SECTION FOR PARASITOLOGY AND AQUATIC DISEASES
FACULTY OF HEALTH AND MEDICAL SCIENCES
UNIVERSITY OF COPENHAGEN

Evaluation of soil microfungi as biological control agents against ascarid eggs

Sundar Thapa [1*], Nicolai V. Meyling [2], Kiran K. Katakam [1], Stig M. Thamsborg [1], Helena Mejer [1]

[1] Section for Parasitology and Aquatic Diseases, Department of Veterinary Disease Biology, Faculty of Health and Medical Sciences, University of Copenhagen, Denmark; [2] Section for Organismal Biology, Department of Plant and Environmental Sciences, Faculty of Science, University of Copenhagen, Denmark; *Email: sundarthp@gmail.com

Introduction

- Thick-shelled ascarid eggs may remain infective in the environment for several years, thus posing a prolonged risk of transmission to animals or humans

➔ **Aim:** to evaluate if microfungi (*Pochonia chlamydosporia* and *Paecilomyces lilacinus*) could reduce the viability of *Ascaridia galli*, *Toxocara canis* and *Ascaris suum* eggs under laboratory conditions

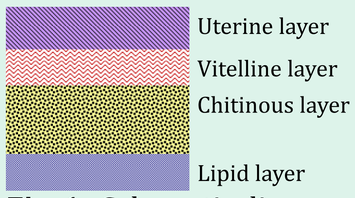


Fig. 1. Schematic diagram of ascarid egg shells.

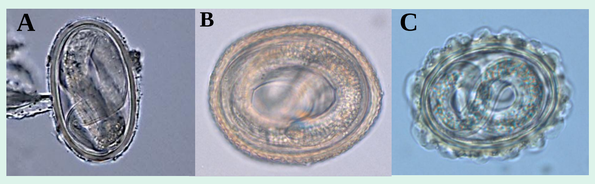
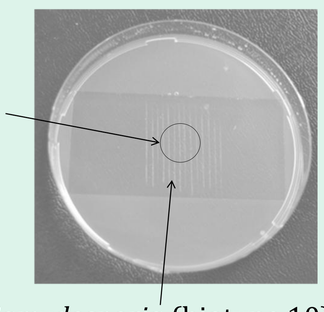


Fig. 2. Embryonated ascarid eggs: A. *Ascaridia galli*, B. *Toxocara canis*, C. *Ascaris suum*.

Materials and methods

4.9 cm^2 premarked area



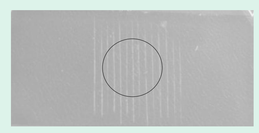
P. chlamydosporia (biotype 10) or *P. lilacinus* (strain 251) inoculated on 2% water agar (1.5 mm thick) covering a glass slide inside a Petri dish

Control Petri dishes were not treated with any fungus

For each parasite species: approx. 150 fresh eggs added to the premarked area on Petri dishes with and without fungi

Approx. number of eggs counted *in situ* using a stereomicroscope (day 0 count)

Incubation at 23°C

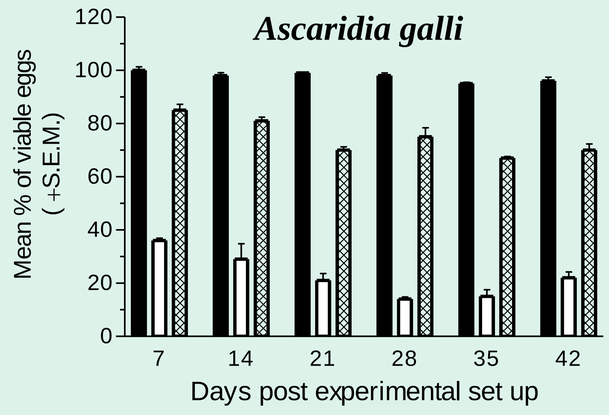


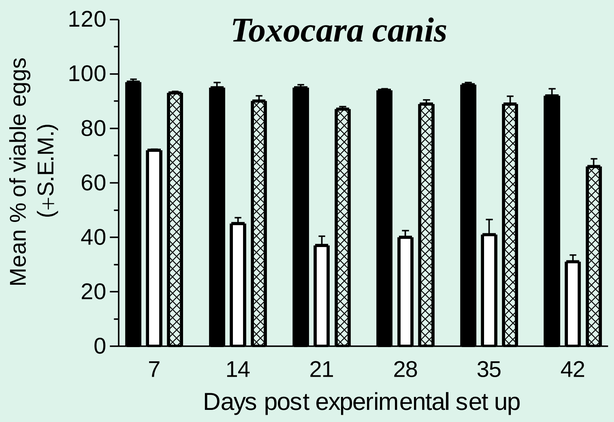
After 7, 14, 21, 28, 35 and 42 days, the glass slide was released from the Petri dish (n=3 for each time point and treatment)

