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## Susceptibility of Poultry Red Mites to Entomopathogens

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**Abstract:** The killing efficacy of the entomopathogenic fungi *Beauveria bassiana* and *Trichoderma album* as well as the bacteria *Bacillus nigateria israelensis* against the poultry red mite, *Dermanyssus gallinae* De Geer, 1778 were experimentally evaluated. In spite of *Dermanyssus gallinae* is a major pest in the poultry production, no information is available on the susceptibility of these mite to entomopathogens. Therefore, infection experiments with two fungi and one bacterial species were carried out in the laboratory. *Beauveria bassiana* and *Trichoderma album* were tested in assays in which the mites were exposed to high doses of conidia. *Beauveria bassiana* and *Trichoderma album* caused high mortalities within the first 5 and 10 days of exposure as well as the bacteria *Bacillus nigateria israelensis*. *Trichoderma album* and *Bacillus nigateria israelensis* proved to be fatal against the poultry red mite *D. gallinae*, to our knowledge, this is the first record. *Trichoderma album* and *Beauveria bassiana* mixture in built-up litter were highly fatal to poultry red mite.

**Key words:** *Dermanyssus gallinae*, poultry red mite, entomopathogenic fungi, *Beauveria bassiana*, *Trichoderma album*, *Bacillus nigateria israelensis*

### INTRODUCTION

The red poultry mite, *Dermanyssus gallinae* De Geer, 1778, is an important haematophagous parasite that attacks the resting hens mainly at night for a short blood meal (Kirkwood, 1968; Lancaster and Meisch, 1986). After feeding, the mite hides in cracks and crevices, where they also mate and lay their eggs (Hearle, 1938). Under favorable warm and moist conditions, the life cycle can be completed in less than one week (Kirkwood, 1968; Nordenfors and Chirico, 1999). Red mite is currently one of the most detrimental ectoparasites in laying birds across several countries. The poultry red mite is the number one pest in poultry egg production and is currently almost impossible to control during the production cycle with traditional measure and decrease their welfare significantly during egg production may result in poorer hen performance, associated with reduced production and cause decreased feed intake and weight loss (Kirkwood, 1968; Williams, 2003).

Scientific information on the effects of poultry red mite on hens is incomplete as information is mainly sourced from the industry and is not well documented. Researchers agree that there are indications for the following effects of poultry red mite, which include, increased water intake in infested hens and lower egg production from the flock overall (Mul *et al.*, 2009).

Poultry red mite infestations have various negative effects on hens, an adult mite ingests approximately 0.2 µl blood (Sikes and Chamberlain, 1954) and high infestation rates of mites may cause anemia and mortality of the hen/host. Infested hens increase their production of new blood cells, but during periods of

rapid mite population growth, blood loss exceeds blood production capacity resulting in severe anaemia (Kilpinen *et al.*, 2005). Other negative effects of poultry red mite include high mortality, stress behaviour (higher levels of preening, head scratching and gentle feather pecking), lower body weight and reduced egg quality due to blood spots (Chauve, 1998). The productivity link could be that a severe mite infestation can increase mortality and as Arkle *et al.* (2005) showed, there is a direct effect of the size of the mite population on bird mortality. This of course means lower hock productivity; however, lower egg production per hen has not been found as a result of a mite infestation (Kilpinen *et al.*, 2005). Heavy infestations may cause considerable economic losses and welfare problems in the poultry production. Besides the direct physiological effect, chicken mites may also act as carrier of several important disease-causing agents, e.g. Salmonella (Zeman *et al.*, 1982), spirochaetosis (Hungerford and Hart, 1937) and encephalitis. Some of these survive in the mite for several months, thus forming a potential source of reinfection of new flocks, as the mite can live without feeding for up to 9 months (Nordenfors *et al.*, 1999). Chicken mites are also known to cause puritic dermatosis in humans (Baselga *et al.*, 1997) and may create serious problems for workers in the poultry industry due to the nuisance of mites crawling on the skin.

