

# Control of *Dermanyssus gallinae* (De Geer 1778) and other mites with volatile organic compounds, a review

## Review

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### Abstract

*Dermanyssus gallinae* (De Geer 1778), commonly named the poultry red mite (PRM), is considered to be the most harmful ectoparasite in poultry farms in Europe. This species feeds on the blood of laying hens, but spends most of its time hidden in cracks and crevices around hen nests. To control PRM populations in poultry houses, chemical pesticides are currently used; however, concern is growing regarding the harmful residues found in eggs and hens, along with the increased resistance of mites against several compounds. Alternatives to synthetic compounds are now being explored, including vaccines, biological control, physical control and semiochemical control based on the chemical ecology of PRM. This review focused on the different volatile organic compounds (VOCs) identified from *D. gallinae* and other mite species that have been discovered to control them. Pheromones (aggregation pheromone, sex pheromone and alarm pheromone) and kairomones promoting attraction behaviour in *D. gallinae* and other mite species are presented, while VOCs from essential oils and plant extracts with repellent properties are also explored. Finally, devices using VOCs on PRM in the field are described, with devices that have been tested on other Acari species being mentioned as potential directions for the future control of PRM.

### Introduction

The poultry red mite (PRM) *Dermanyssus gallinae* (De Geer 1778) is considered to be the most harmful ectoparasite in poultry farms in Europe (Höglund *et al.*, 1995; Sparagano *et al.*, 2014). Individuals mainly feed on the blood of laying hens and wild birds at night (Kirkwood, 1968), and remain hidden the rest of its time in dark places (*e.g.* crevices and cracks around hen nests), where they can survive for up to 9 months without feeding (Chauve, 1998; Nordenfors *et al.*, 1999). PRM can also accidentally infest mammals, including horses, dogs and even humans (Ramsay *et al.*, 1975; Mignon and Losson, 2008; George *et al.*, 2015). The occurrence of *D. gallinae* negatively impacts the general health of hens and the quality of their eggs (*i.e.* blood marks on the shells of eggs), which leads to significant economic losses in the hen egg laying industry (Chauve, 1998; Kilpinen *et al.*, 2005). In particular, PRM parasitism causes skin irritation, anaemia, vascular problems and, even, the death of the host (Chauve, 1998; Cosoroaba, 2001; Kilpinen *et al.*, 2005).

At present, PRM in egg laying hen units is mainly controlled by using different chemical acaricides. However, the repeated use of chemical acaricides is responsible for the presence of residues in eggs and meat (with a potential impact on Public Health), and is associated with the emergence of PRM resistance to different acaricides, as reported in several European countries (Zeman and Zelezny, 1985; Beugnet *et al.*, 1997; Marangi *et al.*, 2009; Sparagano *et al.*, 2009, 2014). This issue is exacerbated when chemical acaricides are used illegally (off-label), as recently demonstrated in Europe (Flochlay *et al.*, 2017).

In addition, resistance to certain acaricides (such as carbaryl, amitraz and permethrin) has emerged in PRM populations over the last 40 years (Zeman and Zelezny, 1985; Beugnet *et al.*, 1997; Marangi *et al.*, 2009; Sparagano *et al.*, 2009, 2014). Therefore, increasing awareness of the general public and changes to legislation have promoted the search for new and eco-friendly ways to control *D. gallinae*, including vaccines, entomopathogenic fungi and essential oils. Simultaneously, the chemical ecology of PRM is being increasingly studied to develop control strategies based on pheromonal communication.

Here, an overview of current information is provided, along with potential future directions, in controlling PRM. The different volatile organic compounds (VOCs) currently identified from *D. gallinae* and other mite species are presented, along with those already used to control them. Specifically, information is presented on (1) pheromones affecting mite behaviour with an attracting effect and (2) the repellent properties of VOCs on PRM. Finally, several devices using VOCs to control *D. gallinae* in field assays are described, as well as designs tested on other Acari species that could potentially be used on PRM.

## Attractive substances

VOCs present an interesting alternative to classical acaricides for controlling acarid populations. Several types of VOCs could be used to manipulate the behaviour of Acari, potentially being integrated in « attract and kill » devices. Here, the potential of several pheromones (Table 1) and kairomones (Table 2) that have already been identified is focused on.

## Pheromones

Pheromones are molecules that are emitted by an individual of a species to influence the behaviour of another individual of the same species (Pageat, 2005). Several pheromones are considered to attract mites, including aggregation pheromones, immature female pheromones, female sex pheromones and, at low doses, alarm pheromones (Sonenshine, 1985; Carr and Roe, 2016). To identify the molecules present in the pheromonal mix released by a species, static headspace analyses (*e.g.* solid phase micro extraction or SPME) (Birkett *et al.*, 2010; Steidle *et al.*, 2014) and dynamic headspace analyses (*e.g.* thermodesorption) (Chen *et al.*, 2019; Makarow *et al.*, 2019) are performed. While dynamic headspace analysis has not yet been performed on mites, to our knowledge, it could prove useful, because it allows small amounts of volatile molecules to be detected and quantified (Pfannkoch and Whitecavage, 2000; Zhang and Li, 2010). The sampling technique must be coupled to gas chromatography-mass spectrometry (GC-MS) analysis to identify the different compounds (Zhang and Li, 2010). Following the identification and quantification of molecules within the pheromonal mix, behavioural tests could be performed, including Y-tube olfactometers bioassays or filter paper contact bioassays, to characterize the behaviour of mites when they are confronted with all or a part of the identified molecules (Entrekin and Oliver, 1982; Steidle *et al.*, 2014).

## Aggregation pheromones

The aggregation pheromones of mites cause non-feeding conspecific mites (male and female) of different developmental stages to aggregate in a safe environment (Carr and Roe, 2016). The aggregation behaviour in PRM was well studied before the aggregation pheromone was identified. Entrekin and Oliver (1982) studied two possible causes for this behaviour: thigmokinesis (*i.e.* increased locomotion in response to changes in contact with the immediate physical environment) and the release of an aggregation pheromone. Both stimuli impact the clustering of mites; however, the chemical ones form aggregates more efficiently. The aggregation phenomenon due to the perception of potential aggregation pheromones by PRM was evaluated by Koenraadt and Dicke (2010). The authors tested the attraction of two groups of conspecific mites (fed and unfed) by performing behavioural experiments. The authors showed that fed and unfed mites were attracted by volatiles emitted by fed conspecifics. This finding was consistent with a previous study showing that the aggregation of mites increases after feeding (Entrekin and Oliver, 1982). Koenraadt and Dicke (2010) proposed that unfed mites are motivated to find a host to feed, while fed ones were searching for a place to hide and reproduce after their blood meal.

Subsequently, Birkett *et al.* (2010) conducted SPME-GC analysis on fed adult PRM, and identified eight molecules, including seven carboxylic acids (including hexanoic acid and dodecanoic acid) and one aldehyde (an octanal) that could be components of the pheromonal mix driving aggregation-forming behaviour in PRM. Behavioural experiments confirmed the attractive effect of each compound, as well as a synergic effect when compounds are associated. It was concluded that the eight compounds attract PRM and could be applied simultaneously, separately or

sequentially to each other (Birkett *et al.*, 2010). It is possible that the aggregation pheromone of PRM is composed of only one or several molecules, as observed in other mite species. For example, Lardolure is the only aggregation pheromone that has been identified in *Caloglyphus polyphyllae*<sup>1</sup> and *Lardoglyphus konoii* mites (Kuwahara *et al.*, 1994; Carr and Roe, 2016). In comparison, the aggregation pheromone of the European House Dust Mite (EHD) *Dermatophagoides pteronyssinus* is mostly composed of neryl propionate, pentadecane and neryl formate (Skelton *et al.*, 2010; Steidle *et al.*, 2014). Finally, neryl formate might be a component of the pheromonal mix of several different species, as it was identified as a major compound of the aggregation pheromone in the American House Dust Mite, *Dermatophagoides farinae* and EHD (Skelton *et al.*, 2010).

## Alarm pheromones

Alarm pheromones are emitted by an individual that has been stressed by an unsafe environment or that has been injured through direct contact (Carr and Roe, 2016). Detection of low concentrations of alarm pheromones causes attraction and clustering behaviour in mites, as part of group survival strategies (*i.e.* formation of a 'safety net'). Above a given concentration, the pheromone becomes repulsive, which causes the strategies of individuals changing (*i.e.* scattering and hiding) (Carr and Roe, 2016). To date, the alarm behaviour or the possible emission of this pheromone has not been studied in PRM; however, several compounds of this pheromone have been identified in other species of Astigmatid mites, including neryl formate and neral (Kuwahara, 2010, 2011; Carr and Roe, 2016). When neral is applied in low doses (1 and 3 ng) on filter paper, mites (*Schwiebia elongata*) are attracted to this compound; however, at higher doses (30 ng), they exhibited repulsive behaviour (Nishimura *et al.*, 2003; Carr and Roe, 2016). Neryl formate was identified as an alarm pheromone for several species, including *Tyrophagus putrescentiae* (Schrank 1781), leading to alarm behaviour (Kuwahara, 2010); however, it attracts also other species, such as *D. farinae* and *D. pteronyssinus* at high doses (10 and 100 ng), and is considered to be an aggregation pheromone (Skelton *et al.*, 2010). Therefore, it is essential to clearly differentiate alarm pheromones that attract mites at low doses from aggregation pheromone that attract mites at all doses. Studies on the PRM alarm behaviour or the potential alarm pheromone emission have yet to be conducted, which could represent an interesting future direction for research.

## Sex pheromones

Two types of sex pheromones have been detected in mites: (1) immature female pheromones, also known as arrestant sex pheromones and (2) female sex pheromones (Sonenshine, 1985; Carr and Roe, 2016). There is a competition between male mites to reproduce with conspecific females. Consequently, males might detect differences between a deutonymph (*i.e.* second nymphal stage) from a protonymph (*i.e.* first nymphal stage) by using the immature pheromone signature emitted by deutonymph females, which is composed of citronellol, farnesol and nerolidol in *Tetranychus urticae* females (Regev and Cone, 1975, 1976, 1980). Following the discovery of a deutonymph female, males exhibit guarding behaviour (*i.e.* association of a male with a deutonymph female) in some acarid species [*e.g.* *Hericia* sp. (Canestrini 1888) or *Tetranychus kanzawai* (Kishida 1927)], until the deutonymph becomes an adult female (Sonenshine, 1985; Witaliński *et al.*, 1992; Yasui, 1992; Fashing, 2003; Oku, 2009; Carr and Roe, 2016). Colloff (2009), cited by Carr and

<sup>1</sup>All complete species names could be found in Table 1.

**Table 1.** Chemical compounds of pheromones tested for their attractive and repulsive effect on mites

Species	Tested substances	Experiments	Effect	References
<i>Acarus immobilis</i> (Griffith 1964)	2-Hydroxy-6-methylbenzaldehyde	Filter paper bioassays Petri dishes experiments	Attraction	Sato <i>et al.</i> (1993)
<i>Aleuroglyphus ovatus</i> (Troupeau 1879)	2-Hydroxy-6-methylbenzaldehyde	Filter paper bioassays Petri dishes experiments	Attraction	Kuwahara <i>et al.</i> (1991)
<i>Caloglyphus</i> sp.	(2R,3R)-Epoxyneral Rosefuran	Petri dishes experiments	Attraction	Mori <i>et al.</i> (1996); Mori <i>et al.</i> (1998)
<i>Caloglyphus polyphyllae</i> (Zakhvatkin 1941)	$\beta$ -Acaridial	Petri dishes experiments	Attraction	Shimizu <i>et al.</i> (2001)
<i>Carpoglyphus lactis</i> (Linnaeus 1767)	Lardolure	Not described	Attraction	Kuwahara <i>et al.</i> (1994)
<i>Cosmoglyphus hughesis</i> (Samsinak 1966)	2-Hydroxy-6-methylbenzaldehyde	Filter paper bioassays Petri dishes experiments	Attraction	Ryono <i>et al.</i> (2001)
<i>Dermanyssus gallinae</i> (De Geer 1778)	Aggregation pheromone Butyric acid Decanoic acid Heptanoic acid Hexanoic acid Nonanoic acid Octanoic acid Pentanoic acid	Petri dishes experiments Y-tube olfactometers 4 arms-olfactometers	Attraction	Entrekin and Oliver (1982); Birkett <i>et al.</i> (2010); Koenaardt and Dicke (2010)
<i>Dermatophagoides farinae</i> (Hughes 1961)	2-Hydroxy-6-methylbenzaldehyde Neryl formate	Filter paper bioassays Petri dishes experiments Y-tube olfactometers	Attraction	Tatami <i>et al.</i> (2001); Skelton <i>et al.</i> (2010)
<i>Dermatophagoides pteronyssinus</i> (Trouessart 1897)	(6Z,9Z)-6,9-heptadecadiene Neral Neryl formate Neryl propionate Pentadecane (Z)-8-heptadecene	Long-range olfactometers Y-tube olfactometers	No effect   Attraction	Skelton <i>et al.</i> (2010); Steidle <i>et al.</i> (2014)
<i>Lardoglyphus konoi</i> (Sasa and Asanuma 1951)	Lardolure	Not described	Attraction	Kuwahara <i>et al.</i> (1994)
<i>Rhizoglyphus robini</i> (Claparède 1869)	$\alpha$ -Acaridial	Petri dishes experiments	Attraction	Mizoguchi <i>et al.</i> (2003)
<i>Rhizoglyphus setosus</i> (Manson 1972)	S-Isorobinal	Petri dishes experiments	Attraction	Mizoguchi <i>et al.</i> (2005)
<i>Schwiebia elongata</i> (Banks 1906)	Neral (low dose) Neral (high dose)	Pasteur pipet experiments	Attraction Repulsion	Nishimura <i>et al.</i> (2003)
<i>Tetranychus urticae</i> (Koch 1836)	Citronellol Farnesol Nerolidol	Petri dishes experiments	Attraction	Regev and Cone (1975), (1976), (1980)

Roe (2016), highlighted that only deutonymphs are able to emit the immature female pheromone. Nevertheless, this pheromone has not been identified in *D. gallinae* so far.

The female sex pheromone is normally released by fed mature females to attract preferably fed males, as the two sexes have a high reproductive success status (Sonenshine, 1985; Carr and Roe, 2016). The female sex pheromone might also be released by males, explaining the mounting behaviour between males

that is sometimes observed (Mori and Kuwahara, 2000; Mizoguchi *et al.*, 2005; Carr and Roe, 2016). Again, the female sex pheromone was not studied in *D. gallinae*, but different compounds have been identified as female sex pheromones in mites (Table 1).

PRM is part of the Acari. The molecules identified in closely related species could be potential candidates for characterizing the sex pheromones of PRM, especially as some compounds are

**Table 2.** Chemical compounds of host kairomones with an attractive effect on mites

Species	Tested substances	Experiments	Effect	References
<i>Dermanyssus gallinae</i> (De Geer 1778)	1-Heptadecene	Feeding experiments Field experiments Y-tube olfactometers	Attraction	Pageat (2005); Harrington <i>et al.</i> (2010); Koenraadt and Dicke (2010); Roy <i>et al.</i> (2018)
	9-Octadecene-ol 1 (oleyl alcohol)			
	Carbone dioxide			
	Chicken skin extracts			
	(E)-non-2-enal			
	Ethyl acetate			
	Feathers (Aged and Fresh)			
	Heptadecane			
	Nonanal			
	Nonanoic acid			
	Oct-1-en-3-ol			
	Octadecane			
Octanal				
<i>Macrocheles muscaedomesticae</i> (Scopoli 1972)	N-Phenyl-N-glucoside	No details available	Attraction	Carr and Roe (2016)
	N-Phenyl-N-mannoside			
<i>Phytoseiulus macropilis</i> (Banks 1904)	VOC from <i>Tetranychus urticae</i>	Y-Tube olfactometers	Attraction	Amin <i>et al.</i> (2010)
<i>Varroa destructor</i> (Anderson and Trueman 2000)	Ethyl palmitate	Mite wind tunnel experiments	Attraction	Calderone and Lin (2001)
	Methyl linolenate			
	Methyl palmitate			
<i>Varroa jacobsoni</i> (Oudemans 1904)	Ethyl palmitate	4 arms-olfactometers	Attraction	Le Conte <i>et al.</i> (1989)
	Methyl linolenate			
	Methyl palmitate			

already shared by different species, including 2-hydroxy-6-methylbenzaldehyde in *Acarus immobilis*, *Aleuroglyphus ovatus*, *Cosmoglyphus hughesi* and *D. farinae* (Kuwahara *et al.*, 1991; