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Opportunities for integrated pest management to control the poultry red mite, Dermanyssus gallinae

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***Dermanyssus gallinae* is the most economically important ectoparasite of laying hens in Europe. Control of *D. gallinae* is already hampered by issues of pesticide resistance and product withdrawal. With the prohibition of conventional cages in 2012 and the resulting switch to more structurally complex housing which favours red mite, the importance of managing this pest is expected to increase. Integrated Pest Management (IPM), as often employed in agricultural pest control, may be a way to address these issues where a combination of different novel control methods could be used with/without conventional management techniques to provide a synergistic and more efficacious effect. Work at in our laboratory has shown that essential oils, including thyme and garlic, may act as effective *D. gallinae* repellents and acaricides, whilst preliminary vaccine studies have demonstrated a significant increase in mite mortality in vitro using concealed antigens. Work elsewhere has considered predators and fungi for *D. gallinae* control and other husbandry techniques such as manipulating temperature and lighting regimes in poultry units. This paper will review the available and emerging techniques for *D. gallinae* control and discuss which techniques might be suitable for inclusion in an integrated management programme (*e.g.* synthetic acaricides and diatomaceous earths).**

Keywords: Dermanyssus gallinae; integrated pest management; control

Introduction

The poultry red mite, *Dermanyssus gallinae,* is currently the most economically deleterious ectoparasite of laying hens in Europe (Chauve, 1998). Research has shown that in the UK between 60% (Fiddes *et al.*, 2005) and 85% (Guy *et al.*, 2004) of commercial egg-laying premises may be infected with *D. gallinae*, with higher mite

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populations typically seen in free range systems compared to cage units (Guy *et al.*, 2004; Fiddes *et al.*, 2005). This is of particular concern given that conventional cages will be prohibited in the EU from 2012 under EU Directive 99/74/EEC and thus the proportion of hens housed in alternative systems such as free-range is likely to increase substantially. *D. gallinae* prevalence in laying flocks worldwide may vary even more than that reported for the UK, from 20 to 90% depending upon the country and production system considered (Sparagano *et al.*, 2009). Evidence for higher infestation rates in free-range systems also appears to be country-dependant on a global scale, and factors such as flock/farm size may also be important in governing infestation rates (Sparagano *et al.*, 2009).

D. gallinae feeds on blood, preferring to feed from birds for a brief period every few days during periods of darkness, while the majority of their life-cycle is spent off the host. Populations of *D. gallinae* are capable of rapid expansion under the amenable conditions found in most poultry houses, where Axtell (1999) observed that complete development from egg to adult can occur in as little as two weeks. The turnover of hens in laying systems typically exceeds 70 weeks, allowing ample time for *D. gallinae* populations to proliferate (Höglund *et al.*, 1995). Where infestations of *D. gallinae* are sufficiently severe, mite feeding may result in significant stress to hens with subsequent declines in bird condition, growth rates, egg quality (through increased shell thinning and spotting) and egg production (Chauve, 1998). In extreme cases, *D. gallinae* population levels may be so high that anaemia and even death of hens can result (Wojcik *et al.*, 2000; Cosoroaba, 2001). It has also been reported that *D. gallinae* may serve as a vector for poultry pathogens and possible zoonoses such as *Salmonella spp*. (Valiente Moro *et al.*, 2009), suggesting a potential risk to human health.

Economic costs associated with both control and production losses due to *D. gallinae* have been estimated at €130 million annually for the EU egg industry (van Emous, 2005), where control of *D. gallinae* has typically been achieved by the use of synthetic contact acaricides including carbaryl, diazinon, dichlorvos and permethin. However, the continued use of these products may be hampered by issues of mite resistance (Beugnet *et al.*, 1997; Kim *et al.*, 2004, Fiddes *et al.*, 2005) and decreasing product availability as a result of tighter legislation regarding their use. Indeed, in Sweden there were no registered compounds available for the control of ectoparasites at the start of the last decade (Chirico and Tauson, 2002). Management of *D. gallinae* by conventional acaricides is further hampered by the behaviour of this pest. When not obtaining a blood meal, mites retreat to small cracks and crevices within the poultry housing system, where they are effectively protected from any acaricide that exerts its effect through contact toxicity.

With increasing reports of *D. gallinae* resistance to synthetic acaricides and changes in legislation and production practises, it is likely that in the future many more of the world's 2.8 billion laying hens (11.7% of which are located in the EU) (Axtell, 1999) will suffer as a result of *D. gallinae* infestation. With this in mind, it is important to identify and evaluate new approaches for the management of *D. gallinae* in poultry systems. Indeed, in 2006 an international seminar was convened to allow *D. gallinae* researchers to discuss current and future research and highlight knowledge gaps in areas such as mite life cycle, production impact, monitoring and control (Mul *et al.*, 2009). Research in the areas of novel acaricides, immunological control, biological control and animal/premise husbandry are all areas in which research is being undertaken in an attempt to control *D. gallinae*. Whilst promising results have been reported in all these areas, work to combine management techniques in an integrated approach to *D. gallinae* control remains wanting. Research focusing on crop pests suggests that such an Integrated Pest

Management (IPM) approach which combines control strategies can have numerous benefits over conventional control (*i.e.* the use of synthetic pesticides alone) (Dent, 1995).

The aim of this paper was to review the main alternatives to synthetic *D. gallinae* control currently being researched and developed and to assess if these would lend themselves to combination with one-another, as well as conventional synthetic acaricides, to achieve an integrated approach to *D. gallinae* management.

Novel acaricides

BIO-PESTICIDES

Bio-pesticides can comprise either bacterial endotoxins or the bacteria themselves. The bacteria *Bacillus thuringiensis*, which has known pathogenicity in insects, has been shown to increase mortality in ticks (Hassanain *et al.*, 1997) and has been used for the control of mites of agricultural crops (van der Geest *et al.*, 2000; Dolinski and Lacey, 2007). Biological insect toxins are now commonly used as crop sprays and have even been incorporated into crop plants via genetic engineering, with *B. thuringiensis* (*Bt)* and *Bt* maize being undoubtedly the best-known examples. Spinosad, a natural product derived from fermentation of the micro-organism *Saccharopolyspora spinosa*, is another such biopesticide that has been used successfully against a range of insect pests, with corresponding research also finding that this product possesses relatively low toxicity to mammals and birds with 70-90% of beneficial insects also left unharmed by exposure (Anastas *et al.*, 1999). Although the toxicity of spinosad to mites *per se* has been reported as being variable and/or reduced in comparison to insect species (Villanueva and Walgenbach, 2006; Holt *et al.*, 2006), experiments using this product against the ectoparasitic cattle tick *Boophilus microplus* have produced some promising results, although repeat applications might be necessary according to the work conducted (Davey *et al.*, 2001, 2005). Pathogenic bacteria of arthropods could provide an alternative means of mite control. This suggests that spinosad, along with other 'new-generation' bio-pesticides, may warrant investigation as premise sprays to manage *D. gallinae* populations.

INERT DUSTS

Inert dusts primarily include diatomaceous earths containing silica (although synthetic amorphous silica-based products also exist). They absorb lipids from the epicuticle of the exoskeleton of pest invertebrates, effectively resulting in death by dehydration. Whilst inert dusts have been well studied for their pesticidal potential and may provide an attractive alternative to synthetic chemicals for pest control, the quality of the raw material has been shown to be an important factor in the efficacy of the dust (Maurer and Perler, 2006). There is therefore a need to evaluate the efficacy of inert dusts under laboratory conditions to ascertain which specific products are likely to have the greatest benefit in *D. gallinae* management. Work by Kilpinen and Steenberg (2009) evaluated a range of commercial inert dusts against *D. gallinae* under laboratory conditions and similarly reported variation in efficacy depending upon the product used, as well as with variations in humidity. High humidity levels (>85%) were shown to reduce the efficacy of inert dust products, suggesting that if used in poultry units with such high humidity levels, measures such as increased application rates would be necessary.

PLANT-DERIVED PRODUCTS

Plant-derived products may offer an alternative to synthetic acaricides for managing *D. gallinae* populations and recent research in this field has produced some promising results (Kim *et al.*, 2004, 2007; Lundh *et al.*, 2005; Maurer *et al.*, 2009, George *et al.*, 2010). Several pesticides based on plant constituents are already used widely in certain areas of pest management (Isman, 2006), including against pests of veterinary significance (George *et al.*, 2008). Products based on extracts from the neem tree (particularly its seeds), for example, are commonly employed in pest management *per se*. Neem oil has been reported to have biocidal effects against some 200 species of arthropod pests (Choi *et al.*, 2004), including ticks (Pathak *et al.*, 2004) and *D. gallinae* (Lundh *et al.*, 2005). Preliminary work by Kim *et al.* (2004) tested 56 plant essential oils for their acaricidal effect on *D. gallinae*. Of these, oils from bay, cade, cinnamon, clove bud, coriander, horseradish, lime dis 5F, mustard, pennyroyal, pimento berry, spearmint and thyme all gave 100% mite mortality in contact toxicity tests at a concentration of 0.07 mg of oil per cm^2. Further experiments by Kim *et al.* (2004) showed that the acaricidal effect of selected essential oils was attributable to action in the vapour phase, an observation supported in later work from the same group (Kim *et al.*, 2007). In similar work by George *et al.* (2010), 50 plant essential oils were assessed for their toxic effect on *D. gallinae*. Twenty of the essential oils chosen gave greater than 80% mite mortality over 24 hours when used at a concentration of 0.14 mg/cm^3, with one in every five of the 50 essential oils used, including thyme, tea tree and garlic, giving 100% mortality.

Vaccines

Observations of mite population levels in poultry flocks infested with *D. gallinae* tend to suggest that birds do not develop resistance to the mites (Nordenfors and Höglund, 2000), while levels of IgY in mite-infested poultry flocks were not related to mite levels (Arkle *et al.*, 2006). Mite-induced immuno-modulation of the host is a possibility (Jackson *et al.*, 2009) and recent research suggests that *D. gallinae* both adapts its reproductive behaviour to the host (Harrington *et al.*, 2010a) and modulates the host initial inflammatory responses (Harrington *et al.*, 2010a,b). An understanding of the immunology of the *D. gallinae*-host relationship is important where vaccination is proposed as a control measure, yet despite the importance of *D. gallinae* in poultry production there is a lack of understanding of how the mite modulates and/or stimulates host immunity.

Vaccines provide attractive alternatives to insecticides for the same reasons as they are preferable to antibiotics, including lack of residues in foodstuffs, no withdrawal periods, no environmental contamination, avoidance of resistance in target populations and ease of administration (Shryock, 2004). Until recently, there were few reports in the literature detailing the development of a *D. gallinae* vaccine or prospective vaccine candidates.

Immunisation of birds with somatic *D. gallinae* antigens has met with variable success. Arkle *et al.* (2008) found no significant difference in *D. gallinae* mortality when fed *in vitro* on blood from birds immunised with *D. gallinae* proteins and controls. However, both Wright *et al.* (2009) and Harrington *et al.* (2009a) reported a significant 7.5% or 50.6%, respectively, increase in *in vitro* mite mortality when fed blood spiked with egg-extracted antibodies from *D. gallinae* immunised birds. However, both authors used different methods of protein extraction; Harrington *et al.* (2009a) used a urea-based method, whilst Wright *et al.* (2009) identified significant mite mortality using PBS-extracted proteins but reported no significant effect of urea-extracted *D. gallinae* proteins on mite mortality.

A genomics approach has also been undertaken to investigate *D. gallinae* vaccine candidates. Bartley *et al.* (2009) identified an orthologue of tick histamine release factor (HRF) in *D. gallinae* and suggested the protein could have a regulatory function in mites. Following production of the recombinant protein *Dg*-HRF-1, *in vitro* testing against *D. gallinae* demonstrated a significant 7% increase in mite mortality compared to controls. Harrington *et al.* (2009b) immunised laying hens with recombinant proteins derived from ticks (Bm86) or mosquitoes (subolesin). Bm86 has well documented activity against *Boophilus microplus* (de la Fuente *et al.*, 2007), although non-*Boophilus* spp. activity is variable (Willadsen, 2004), whilst subolesin has been shown to be highly conserved across a number of arthropods, including ticks, and can affect blood meal digestion and reproduction in ticks (de la Fuente *et al.,* 2006; Kocan *et al.*, 2007). Harrington *et al.* (2009b) demonstrated a significant 35.1% increase in *D. gallinae* mortality in subolesin-immunised versus control birds in an *in vitro* assay, whilst immunisation with Bm86 resulted in a non-significant, but numerical 23% increase in mite mortality.

The development of arthropod vaccines is notoriously difficult due to the limiting step of identification and characterisation of new protective antigens, and this has restricted the availability of commercial vaccines against arthropod ectoparasites (reviewed by Willadsen, 2004). The recent research on *D. gallinae* antigens has demonstrated that there is potential to develop a vaccine to control the poultry red mite. However, the approaches undertaken have so far relied upon the concealed antigen approach, whereby the host immune system is not normally exposed to mite antigens, for example gut epithelial cell, and as such the parasite has not developed hot-immune avoidance mechanisms. This approach is not without pitfalls, most notably the necessity to repeatedly vaccinate animals due to the lack of stimulation of host immunity by these concealed antigens.

