The Old School, Brewhouse Hill, Wheathampstead,
Hertfordshire AL4 8AN, UK

Animal Welfare 2010, 19: 265-273
ISSN 0962-7286

The effect of essential oils showing acaricidal activity against the poultry red mite (Dermanyssus gallinae) on aspects of welfare and production of laying hens

DR George*†‡, OAE Sparagano‡, G Port§, E Okello‡, RS Shiel‡ and JH Guy‡

† Lancaster Environment Centre, Lancaster University, Lancaster LA1 4YQ, UK
‡ School of Agriculture, Food and Rural Development, Newcastle University, Newcastle upon Tyne NE1 7RU, UK
§ School of Biology, Newcastle University, Newcastle upon Tyne NE1 7RU, UK
* Contact for correspondence and requests for reprints: d.george@lancaster.ac.uk

Abstract

*The poultry red mite (*Dermanyssus gallinae*) causes severe welfare concerns for laying hens arising from anaemia and disease transmission, and has been identified as an associated risk factor in cannibalistic feather pecking. Previous work suggests that essential oils may offer an alternative to synthetic acaricides to control* D. gallinae*. Such alternatives are needed due to the limitations of synthetic acaricides (eg availability, resistance, environmental concerns and product residues). The aim of the current study was to ensure that selected essential oils have no negative impact on either hen welfare or egg production. To achieve this aim, small groups of laying hens were confined in poultry huts for a period of eight weeks during which time the interior of the huts was sprayed at weekly intervals with one of four different treatments: i) Thyme essential oil at 5× the LC_{90} level (the concentration of oil previously found to kill 90% of* D. gallinae *under laboratory conditions) for* D. gallinae *in 500 ml of water; ii) Pennyroyal essential oil at 5× the LC_{90} level for* D. gallinae *in 500 ml of water; iii) Solvent-only (huts treated with 500 ml of water); and iv) Pseudo-spray where huts were not treated with any product, but subjected to sham-spraying. The results suggest that pennyroyal essential oil would not be suitable for further development as an acaricide for* D. gallinae*, since this treatment had to be terminated early in the study period as a result of concerns about the welfare of hens exposed to this oil. Conversely, there were few differences in feather condition, hen weight, feed intake, feeding efficiency, egg production or egg weight between thyme-treated huts and huts that were either pseudo-sprayed or sprayed with solvent-only (water). It is concluded that thyme essential oil is a promising candidate for further development as an acaricide for* D. gallinae *to help safeguard the welfare of laying hens in commercial poultry systems.*

Keywords: *acaricide, animal welfare,* Dermanyssus gallinae*, egg production, essential oil, laying hen*

Introduction

The poultry red mite (*Dermanyssus gallinae*, De Geer), although not entirely species-specific, is seen most frequently in systems for laying hens, largely due to the lengthy turnover of the flock (approximately 72 weeks) which allows time for large mite populations to become established (Höglund *et al* 1995). The consequences of infestation by *D. gallinae* are severe for the welfare of laying hens. The feeding mite can cause irritation, restlessness and either mild or severe anaemia, occasionally resulting in death (Wojcik *et al* 2000; Cosoroaba 2001). *D. gallinae* are also a threat in the spread of disease, since they may act as a vector for a number of pathogenic poultry infections, both viral and bacterial (Chirico *et al* 2003; Valiente Moro *et al* 2009). Behavioural observations have also shown increases in cannibalistic feather pecking associated with *D. gallinae* infestation (Kilpinen 1999). In a study of a caged housing system, mortality of birds rose from 1 to 4% due to parasitism by *D. gallinae* and egg production was reduced by approximately 10% (Wojcik *et al* 2000). Similar figures from another study of caged hens recorded a significant decrease in egg production levels (95 to 75%) and an increase in mortality from 5 to 52% (Cosoroaba 2001) associated with infestation by *D. gallinae*. Economic sustainability is also affected through reduced growth rates of growing hens, reduced egg production and reduced egg quality (poor shell integrity and blood staining of the shell surface) (Urquhart *et al* 1996; 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 populations typically seen in free-range systems compared to cage units (Guy *et al* 2004; Fiddes *et al* 2005; Arkle *et al* 2006). This is of particular concern given that conventional cages will be prohibited in the EU from 2012 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 more significantly (20–90%) 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).

The most common form of control of *D. gallinae* is by the application of synthetic pesticides. However, the number of effective pesticides registered for application in poultry houses is relatively low for a number of reasons, including development of mite resistance (Beugnet *et al* 1997; Kim *et al* 2004; Fiddes *et al* 2005), chemical and antibiotic residues in food and undesirable environmental effects (Dalton & Mulcahy 2001). In addition, the tendency of *D. gallinae* to occupy small cracks and crevices in the poultry house and their ability to survive for extended periods without taking a blood-meal (Axtell 1999), alongside their prolific reproduction capacity and short lifecycle, make eradication very challenging (Kilpinen 2001). As a result, in addition to being a welfare issue for hens, *D. gallinae* is considered to be the most economically deleterious parasite of laying hens in Europe (Chauve 1998) which costs the EU industry an estimated €130 million per annum in control and production losses (van Emous 2005). If viable alternatives to synthetic control products are not sought 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. Research has also suggested that *D. gallinae* may obtain a blood-meal from a range of alternative hosts, including horses (Mignon & Losson 2008), rodents (Lucky *et al* 2001) and man (Bruneau *et al* 2001), where these mites have recently been linked to dermatological disorders such as pseudoscabies (Cafiero *et al* 2008). Effective means of controlling this pest may thus be of importance for the welfare of other species as well as hens, ourselves included.

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; George *et al* 2009, 2010a,b; Maurer *et al* 2009). Several pesticides based on plant constituents of one kind or another are already used widely in certain areas of pest management (Isman 2006), including against pests of veterinary significance (George *et al* 2008a). 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 *D. gallinae* (Lundh *et al* 2005).

