# Assessing Nitrogen's Relationship with the Formation of Toxocysts in Oyster Mushrooms

Brian Kearns, Karel Pluhar, & Beth Wilkes

# Oyster Mushrooms & Toxocysts

Certain species of fungi grow on substrates with little available nitrogen. These fungi have adapted methods to acquire the nutrients they need. The hyphae of the fungi are the filamentous cells that grow in a vast network (called mycelium) throughout their environment, similar to the roots of a tree. Oyster mushrooms (*Pleurotus ostreatus*) have evolved the capability of killing nematodes using toxocysts [fig. 1&2] to acquire nitrogen from their decomposition.





### Nematodes



Some nematode species are beneficial to humans. They kill grubs and other harmful invertebrates. However, other nematode species are pests that destroy plant roots, and some can even infect humans.

A toxocyst is a chemical-filled sphere which develops on the hyphae of certain fungi such as *P. ostreatus*. They are fragile and will break at a slight touch, such as a nematode coming into contact. This releases the volatile chemicals within, which causes necrosis in nematode cells.

# Experiment



Malt extract agar with nitrogen fertilizer solution in concentrations of 1%, 2%, and 4% was used as the substrate for the mycelium. By growing *P. ostreatus* mycelium in agar containing varying concentrations of nitrogen, the effect of nitrogen on toxocyst production quantity was evaluated.

Contamination was a recurring issue [fig. 4&5]. Stricter sterilization methods were put in place and new agar plates were inoculated with a *P. osteratus* spore solution [fig. 7]

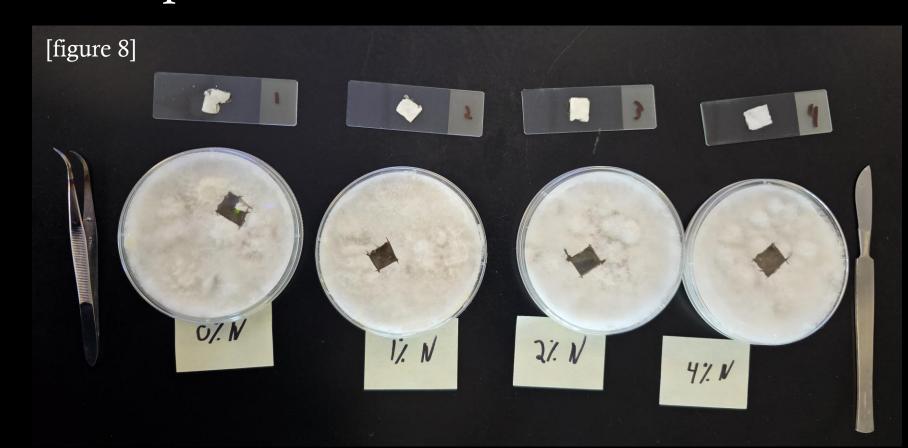




## Agar Plates and Mycelium



After approximately 3 weeks, the mycelium had propagated enough to view a sample under a microscope [fig. 7]. This allowed enough time for full toxocyst development.



Sections of the mycelium were cut out and carefully scooped onto slides for viewing under a microscope [fig. 8]. Care had to be taken not to break the toxocysts during cutting and transfer. All microscopic images were taken at 400x magnification.

# Results & Conclusion

Concord's Community College

Observations showed a noticeable trend in the development of toxocysts on P. ostreatus at varying nitrogen concentrations. In the mycelium from the 0% nitrogen plate, toxocysts were very common and densely packed [fig. 9]. Samples from the 1% & 2% nitrogen concentrations had similar toxocyst growth, however less densely arranged [fig. 10 & 11]. The 4% nitrogen concentration sample had a noticeable difference in toxocyst development [fig.12]. Fewer fully formed toxocysts were noticed, instead more spitzenkörper-like structures were seen [fig.12-a]. Further study is needed to determine if the spitzenkörper-like structures are in fact spitzenkörper (new hyphae growth) or if they are a result of delayed/ dormant toxocyst growth. Further study and observation is also needed to determine quantifiably if there were fewer toxocysts at higher concentrations. With this insight, the potential of using fungi for nematode population control can be further studied, realized, and applied.

#### Citations

Truong, B.-N., Suzuki, A., Truong, B.-N., Okazaki, K., Fukiharu, T., Takeuchi, Y., Futai, K., Le, X.-T., & Suzuki, A. (2007). Characterization of the nematocidal toxocyst in pleurotus subgen. Coremiopleurotus. *Mycoscience*, 48(4), 222–230. https://doi.org/10.1007/s10267-007-0358-4

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