Biological control

NATURAL ENEMIES

Natural enemies in the form of predators and parasitoids can be of great benefit in controlling crop pests (Gurr *et al.*, 2004). Where naturally occurring populations are insufficient to provide pest management benefits, mass releases of artificially reared individuals can be used to reduce pest populations. Such releases are especially effective in enclosed systems (such as greenhouses), where the natural enemies are confined to the release site and so must necessarily feed/parasitize the target pest in order to survive. Whilst little work has been done to assess the potential of predators and parasitoids to control *D. gallinae* populations, the fact that hens are typically housed in at least partly enclosed systems suggests that such control might be achievable if suitable candidates are identified for mass rearing and release.

Research data exists that suggest that suitable candidate predators may exist for use against *D. gallinae* in poultry systems. Numerous authors have reported the occurrence of the predatory mite *Cheyletus eruditus* in poultry houses where this species has been observed feeding on juvenile *D. gallinae* (Lesna *et al.*, 2009). Releases of *C. eruditus* have not provided control of *D. gallinae* in experiments to date, although additional research has identified a further two predatory mite species (*Hypoaspis aculeifer* and *Androlaelaps casalis*) with *D. gallinae* management potential (Lesna *et al.*, 2009). It is possible that other predatory species aside from mites would also prey upon *D. gallinae*, although mass-rearing and release of species such as predatory beetles would be more difficult to achieve. Similarly, whilst parasitoids are often mentioned with regard to crop pest control, many of these species (such as the parasitic Hymenoptera) require additional resources to survive and reproduce (such as pollen and nectar) that would not be present within a poultry unit.

ENTOMOPATHOGENIC FUNGI

Entomopathogenic fungi are currently used worldwide for control of a wide range of arthropod pests, particularly pests in protected crops or field crops. *D. gallinae* has been found to be susceptible to infection by isolates of each of the species *Beauveria bassiana*, *Metarhizium anisopliae* and *Paecilomyces fumosoroseus* when mites were inoculated with high doses of conidia (Steenberg and Kilpinen, 2003). In subsequent work on the same project isolates from *B. bassiana* consistently proved to be most virulent and persistent over time, with up to 80% transmission when 5% of mites had been treated (Steenberg *et al.*, 2006). Nevertheless, semi-field experiments showed that although the fungus was capable of reducing *D. gallinae* population growth, control levels were not satisfactory (Steenberg *et al.*, 2006). This probably results from the fact that the time required to kill *D. gallinae* was sufficiently long to allow females to oviposit, where treatment was reported not to have effected oviposition (Steenberg *et al.*, 2006). Tavassoli *et al.* (2008) reported that while three strains of *M. anisopliae* were effective in killing adult *D. gallinae*, pathogenicity against different mite developmental stage varied according to fungal strain as well as being dose and time dependent. Use of entomopathogenic fungi may also be dependent on maintaining adequate humidity levels to ensure fungal transmission and it remains to be seen if this method can be successfully developed and employed in the field for *D. gallinae* control.

Animal husbandry

Higher mite populations are typically seen in free range systems compared to cage units (Guy *et al.*, 2004; Fiddes *et al.*, 2005; Arkle *et al.*, 2006) probably as these systems provide both a more favourable habitat for the mites and a greater challenge for decontamination between flocks. Such systems often contain a large number of sites highly suited as *D. gallinae* refugia. When not obtaining a blood meal, mites retreat to these small cracks and crevices where they can easily survive unfed for many months in areas that are difficult to clean down between flocks. It should be possible to design poultry facilities that are less ‘mite-friendly’ than is currently the norm, although removing all refugia is clearly unrealistic. Recent research suggests that using a Hazard Analysis and Critical Control Point method (HACCP) can be potentially beneficial in preventing *D. gallinae* establishment in all types of hen housing where Mul and Koenraadt (2009) have adapted this technique from that first development by NASA in the 1960s. Forty-one potential infestation hazards are cited, such as introduction and spread of mites by birds, rodents and employees, with suggested corrective actions to minimise *D. gallinae* population establishment. The system has been trialled in Holland and the UK and described as a useful and feasible management tool by poultry farmers.