In previous work by George *et al* (2010a), 50 plant essential oils were assessed for their toxic effect on *D. gallinae*. In this same work, around half of these oils were selected based on their high initial toxicity to *D. gallinae* and taken forward for testing at various concentrations. The most promising seven essential oils were then tested at different environmental parameters and against brine shrimp, mealworm beetles and juvenile as well as adult mites, to assess their effect on *D. gallinae* life-stages and non-target organisms (George *et al* 2010b). From these data, and in conjunction with data obtained on the repellence of various essential oils to *D. gallinae* (George *et al* 2009), it appears that thyme and pennyroyal essential oils could be promising *D. gallinae* pest-management products. Whilst this is encouraging, especially as essential oils are typically environmentally non-persistent and many are known to possess extremely low mammalian toxicities (Isman 2006), research is still needed to confirm that these essential oils have no negative effect on hen welfare before they can be recommended for development as *D. gallinae* acaricides and/or repellents. Therefore, the aim of this experiment was to evaluate the effect of thyme and pennyroyal essential oil on hen welfare and egg production.

Materials and methods

The experiment described below was conducted under Home Office Licence and local guidelines in place at Newcastle University, UK.

Experimental design

Huts used to house experimental hens were arranged in an open-fronted shed at Cockle Park Farm (Newcastle University, Morpeth, UK) in a randomised block design according to treatment. Four treatments were used as follows: i) Thyme — huts treated weekly with thyme essential oil at 5× the LC_{90} level for *D. gallinae* in 500 ml of water; ii) Pennyroyal — huts treated weekly with pennyroyal essential oil at 5× the LC_{90} level for *D. gallinae* in 500 ml of water; iii) Solvent-only — huts treated weekly with 500 ml of water; and iv) Pseudo-spray — huts not treated with any product, but subjected to sham-spraying. The LC_{90} for thyme and pennyroyal (ie the amount of these oils required to kill 90% of *D. gallinae* over 24 h) had been previously identified in laboratory assays by George *et al* (2010a) as 0.044 and 0.105 µL cm^{-3}, respectively. Essential oils were sourced from New Directions, Southampton, UK. All huts were separated from one another by a space of approximately 1 m.

Materials and animals

Sixty-four Lohmann Brown pullets, previously reared by a commercial supplier in a deep litter house with access to perches, were used for this experiment. Birds were exactly 16 weeks old when delivered and had been beak-trimmed at eight days of age and vaccinated against a range of diseases (including *Salmonella*, Newcastle disease and coccidiosis).

The birds were housed in 16 poultry huts. Huts measured 120 × 90 × 82 cm (length × depth × height) at the front (with a roof that sloped down towards the back of the hut to a height of 66 cm). Huts were of the 'Silver Jubilee Free Range Poultry House' type (WS Hodgson and Co Ltd, Cotherstone, UK), and were treated with Spray and Protect Timbercare (Wickes, UK). Each hut was fitted with a 40 W lamp (attached to a wire-mesh ventilation window above the access door) where light regimes were set at 14: 10 h,

https://doi.org/10.1017/S0962728600001640 Published online by Cambridge University Press

light:dark with lights coming on at 0700h to allow for periods of low light in the huts before and after artificial lights were turned on and off. Huts also contained a perch (90-cm long), a nest box (approximately 60-cm wide and divided into two compartments) and a pop hole. The pop-hole was fixed open and covered with wire mesh to improve ventilation. Birds were not permitted to leave the hut during the experiment to ensure equal exposure to treatments in all huts. A 3 L circular drinker and 3 kg feed hopper, attached to the inside of the access door, were provided in each hut. Feed (a commercial pelleted layer ration of 18% Farmgate Layers Pellets from BATA, North Yorkshire, UK) and water were available *ad libitum*. A covering of sawdust (1–2-cm deep) was used as a substrate in each hut and replaced at the end of each experimental week.

Experimental procedure

Groups of four pullets were chosen at random, placed into each of the 16 poultry huts and fitted with 12-mm coloured plastic leg rings for identification. Due to constraints on the equipment needed to run the experiment, it was necessary to stagger the days on which data were collected so that each week half of the huts (selected at random on a per block basis) were treated on one day and the other half the following day. Spray treatments were applied between 1100 and 1200h and the drinkers were removed during spraying to minimise the risk of contamination of the birds' water supply. The walls (excluding the back of the door) and ceiling of the hut interior were sprayed during treatment application, where the total volume of liquid used dictated that hut interiors were sprayed to the point of run-off. The substrate and nest box were not sprayed directly, although they may have been subject to drift during the treatment process. This procedure for spray application was chosen to mimic the way in which conventional acaricides are applied to poultry facilities, where nest boxes are generally inaccessible for direct treatment, especially when birds are in lay. For the first two spray dates, all huts were pseudo-treated (no product in atomisers) to acclimatise birds to the treatment process and allow baseline data to be collected. After this time, huts were sprayed with product according to treatment, except for pseudo-spray control huts which continued to be pseudo-sprayed, on a weekly basis for six consecutive weeks.

Data collection

Upon delivery, hens were given one week to settle in prior to the start of the study period. At this time (week 0), and at weekly intervals thereafter for the remainder of the study period (weeks 1–8), hens were weighed and their feather condition scored using a subjective scale from 0 (intact feathers) to 5 for 11 independent body regions (where a maximum score of 55 could theoretically be obtained) according to the method presented by Bilčík and Keeling (1999). Feed remaining in the trough at this time and at weekly intervals thereafter was weighed to provide data on feed intake (where the feed added to the trough throughout the week was recorded), estimated from feed disappearance from the trough, and to allow for subsequent estimation of feed efficiency. Feed efficiency was calculated on a weekly basis by dividing the total weight gained per hut by the total feed intake from that hut to provide a feed efficiency index. From the start of the study period, daily records were made of the maximum and minimum temperatures in each hut (all huts were fitted with a max/min thermometer) and the number of eggs laid in each hut. Eggs collected from huts were returned to the laboratory and weighed. Daily maximum/minimum temperatures during the study period, averaged across all huts, peaked at 26 and 9ºC, respectively. Welfare data (feather condition and bodyweight) were collected each week on the day prior to product application, with the last data collection taking place six days after the final spray date, at which time the experiment was terminated.

Statistical analysis

All statistical analyses were performed using Minitab (v 15, Minitab Inc, State College, USA). Differences over time in the measured variables were not considered as it was expected that these would vary throughout the study due to ongoing development/growth of the young hens used. Furthermore, excluding time as a factor in any global analysis allows easier interpretation of data from individual weeks in isolation. Individual sampling dates were thus considered separately from one another in the analyses.