Furthermore, it is likely that the mite act as reservoirs for zoonotic bacteria since the mite will hide in the structure and thus be out of reach of the sanitation measures carried out between flocks (Sternberg *et al.*, 2005). Poultry red mite can have a serious impact upon human

health, apart from causing skin irritation and itching, the mites can cause allergic skin reactions (Sahibi *et al.*, 2008; Potenza *et al.*, 2008). The chemicals used to control poultry red mite may have adverse effects on humans as well, both directly, for workers exposed to chemicals and indirectly through consumption of poultry eggs containing pesticide residues (Hamscher *et al.*, 2003). Moreover, the eggs may have blood spots on the shells and would therefore be downgraded. This is caused when eggs roll over led red mites.

Control of red mite is typically undertaken by manual spraying of acaricides, although this method has limitations due to mite resistance and market withdrawal of many acaricides for environmental and toxicity concerns (Chirico and Tauson, 2002; Arthur and Daniel, 2003).

Scientists are attempt to develop a novel control method based on entomopathogenic fungi and semiochemicals to be combined in a trapping system, in which mites are inoculated with fungus and then disseminate the fungus to conspecifics. Entomopathogenic fungi seemed good candidates, recent development in pest control research have proves the urgent need for developing biological control methods with use of microbial pathogens (Steenberg and Kilpinen, 2003; Bhattacharyya *et al.*, 2004; Sternberg *et al.*, 2005). At present, an isolate of *Beauveria bassiana* has been selected and its mass production is being optimized (Sternberg *et al.*, 2005). To control poultry red mite, a fungus is needed that affects poultry red mite and/or its eggs and thus prevents their multiplication (Mul *et al.*, 2009). A very important aspect is safety to non-targets, such as humans, poultry and eggs, but the record of these fungi is excellent in this (Vestergaard *et al.*, 2003) and safe isolates will be available. The selected fungi should be able to survive in poultry red mite and the ecosystem of poultry red mite. *Trichoderma* spp. is a soil-borne fungal genus; fungal antagonists have been applied to strawberry flowers to inhibit infection by *Botrytis cinerea* in field experiments in Norway over several years (Strømeng *et al.*, 2005).

So, the objective of this study was a trial to evaluate the efficacy of *Beauveria bassiana*, *Trichoderma album* and *Bacillus Nigateria israelensis* on *Dermanyssus gallinae* stages as entomopathogens.

## MATERIALS AND METHODS

### Pathogenic microorganisms:

1. *Trichoderma album*. Source: Biozeid (Local) Company. It contains  $25 \times 10^6$  spores per mg.
2. *Beauveria bassiana*, Biosect. Source: KZ Company. It contains  $32 \times 10^6$  spores per mg. Quantification the spores were stained with neutral red according to La Peyer *et al.* (1995) the bright violet spores were considered as viable ones.
3. *Bacillus Nigateria israelensis* (Bioark, has a potency of  $32 \times 10^6$  per mg).

**Experiment I:** (Steenberg and Kilpinen, 2003) Two fungal species (*Beauveria bassiana* and *Trichoderma album*) and the bacteria *Bacillus nigateria israelensis* were tested against poultry red mite (*D. gallinae*).

Adult female chicken mites were selected from collected colonies of caged poultry farm to carry out the experiment (blood-fed 24 h prior to the exposure). Groups of 100 chicken mites (10 replicates per species of pathogens) were placed in Petri dishes with sporulating fungal cultures for 2-3 min. This ensured maximum exposure to fungal spores and bacterial cells. Control mites were left untreated. The mites were then transferred to glass tubes plugged with cotton wool and incubated in plastic boxes at 25°C/85 % RH for 16 days. Mortality was recorded at 1-3 day intervals.

### Experiment II:

- Effect of the fungi on poultry red mite in the built-up litter material inoculated by fungal spores: *Beauveria bassiana*, *Trichoderma album* and mixture of *Beauveria bassiana* and *Trichoderma album*.
- Effect of *Bacillus Nigateria israelensis* (Bioark, has a potency of  $32 \times 10^6$  per mg) on poultry red mite in the built-up litter material inoculated by *Bacillus cells*.

One ml of distilled water containing one mg of the tested organism was sprayed to 10 gms of sterile built-up litter (autoclaved) containing 200 adult red mites. The experiment was replicated 4 times. 200 adult red mites were kept as control. The glass tubes containing the litter and mites were plugged with cotton wool and incubated in plastic boxes at 25°C/85 % RH for 16 days; the mortality was recorded at 3 days intervals.