Egg viability (= ability to embryonate) examined using compound and differential interference contrast microscopes

Percentage of viable eggs calculated in relation to day 0 count

Results





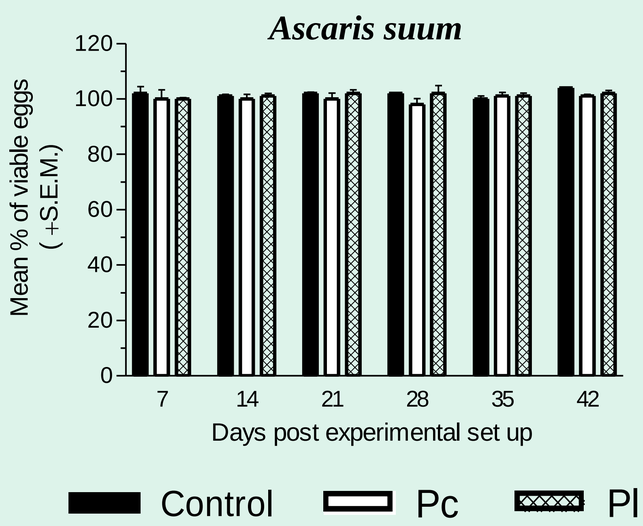


Fig. 3. Mean percentages (n=3, +S.E.M) of viable *A. galli*, *T. canis* and *A. suum* eggs on control, or a fungus (*P. chlamydosporia* (Pc) or *P. lilacinus* (Pl)) treated plates.

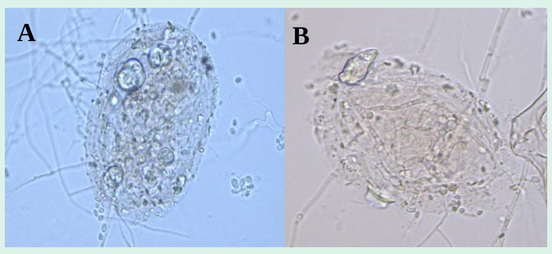


Fig. 4. *A. galli* eggs destroyed by *P. chlamydosporia* (A, B). Both shell and contents are degenerated.

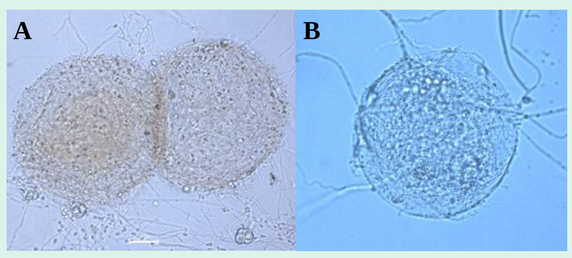


Fig. 5. *T. canis* eggs destroyed by *P. chlamydosporia* (A) and *P. lilacinus* (B). Both shell and contents are degenerated.

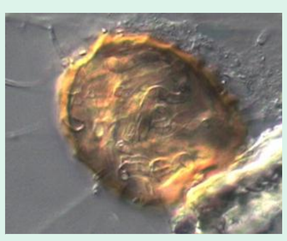


Fig. 6. *A. suum* egg destroyed by *P. chlamydosporia* . Most of the shell is intact whereas the content is degenerated and colonized by hyphae.

Discussion and conclusion

- *P. chlamydosporia* had a large negative impact on viability of *A. galli* and *T. canis* eggs whereas *P. lilacinus* had only a limited effect ➔ interspecies differences in the ability of fungi to destroy eggs
- *A. suum* eggs more resistant than *A. galli* and *T. canis* eggs to both fungi ➔ the thick uterine layer in *A. suum* eggs is perhaps important in protecting against the microfungi
- Shells and contents of *A. galli* and *T. canis* eggs were completely degraded ➔ enzymatic degradation of egg shell protein and chitin (Khan et al., 2004; Zhang et al., 2009) probably the primary mechanism of both fungi

➔ ***P. chlamydosporia* may be a potential biological control agent against *A. galli* and *T. canis* eggs in the environment**

Acknowledgements

Rothamsted Research (UK) and Prophyta (Germany) are thanked for kindly providing the fungal isolates *Pochonia chlamydosporia* biotype 10 and *Paecilomyces lilacinus* strain 251, respectively. The study was funded by the Green Development and Demonstration Programme (GUDP), the Danish AgriFish Agency, Danish Ministry for Food, Agriculture and Fisheries.

References

Khan, A., Williams, K.L., Nevalainen, H.K.M., 2004. Effects of *Paecilomyces lilacinus* protease and chitinase on the eggshell structures and hatching of *Meloidogyne javanica* juveniles. Biol. Control 31, 346-352.

Zhang, C., Wu, X., Cai, X., 2009. Effect of chitinases produced by *Pochonia chlamydosporia* on egg-hatching of *Meloidogyne incognita*. Sci. Agri. Sin. 42, 3509-3515.