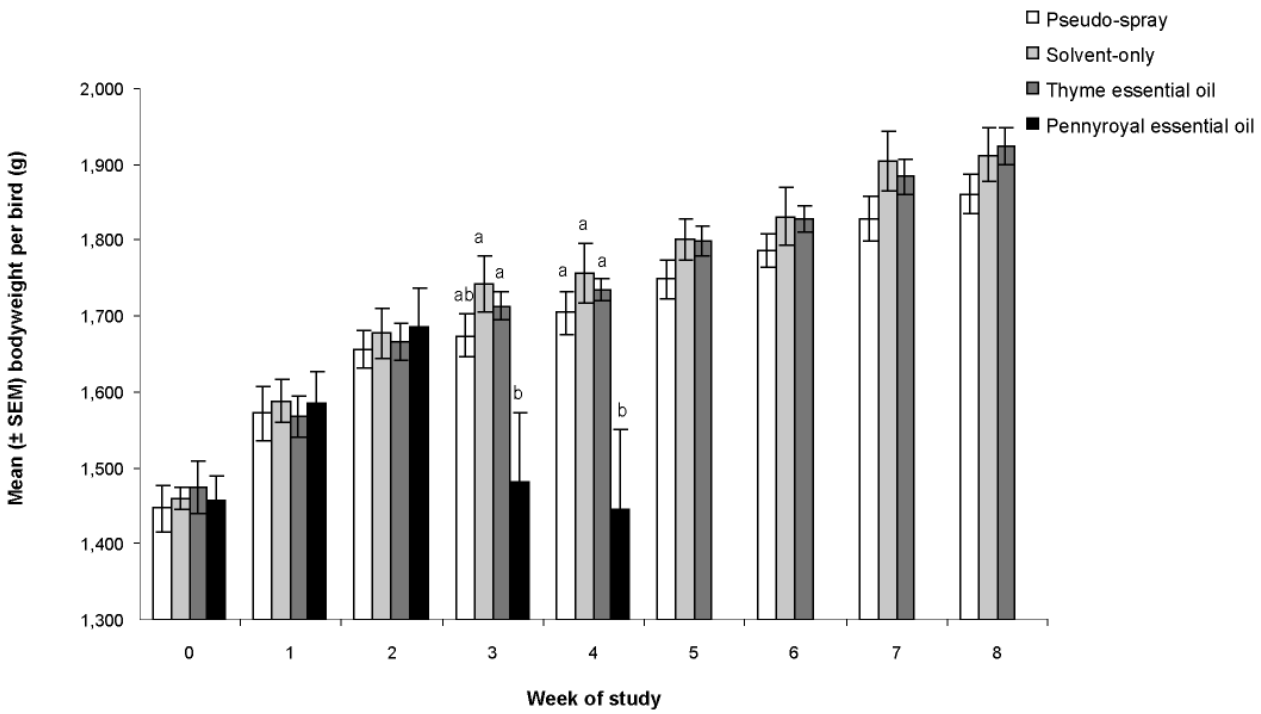
For feather condition, the average weekly score from all birds in a given hut was calculated and analysed between treatments using the Kruskal-Wallis test (adjusted for ties) as data could not be considered continuous for parametric analysis. Scores for all body regions for any individual hen were totalled. Feather scores were notably low (zero) in weeks 0–1 of the study period. As a result, only data from weeks 2–8 were analysed. For bodyweight and body-weight gain, the average weekly weight and weight gain of hens per hut were taken and subjected to analysis of variance (ANOVA) where treatment and experimental block were considered as factors. Results from week 5 for bodyweight gain required cube-root transformation to fit the residuals from the ANOVA to a normal distribution. Results on feed intake (per bird per day) were analysed as for hen weight. In no instance did this data require transformation. Results on feed efficiency (weight gained/feed intake per hut) were also analysed in this way. Indices calculated from data obtained in weeks 1 and 5 were cube-root transformed prior to analysis.

Results on egg production were not considered to be continuous in nature and so were analysed by non-parametric methods. The average number of eggs produced per bird per day from huts under different treatments was compared using the Kruskal-Wallis test (adjusted for ties). Data on egg weight were analysed by obtaining an average egg weight (per week per hut) and then subjecting this data to ANOVA as described previously. Egg laying did not commence in some huts until into the third week of the study, so only data from weeks 3 to 8 were analysed.

Where a significant difference was identified between treatments using ANOVA, *post hoc* testing was conducted using Tukey's test. Where *post hoc* testing was required following the Kruskal-Wallis test this was done using Mann-Whitney *U* tests.

https://doi.org/10.1017/S0962728600001640 Published online by Cambridge University Press

Figure 1



Mean (± SEM) bodyweight for hens housed in poultry huts subjected to different spray treatments (n = 4). Within a given week, means not sharing a common letter are significantly different according to the Tukey's test ($P < 0.05$).

Results

Where data required transformation prior to analysis, all graphs displayed show means and individual treatment standard errors derived from the original data. Due to concerns over hen welfare, it was necessary to terminate the pennyroyal treatment soon after the commencement of spraying with this product (directly after treating in week 4 of the study period). Results obtained from pennyroyal-treated hens are thus not available after week 4 of the study. For reference, data collected after spraying with product commenced are shown from week 3 onwards, where product-spraying was begun the day after collection of week 2 data.

Mortality

Mean mortality (% birds per hut which died) was 12.5% for the pennyroyal treatment, and 0.0% for the other three treatments. However, all birds in the pennyroyal treatment were subsequently euthanased in week 4 due to concerns over toxicity.

Feather condition

At no point during weeks 2–8 of the study period was there any significant difference ($P < 0.05$) in feather condition between treatments. Mean feather scores per bird were low in weeks 1–4 (< 2), although they had increased by week 8 (6–7) in all treatments.

Bodyweight

Significant differences between treatments in the bodyweight of hens were recorded in weeks 3 ($F_{3,9} = 5.07$, $P < 0.05$) and 4 ($F_{3,9} = 6.92$, $P < 0.05$). In both cases, this result was due to a decline in the bodyweight of hens subjected to treatment with pennyroyal essential oil (Figure 1).

Bodyweight gain

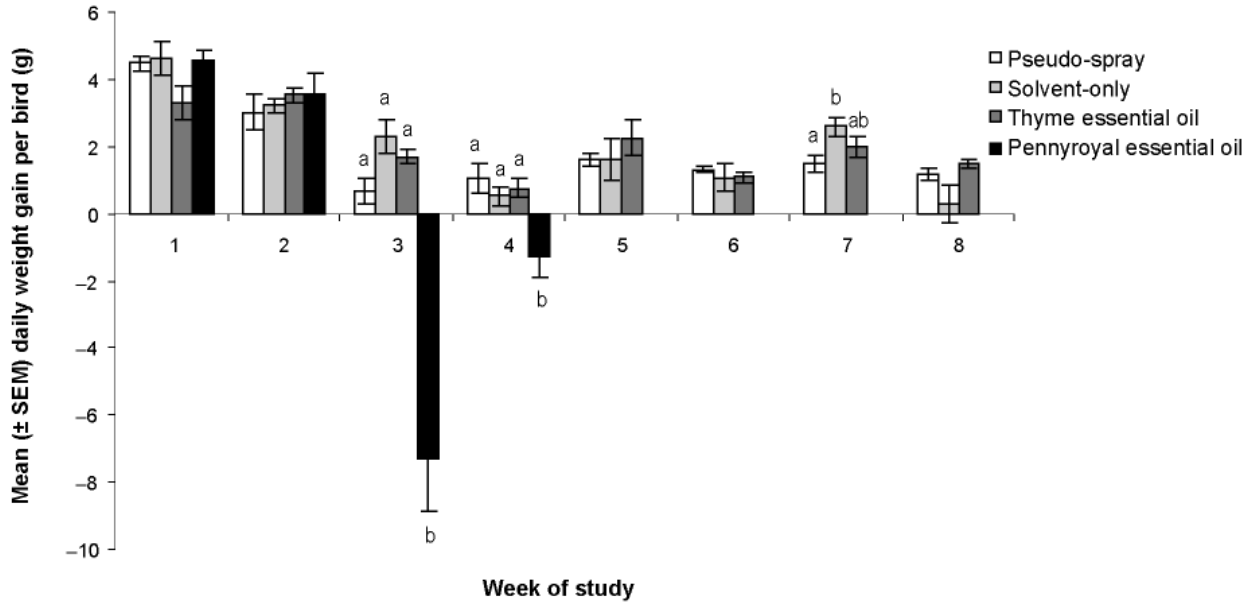
There was a significant difference between treatments in the average weight gained by hens for week 3 ($F_{3,9} = 26.49$, $P < 0.001$), 4 ($F_{3,9} = 6.83$, $P < 0.05$) and 7 ($F_{2,6} = 5.42$, $P < 0.05$) data. For week 3 and 4 data, this resulted from weight loss in birds subjected to pennyroyal essential oil, where birds in all other treatment groups experienced weight gain (Figure 2). For week 7, hens under the solvent-only treatment gained significantly more weight that those of the pseudo-spray treatment.

Feed intake and feed efficiency

There were significant differences between treatments in feed intake for weeks 5, 6 and 7 ($F_{2,6} = 13.35$, 7.06 and 8.95, $P < 0.01$, 0.05 and 0.05, respectively) (Figure 3). In all cases, this difference was due to higher feed intake ($P < 0.05$) in the pseudo-spray treatment compared to either the other two treatments in week 5 or the solvent-only treatment in weeks 6 and 7 (Figure 3). There was also a significant effect of block on the data (weeks 5 and 7;

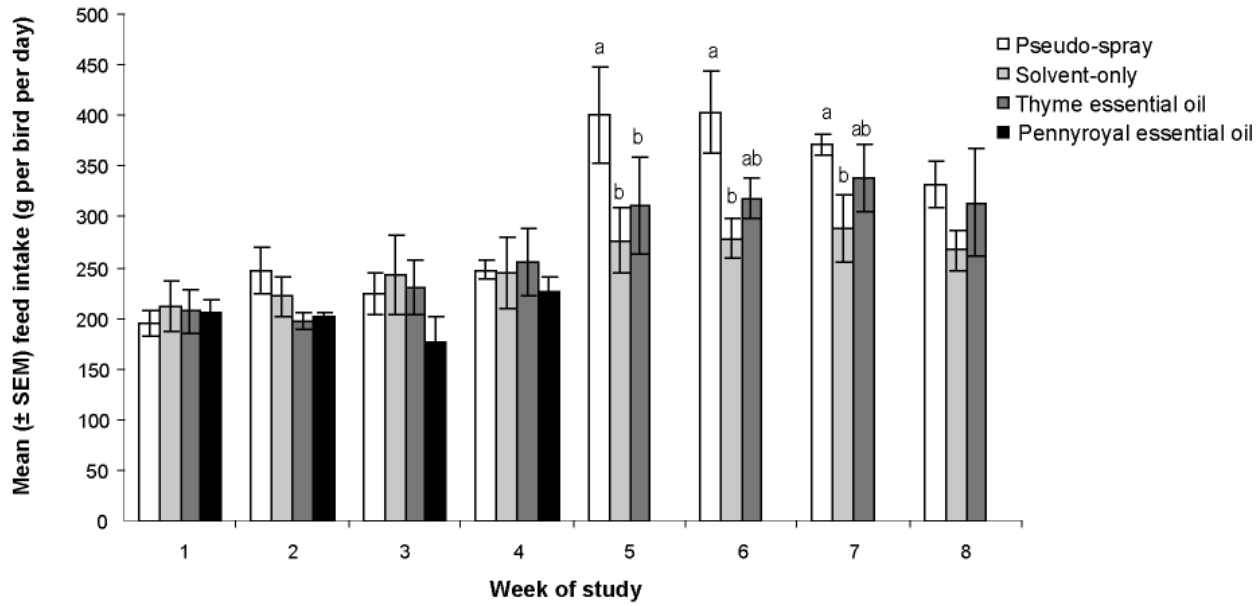
https://doi.org/10.1017/S0962728600001640 Published online by Cambridge University Press

Figure 2



Mean (± SEM) daily weight gain for hens housed in poultry huts subjected to different spray treatments (n = 4). Within a given week, means not sharing a common letter are significantly different according to the Tukey's test ($P < 0.05$).