## RESULTS AND DISCUSSION

*Beauveria bassiana* caused 65% mortality within 5 days of inoculation in Experiment I (Fig. 1) and 80% mortality after 10 days of incubation in Experiment II (Fig. 2). *Beauveria bassiana* has potential effect on poultry red mite *D. gallinae*; Biosect (*Beauveria bassiana*) has a powerful effect against the poultry red mite after more than 10 days post exposure. Recent development in pest control research have proves the urgent need for developing biological control methods with the use of microbial pathogens (Steenberg and Kilpinen, 2003; Bhattacharyya *et al.*, 2004; Sternberg *et al.*, 2005). At present, an isolate of *Beauveria bassiana* has been selected and its mass production is being optimized (Sternberg *et al.*, 2005).

Steenberg and Kilpinen (2003) and Mul *et al.* (2009) suggested that, Entomopathogenic fungi infect their hosts through the cuticle, penetrate them and spread through the body and after the fungus has killed the mite, it can grow out of the mite cadaver and produce more spores, increasing the chance for other poultry red mite to be infested.

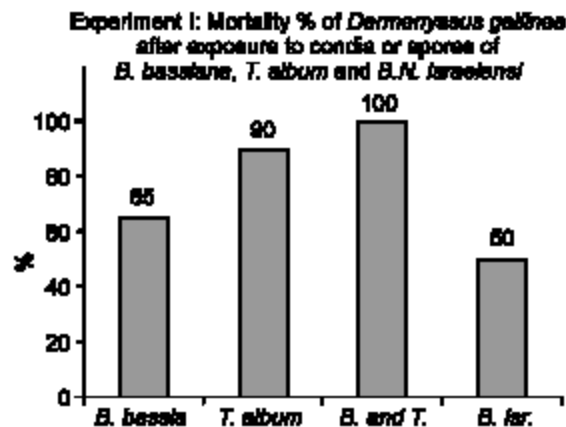


Fig. 1: Effect of the fungi (*Beauveria bassiana*, *Trichoderma album*) and the bacteria *Bacillus nigateria israelensis* on poultry red mite exposed to fungal spores and bacterial cells in Petri dishes containing sporulating cultures for 2-3 min

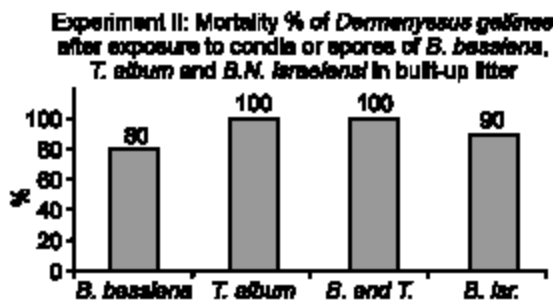


Fig. 2: Effect of the fungi on poultry red mite in the built-up litter material inoculated by fungal spores: *Beauveria bassiana*, *Trichoderma album* and mixture of *Beauveria bassiana* and *Trichoderma album* and the bacteria *Bacillus nigateria israelensis*

*Trichoderma album* caused 100% mortality within 5 days of inoculation in Experiment II and 90% mortality after 10 days of incubation in Experiment I.

*Trichoderma album* proved to be fatal against the poultry red mite *D. gallinae*, to our knowledge, this is the first record.

With respect to *Trichoderma album* we think that, the spores of the fungi germinate on the host cuticle and legs, penetrate them and spread through the body (Fig. 3). In addition, lytic enzymes secreted by the fungus may be played a role in the process of damage. After the fungus has killed the mite, it can grow out of the mite cadaver and produce more spores, increasing the chance for others.

Fungi in the genus *Trichoderma* have been used for the production of lytic enzymes and to control a wide range of plant pathogenic fungi. Dean *et al.* (1998) reported that the hydrolysis products from swollen chitin generated by *T. fungi* were chitinolytic enzymes. *Trichoderma fungi* have been applied to control tests of soil-borne plant pathogens. Their effectiveness is reported in many papers; fungal antagonists have been applied to strawberry flowers to inhibit infection by *Botrytis cinerea* in field experiments in Norway over several years (Strømeng *et al.*, 2005).

Héraux *et al.* (2005) evaluated the weed management potential of two cultural weed management techniques; cover cropping and fertility management, with allelochemical-releasing organism compared to herbicides. *Trichoderma virens* (= *Gliocladium virens*), which releases the herbicidal molecule viridiol, is applied in Composted Chicken Manure (CCM), *T. virens*-inoculated CCM provided significant weed control. Abd-Aziz *et al.* (2008) Found that, the *Trichoderma virens* UKM1 secretes a significant amount of exochitinase which degraded the chitinous materials of the shrimp shells.

*Trichoderma album* and *Beauveria bassiana* mixture ( $32 \times 10^6$  spores per mg,  $25 \times 10^6$  spores per mg, respectively) were highly fatal to poultry red mite (the mortality reached 100% within 7 days of inoculation in Experiment II), so *D. gallinae* appears susceptible to members of this particular group of entomopathogens. Entomopathogenic fungi infect their hosts through the cuticle and several species have wide host ranges. Therefore, entomopathogenic fungi may be expected to

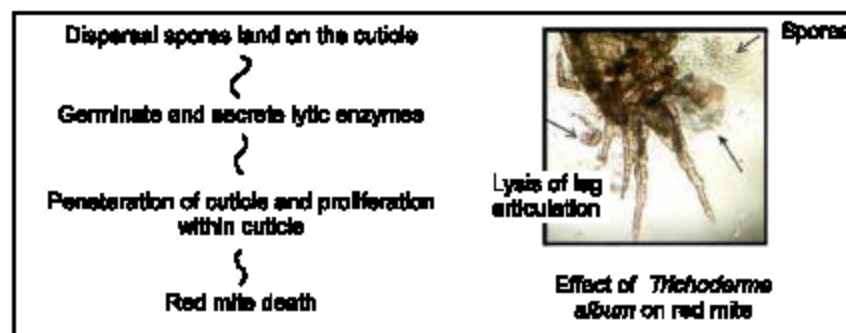


Fig. 3: Effect of *Trichoderma album* on poultry red mite (X100)

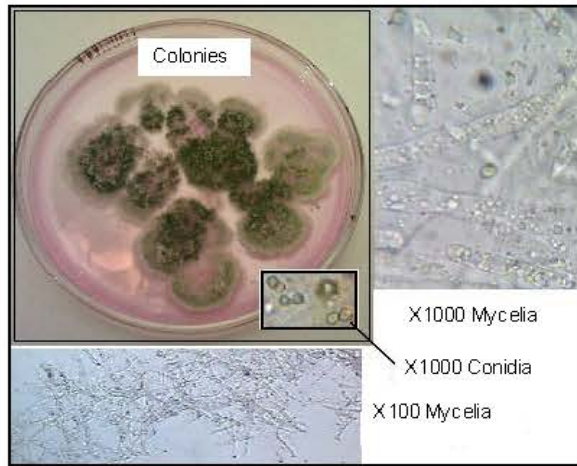


Fig. 4: *Trichoderma album* on culture and under microscope (X100 and 1000)

have a high control potential for blood-feeding mites like *D. gallinae* (Kilpinen, 2005).

*Bacillus Nigateria israelensis* caused 50% mortality within 5 days of inoculation in Experiment I (Fig. 1) and 90% mortality after 10 days of incubation in Experiment II (Fig. 2). Adult stage of the poultry red mite *D. gallinae* was susceptible to *Bacillus Nigateria israelensis* and the mortality of the Adult stage of the poultry red mite *D. gallinae* reached 90%.

**Conclusion:** Our data indicated that, (1) *Trichoderma album* and *Bacillus nigateria israelensis* proved to be fatal against the poultry red mite *D. gallinae*, to our knowledge, this is first record. (2) *Beauveria. Bassiana* has potential effect on poultry red mite *D. gallinae*. (3) *Trichoderma album* and *Beauveria bassiana* mixture in built-up litter were highly fatal to poultry red mite. (4) Biocontrol appears to have the potential to provide a successful eradication strategy for the future.

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