of this study was to compare different formulations of amaranth-based media for the cultivation of *P. ostreatus* mycelium with increased protein content. Formulations containing amaranth seed flour, glucose, calcium carbonate (CaCO<sub>3</sub>), leucine, urea, yeast, and malt extract were used for the culture media. *P. ostreatus* (strain HK35) mycelial plug was inoculated aseptically into Erlenmeyer flasks (250 mL) containing different formulations of sterilized liquid media (50 mL). The flasks were incubated at 25 ± 1 °C for up to 17 days after which the mycelium was harvested, and the yield recorded. Mycelium was frozen at -80 °C and lyophilized at -40 °C (Telstar LyoQuest) for 24 h. The nitrogen/protein content (dry basis) was determined using the TruMac Leco, Protein Analyser, operating on the Dumas combustion method according to AOAC 992.15. The results demonstrated that among tested media, CaCO<sub>3</sub>, malt and yeast extract best promoted mycelial growth (8 days), whereas the highest yield (4.26 g) and protein content (34%) of the mycelium were observed on the media optimized with leucine and urea. The properties of the medium can be attributed to the concentration and type of carbon and nitrogen sources. In conclusion, this study confirmed that the protein content of *P. ostreatus* can be increased by using specific media, thereby providing a sustainable source of protein and novel ingredients that could contribute to the world's food supply. In addition, the cultivation of mycelium in controlled conditions could lead to the production of high-value (proteins) and safe biomass product. Nonetheless, further research should be conducted by the Food and Agriculture industry to increase the availability of cultivated mycelium products as vegan protein sources or meat analogues.

### PRESENTER BIOGRAPHY: KAYISE MASEKO

Kayise Maseko is a 2nd year PhD student from the Department of Biotechnology and Food Technology, at Tshwane University of Technology, Arcadia campus. Her thesis is titled "Evaluation of biomaterials for mycelium production and the development of hybrid cell-cultured meat", and is under the supervision of Dr B. Meiring, Professor T. Regnier, and Dr P. Bartels.