Figure 3



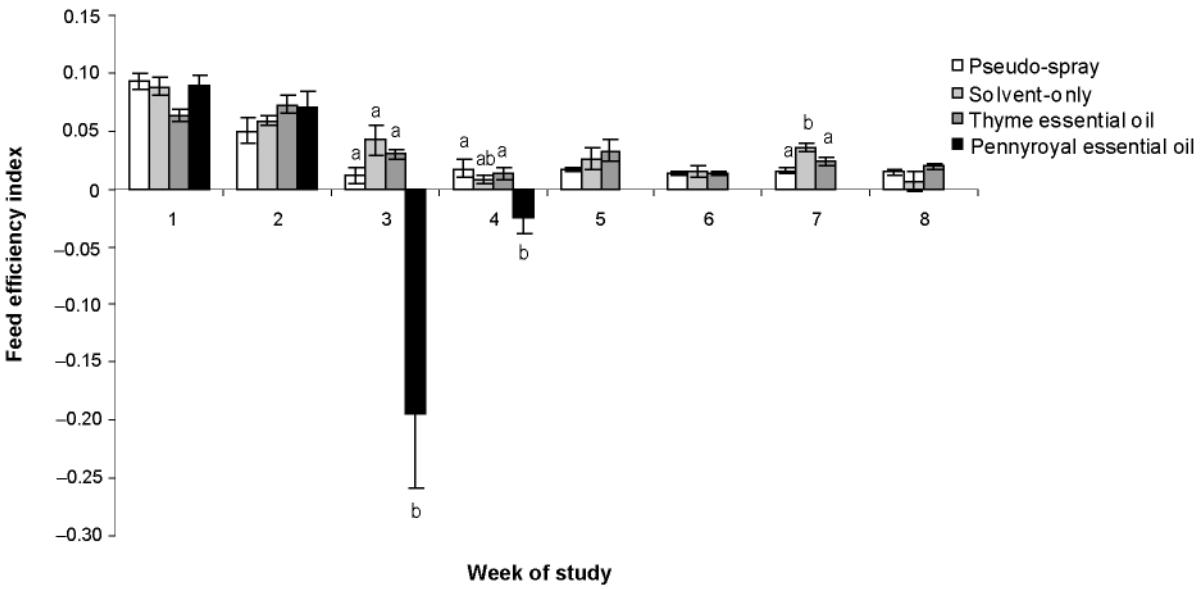
Mean (± SEM) daily feed intake for hens housed in poultry huts subjected to different spray treatments (n = 4). Within a given week, means not sharing a common letter are significantly different according to the Tukey's test ($P < 0.05$).

$F_{3,6}$ = 16.10 and 9.49, $P < 0.01$ and 0.05, respectively). Feed intake was increased in block 1 compared to all other blocks ($P < 0.05$) in week 5 of the study and increased ($P < 0.05$) in block 1 compared to blocks 2 and 3 in week 7.

There was a significant difference in feeding efficiency indices between treatments for weeks 3 ($F_{3,9}$ = 11.95, $P < 0.01$), 4 ($F_{3,9}$ = 5.89, $P < 0.05$) and 7 ($F_{2,6}$ = 15.56, $P < 0.01$) (Figure 4). For week 3 and 4, this resulted from a reduced digestive efficiency index in birds subjected to pennyroyal essential oil (Figure 4). For week 7, the feeding efficiency index was higher in the solvent-only treatment compared to the other two treatments ($P < 0.05$).

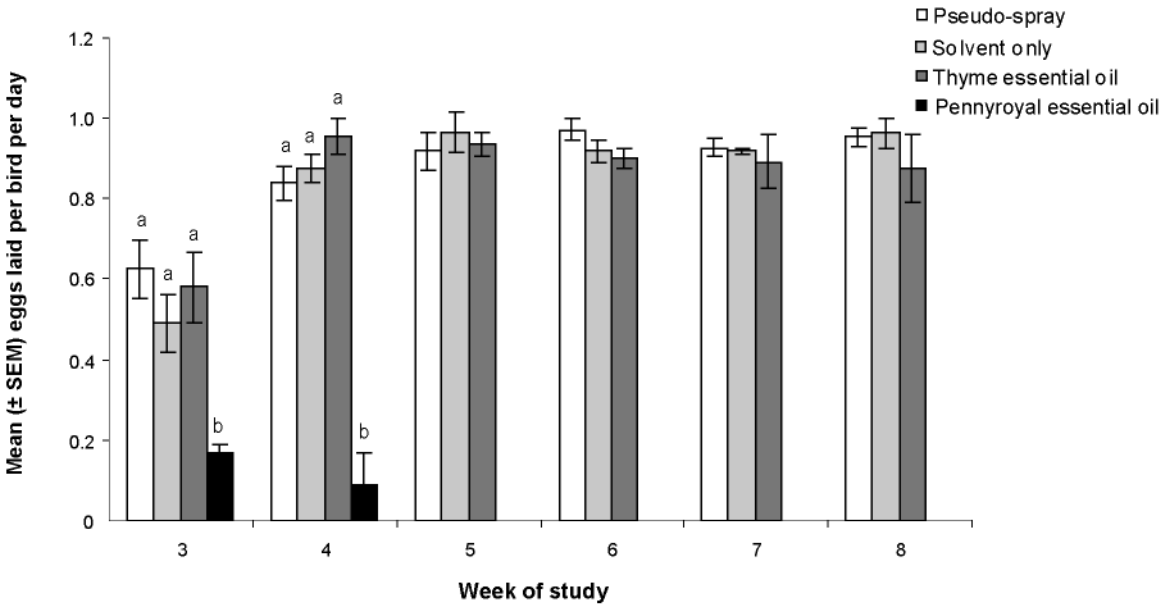
https://doi.org/10.1017/S0962728600001640 Published online by Cambridge University Press

Figure 4



Mean (± SEM) weekly feed efficiency indices for hens housed in poultry huts subjected to different spray treatments (n = 4). Within a given week, means not sharing a common letter are significantly different according to the Tukey's test ($P < 0.05$).

Figure 5



Mean (± SEM) daily eggs laid per bird for hens housed in poultry huts subjected to different spray treatments (n = 4). Within a given week, means not sharing a common letter are significantly different according to Mann-Whitney *U* tests ($P < 0.05$).

Egg production and weight

There was a significant difference in egg production between treatments for weeks 3 ($H = 9.79$, $P < 0.5$) and 4 ($H = 10.39$, $P < 0.05$). In both cases, this resulted from reduced egg production by birds subjected to pennyroyal essential oil (Figure 5). At no other point during the study was there any significant difference in the weekly egg production of hens under different treatments (Figure 5).

There was no significant difference between the mean weight of eggs laid from hens under different treatments at any point during weeks 3 to 8 of the study. In week 3 mean egg weight was approximately 45 g in all treatments, this rising to ~60 g by week 8.

https://doi.org/10.1017/S0962728600001640 Published online by Cambridge University Press

Discussion

The aim of this experiment was to evaluate the effect of thyme and pennyroyal essential oils on welfare and egg production of groups of laying hens. After commencing spray applications in week 2, it became apparent in weeks 3 and 4 of the study that pennyroyal essential oil was negatively affecting both the welfare (two birds died and others experienced weight loss not seen in other treatments) and productivity (egg production) of hens. Therefore, it was decided that the pennyroyal treatment should be discontinued as of week 4 of the study. Hens from huts treated with pennyroyal were sent for post mortem examination at a veterinary pathology laboratory. Gross post mortem examination did not reveal any lesions suggestive of toxicity, although such a pathological effect would only be determined by more detailed investigations, such as histopathology (JP Duff, personal communication 2008). Expert veterinary opinion was that the treatment should be terminated based on the clinical evidence presented; namely a substantial drop in bodyweight and egg production and depressed behaviour.

Previous work in our laboratory (George *et al* 2009, 2010b) has demonstrated that pennyroyal essential oil has several characteristics which indicate that it could be an acaricide for *D. gallinae*, including high levels of toxicity to *D. gallinae* at all life stages, environmental stability and relative non-toxicity to brine shrimp (George *et al* 2010a,b). However, pennyroyal essential oil is known to be relatively more toxic to vertebrates than thyme essential oil, where respective oral rat LD_{50} values have been reported as 400 and 2,840 mg kg^{-1} bodyweight (Golob *et al* 1999). Pulegone, the main constituent of pennyroyal essential oil, has been shown to cause slackness, depression, decreased food consumption and bodyweight and increased liver weight when administered orally to rats (Mølck *et al* 1998). Mølck *et al* (1998) also described fatal cases of pennyroyal and pulegone poisoning in humans resulting in mental disturbance, cerebral oedema, acute ischaemic necrosis, vacuolisation of the white matter, hepatic necrosis and renal failure. Whilst the previous work in our laboratory using a brine shrimp assay (a common method to assess potential mammalian toxicity) did not identify any potential deleterious effects of pennyroyal, in the present study pennyroyal was applied to huts at a substantially higher rate than that used in the brine shrimp assay and which could be recommended for commercial control of *D. gallinae*. This was to ensure that the effect of exposure to very high levels of oil, even by accident, on hen welfare could be determined. More detailed laboratory analyses, outwith the scope of this current study would be required to identify exactly why exposure to pennyroyal essential oil exerted a negative effect on hens in the current study. Nevertheless, the current study demonstrates that pennyroyal essential oil could not be recommended as a *D. gallinae* acaricide for use in the poultry industry and emphasises the need to carefully screen any new plant-derived products for potential toxic effects on hens. It is likely that the cumulative effect of increased dosage and higher vertebrate toxicity *per se* caused the observed negative effects in hens following pennyroyal essential oil application, where such effects were absent altogether where thyme essential oil was used.

At no point during the 8-week study period was there any difference in feather condition, bodyweight or weight gain between hens treated with thyme essential oil and those that were either pseudo-sprayed or treated with solvent alone. This shows that not only did treating with thyme essential oil have no immediate effect on welfare (after product spraying at the start of week 3), but also that there was no cumulative effect of treating at weekly intervals (for up to six consecutive weeks).

There was also no effect of thyme essential oil on egg production and egg weight, reassuring egg producers that the use of this product to control *D. gallinae* and safeguard hen welfare is unlikely to have any negative impact on production parameters. Such a result, whilst beneficial to the development of thyme essential oil as a *D. gallinae* acaricide, is not surprising in light of work elsewhere in the literature. In work by Radwan *et al* (2008), inclusion of herbs in hen diets (including thyme at 1.0%) improved production performance, with observed increases in egg number and mass and improved feed conversion. Radwan *et al* (2008) attributed this, along with similar results from earlier work with chicks using thyme and rosemary at 0.5% (Radwan 2003), to the essential oil component of the herbs having antimicrobial, antifungal and antioxidant properties that may have improved utilisation of dietary nutrients. Similarly, Poráčová *et al* (2007) reported an increase in the weight of eggs laid by hens whose diet was enriched with 0.1% chamomile essential oil. There was, however, no evidence in the current work that treating hen housing as opposed to feed with thyme essential oil had any beneficial effect on growth or production parameters.

Some significant effects of treatment were seen for data on feed intake and feed efficiency between thyme and the pseudo-spray/solvent-only treatments. There was a trend for feed intake in weeks 5, 6 and 7 to be greater for birds that were pseudo-sprayed. Even then, however, only in week 5 was feed intake from thyme-treated and pseudo-spray huts significantly different from one another (whilst data from pseudo-spray and solvent-only huts were significantly different from each other on all three dates). This suggests that any effect seen was not the result of spraying with thyme essential oil, but perhaps resulted from spraying *per se* for reasons unknown. As the huts in three of the four treatments were sprayed with liquid to the point of run-off, it is possible that there was an increase in humidity/moisture in these huts when compared to pseudo-spray treatment which might have affected feed intake. The results were nevertheless inconsistent and further study would be needed to determine if this was indeed the case. Feed intake data, if data from pennyroyal-treated hens are discounted, did not differ between treatments for any other weeks of the study (weeks 1 and 2 prior to spraying with any product and weeks 3, 4 and 8 after product applica-

https://doi.org/10.1017/S0962728600001640 Published online by Cambridge University Press

tion). Similarly, again with pennyroyal data aside, data on feed efficiency only differed in week 7 of the study period. Again, this was not the result of data from thyme-treated huts differing from both pseudo-spray and solvent-only treated huts, suggesting that treating with thyme essential oil *per se* was again not the cause of this difference. Where feed efficacy was reduced in pennyroyal-treated huts it can be speculated that this resulted from birds diverting nutrients from production (egg output) to homeostasis (response to the toxicity challenge from pennyroyal) since feed intake was not significantly reduced.

It therefore seems that thyme essential oil, if developed and deployed as an acaricide for use against *D. gallinae*, could be applied at an effective concentration and reapplied on multiple occasions if necessary with no negative impact on hen welfare or egg production. Other work on essential oils has suggested that lavender essential oils may be short-lived in their toxicity to *D. gallinae* (George *et al* 2008b) and may thus require such repeat application to be effective as acaricides. Research also indicates, however, that thyme essential oil might display relatively fast knock-down to *D. gallinae* (Olatunji *et al* 2008) and be repellent to adult *D. gallinae* for relatively long periods (of up to several weeks) (George *et al* 2009). If used as an acaricide application, thyme essential oil could therefore both kill any *D. gallinae* present in a poultry unit and serve to minimise re-colonisation by *D. gallinae* following application to further safeguard hen welfare.

The potential of thyme essential oil as a pest control agent is also supported elsewhere. Essential oil from thyme (or thymol, its main chemical constituent) has been researched for pest control against *D. gallinae* (Kim *et al* 2004), the bee mite species, *Varroa jacobsoni* (Oudemans) (Calderone *et al* 1997), *Acarapis woodi* (Rennie) (Rice *et al* 2002) and *Varroa destructor* (Anderson & Trueman) (Rice *et al* 2002; Floris *et al* 2004) and the parasitic mite *Psoroptes cuniculi* (Delafond) (Perrucci *et al* 1995). In displaying toxicity to a range of pest species in this way, it may be the case that thyme essential oil could be deployed in poultry units against a multitude of pests in a single application. Mites (including species other than *D. gallinae*), lice, bedbugs, fleas, ticks and various species of Diptera may all serve as pests to varying degrees in poultry systems (Axtell 1999) and could potentially be targeted alongside *D. gallinae*.

Animal welfare implications

The results of the current study confirm that thyme essential oil, a plant-based product with promising potential for development as either a *D. gallinae* acaricide, repellent or both, could be used in commercial laying hen systems with no apparent risk to hen welfare or productivity. This will hopefully allow for further field-scale testing and development of thyme essential oil-based products for use against *D. gallinae* in laying hen systems.

Conclusion

In the absence of any negative effect on hen welfare and egg production, data from this pilot study provides evidence that thyme essential oil may be considered a promising candidate as an acaricide and/or repellent for *D. gallinae*. More work will nevertheless be required to both confirm these results on a commercial scale and identify the best options for deploying essential oils in commercial practice.

Acknowledgements

This work was carried out as part of the MITEeHEN project to develop alternative, plant-based acaricides to control *D. gallinae* for which funding from Defra is gratefully acknowledged. The authors are also grateful to representatives of the UK poultry industry, including BFREPA, NFU and Noble Foods for providing technical advice on the development of alternative control strategies for *D. gallinae*.

References

Arkle S, Guy JH and Sparagano O 2006 Immunological effects and productivity variation of red mite (*Dermanyssus gallinae*) on laying hens — implications for egg production and quality. *World's Poultry Science Journal 62*: 249-257

Axtell RC 1999 Poultry integrated pest management; status and future. *Integrated Pest Management Reviews 4*: 53-73

Beugnet F, Chauve C, Gauthey M and Beert L 1997 Resistance of the red poultry mite to pyrethroids in France. *Veterinary Record 140*: 577-579

Bilčík B and Keeling LJ 1999 Changes in feather condition in relation to feather pecking and aggressive behaviour in laying hens. *British Poultry Science 40*: 444-451

Bruneau A, Dernburg A, Chauve C and Zenner L 2001 First *in-vitro* cycle of the chicken mite, *Dermanyssus gallinae* (De Geer 1778), utilizing an artificial feeding device. *Parasitology 123*: 583-589

Cafiero M, Camarda A, Circella E, Santagada G, Schino G and Lomuto M 2008 Pseudoscabies caused by *Dermanyssus gallinae* in Italian city dwellers: a new setting for an old dermatitis. *Journal of the European Academy of Dermatology and Venereology 22*: 1382-1383

Calderone NW, Wilson WT and Spivak M 1997 Plant extracts used for control of the parasitic mites *Varroa jacobsoni* (Acari: Varroidae) and *Acarapis woodi* (Acari: Tarsonemidae) in colonies of *Apis mellifera* (Hymenoptera: Apidae). *Journal of Economic Entomology 90*: 1080-1086

Chauve C 1998 The poultry red mite *Dermanyssus gallinae* (De Geer, 1778): current situation and future prospects for control. *Veterinary Parasitology 79*: 239-245

Chirico J, Eriksson H, Fossum O and Jansson D 2003 The poultry red mite, *Dermanyssus gallinae*, a potential vector of *Erysipelothrix rhusiopathiae* causing erysipelas in hens. *Medical and Veterinary Entomology 17*: 232-234

Choi W, Lee S, Park H and Ahn Y 2004 Toxicity of plant essential oils to *Tetranychus urticae* (Acari: Tetranychidae) and *Phytoseiulus persimilis* (Acari: Phytoseiidae). *Journal of Economic Entomology 97*: 553-558

Cosoroaba I 2001 Massive *Dermanyssus gallinae* invasion in battery-husbandry raised fowls. *Revue de Médecine Vétérinaire 152*: 89-96

Dalton JP and Mulcahy G 2001 Parasite vaccines-a reality? *Veterinary Parasitology 98*: 149-167

Fiddes MD, Le Gresley S, Parsons DG, Epe C, Coles GC and Stafford KA 2005 Prevalence of the poultry red mite (*Dermanyssus gallinae*) in England. *Veterinary Record 157*: 233-235

https://doi.org/10.1017/S0962728600001640 Published online by Cambridge University Press

Floris I, Satta A, Cabras P, Garau V and Angioni A 2004 Comparison between two thymol formulations in the control of *Varroa destructor*: effectiveness, persistence, and residues. *Journal of Economic Entomology 97*: 187-191

George DR, Callaghan K, Guy JH and Sparagano OAE 2008b Investigating the residual toxicity of lavender essential oils as acaricides against the poultry red mite (*Dermanyssus gallinae*). *Research in Veterinary Science 85*: 440-442

George DR, Guy JH, Arkle S, Harrington D, De Luna C, Okello EJ, Shiel RS, Port G and Sparagano OAE 2008a Use of plant-derived products to control arthropods of veterinary importance; a review. *Annals of the New York Academy of Science 1149*: 23-26

George DR, Sparagano OAE, Port G, Okello E, Shiel RS and Guy JH 2009 Repellence of plant essential oils to *Dermanyssus gallinae* and toxicity to the non-target invertebrate *Tenebrio molitor*. *Veterinary Parasitology 162*: 129-134

George DR, Sparagano OAE, Port G, Okello E, Shiel RS and Guy JH 2010a Environmental interactions with the toxicity of plant essential oils to the poultry red mite *Dermanyssus gallinae*. *Medical and Veterinary Entomology 24*: 1-8

George DR, Sparagano OAE, Port G, Okello E, Shiel RS and Guy JH 2010b Toxicity of plant essential oils to different life stages of poultry red mite (*Dermanyssus gallinae*) and non-target invertebrates. *Medical and Veterinary Entomology 24*: 9-15

Golob P, Moss C, Dales M, Fidgen A, Evans J and Gudrups I 1999 The use of spices and medicinals as bioactive protectants for grains. *FAO Agricultural Services Bulletin No 37, Food and Agriculture Organisation of the United Nations*. Rome, Italy

Guy JH, Khajavi M, Hlalele MM and Sparagano O 2004 Red mite (*Dermanyssus gallinae*) prevalence in laying units in Northern England. *British Poultry Science 45*: S15

Höglund J, Nordenfors H and Uggla A 1995 Prevalence of the poultry red mite, *Dermanyssus gallinae*, in different types of production systems for egg layers in Sweden. *Poultry Science 74*: 1793-1798

Isman MB 2006 Botanical insecticides, deterrents, and repellents in modern agriculture and an increasingly regulated world. *Annual Review of Entomology 51*: 45-66

Kilpinen O 1999 Problems caused by the chicken mite, *Dermanyssus gallinae*, in the Danish egg production. In: Good M, Hall MS, Losson B, Pithan K and Sol J (eds) *Mange and Myiasis of Livestock* pp 61-65. Commission of the European Communities: Brussels, Belgium

Kilpinen O 2001 Activation of the poultry red mite, *Dermanyssus gallinae* (Acari: Dermanyssidae), by increasing temperatures. *Experimental and Applied Acarology 25*: 859-867

Kim S, Na Y, Yi J, Kim B and Ahn Y 2007 Contact and fumigant toxicity of oriental medicinal plant extracts against *Dermanyssus gallinae* (Acari: Dermanyssidae). *Veterinary Parasitology 145*: 377-382

Kim S, Yi J, Tak J and Ahn Y 2004 Acaricidal activity of plant essential oils against *Dermanyssus gallinae* (Acari: Dermanyssidae). *Veterinary Parasitology 120*: 297-304

Lucky AW, Sayers C, Argus JD and Lucky A 2001 Avian mite bites acquired from a new source: pet gerbils: report of 2 cases and review of the literature. *Archives of Dermatology 137*: 167-170

Lundh J, Wiktelius D and Chirico J 2005 Azadirachtin-impregnated traps for the control of *Dermanyssus gallinae*. *Veterinary Parasitology 130*: 337-342

Maurer V, Perler E and Heckendorn F 2009 In vitro efficacies of oils, silicas and plant preparations against the poultry red mite *Dermanyssus gallinae*. *Experimental and Applied Acarology 48*: 31-41

Mignon B and Losson B 2008 Dermatitis in a horse associated with the poultry mite (*Dermanyssus gallinae*). *Veterinary Dermatology 19*: 38-43

Mølck A, Poulsen M, Tingard Lauridsen S and Olsen P 1998 Lack of histological cerebellar changes in Wistar rats given pulegone for 28 days. Comparison of immersion and perfusion fixation. *Toxicology Letters 95*: 117–122

Olatunji G, Guy JH, George DR and Sparagano OAE 2008 Effect of plant essential oils as acaricides against the poultry red mite, *Dermanyssus gallinae*, with special focus on exposure time. *British Society for Parasitology Spring Meeting* pp 166. 30th March-2nd April 2008, Newcastle upon Tyne, UK

Perrucci S, Macchioni G, Cioni PL, Flamini G and Morelli I 1995 Structure/activity relationship of some natural monoterpenes as acaricides against *Psoroptes cuniculi*. *Journal of Natural Products 58*: 1261-1264

Poráčová J, Blasčáková M, Zahatňanská M, Taylorová B, Sutiaková I and Sály J 2007 Effect of chamomile essential oil application on the weight of eggs in laying hens Hisex Braun. *Acta Horticulturae 749*: 203-206

Radwan NL 2003 *Effect of using some medicinal plants on performance and immunity of broiler chicks*. PhD Thesis, Cairo University, Egypt

Radwan NL, Hassan RA, Qota EM and Fayek HM 2008 Effect of natural antioxidant on oxidative stability of eggs and productive and reproductive performance of laying hens. *International Journal of Poultry Science 7*: 134-150

Rice ND, Whittington R and Higo HA 2002 Comparison of release mechanisms for botanical oils to control *Varroa destructor* (Acari: Varroidae) and *Acarapis woodi* (Acari: Tarsonemidae) in colonies of honey bees (Hymenoptera: Apidae). *Journal of Economic Entomology 95*: 221-226

Sparagano O, Pavlićević A, Murano T, Camarda A, Sahibi H, Kilpinen O, Mul M, van Emous R, le Bouquin S, Hoel K and Cafiero MA 2009 Prevalence and key figures for the poultry red mite *Dermanyssus gallinae* infections in poultry farm systems. *Experimental and Applied Acarology 48*: 3-10

Urquhart GM, Armour J, Duncan JL, Dunn AM and Jennings FW 1996 *Veterinary Parasitology, Second Edition*. Blackwell Scientific Publications: Oxford, UK

Valiente Moro C, De Luna CJ, Tod A, Guy JH, Sparagano OAE and Zenner L 2009 The poultry red mite (*Dermanyssus gallinae*): a potential vector of pathogenic agents. *Experimental and Applied Acarology 48*: 93-104

van Emous R 2005 Wage war against the red mite! *Poultry International 44*: 26-33

Wojcik AR, Greygon-Franckiewicz B, Zbikowska E and Wasielewski L 2000 Invasion of *Dermanyssus gallinae* (De Geer, 1778) in poultry farms in the Torun Region. *Polish Parasitological Society 46*: 511-515

https://doi.org/10.1017/S0962728600001640 Published online by Cambridge University Press