Mul and Koenraadt (2009) noted the potential of temperature manipulation to control *D. gallinae*, where heating the house between flocks to 55°C was suggested as a control measure. *D. gallinae* population development is known to be temperature dependant, where research suggests that the stage-specific survival of immature mites decreases rapidly outside the range of 10-37°C (Maurer and Baumgärtner, 1992). Tucci *et al.* (2008) similarly reported high mortality of *D. gallinae* at 35°C indicating that this temperature had adverse effects on mite development. Complete mortality of *D. gallinae* can be expected at greater than 45°C and below -20°C (Nordenfors *et al.*, 1999). This suggests that temperatures of 55°C, which would be expensive to reach and maintain for any length of time, may be excessive for controlling *D. gallinae* and that similar levels of control might be achievable at a reduced temperature (although this might need to be maintained for longer).

It may be possible to control *D. gallinae* populations by manipulating lighting regimes within poultry units. Research suggests that short-cycle intermittent lighting regimes can markedly reduce *D. gallinae* numbers compared with more standard regimes, although lighting intensity (Zoons, 2004; Stafford *et al.*, 2006). This is likely caused by disruption to the mites' normal nocturnal feeding behaviour, although exact reasons remain unclear. However, current EU legislation on hen welfare requires a statutory 8 hour dark period, making it difficult to envisage how intermittent lighting regimes could be employed in practise for *D. gallinae* control.

IPM potential for *Dermanyssus gallinae* control

Many of the different *D. gallinae* management approaches previously described would be amenable to use alongside one-another, as well as in combination with conventional currently used hygiene and chemical control methods. It is likely, for example, that a *D. gallinae* vaccine could, and depending upon the antigen it might be preferable, to use in conjunction with any other control method described. Similarly, the use of novel pesticides would unlikely be unaffected by advances in animal husbandry techniques and *vice versa*, with both being compatible with conventional *D. gallinae* control. Nevertheless, such compatibility would not be true of all the *D. gallinae* control methods described.

Novel pesticides may be just as toxic to invertebrate natural enemies as they are to the target pest. Inert dusts work via the invertebrate cuticle and so can be expected to exert a broad-spectrum effect on all invertebrates. Plant essential oils are thought to disrupt binding of invertebrate nerve cord proteins, specifically ^{3}H-octopamine (according to work done with the American cockroach *Periplaneta Americana*) (Enan *et al.*, 1998). As such, any product giving high levels of mortality in *D. gallinae* might similarly be expected to do so in predators and parasitoids (also possessing an invertebrate-specific octopaminergic nervous system), although some work exists to suggest that toxicity to invertebrates may depend on the species considered (Isman, 2000; George *et al.*, 2009). Bio-pesticides may be more species specific and amenable to use alongside natural enemy releases. For example, research suggests that predatory invertebrate species may be relatively tolerant to spinosad, although parasitoids are at much greater risk from exposure (Williams *et al.*, 2003). Use of invertebrate-specific bio-pesticides (and inert dusts) is also likely to be amenable with treating *D. gallinae* infestations with entomopathogenic fungi (although it is plausible that dusts could reduce premise humidity levels). The same may not be true for essential oil-based acaricides, however, where these products are often highly fungicidal. Similarly, animal husbandry techniques involving raising premise temperatures to levels lethal to *D. gallinae* between flocks would likely have an adverse effect on any beneficial organisms present making these techniques incompatible with one-another.

Conclusions

It remains to be seen how many of the new and emerging *D. gallinae* management techniques cited herein will be as useful in practice as preliminary results suggest. It will also be interesting to observe if any synergistic effect could be gained by combining any compatible techniques in an IPM approach to *D. gallinae* control. Even where techniques are amenable to combination with one-another, this will not automatically infer any pest management benefit in using multiple techniques together. In work by Maurer *et al.* (2009), for example, mortality of *D. gallinae* exposed to a diatomaceous earth was comparable with or without the addition of 2% pyrethrum extract. Whilst beyond the scope of this paper, as an integrated approach to pest management can also encompass the use of 'conventional' acaricides, the development of new synthetic products such as Phoxim (marketed as 'ByeMite®') should not be overlooked when considering IPM for the control of *D. gallinae*.

Although any consideration of IPM for use against *D. gallinae* remains in its infancy, according to pest management research in other areas, developing methods that could be combined in an integrated approach could be beneficial to *D. gallinae* management in the future. With continued research into new technologies and techniques for *D. gallinae* control an IPM approach to control this important poultry pest has the potential to be realised.

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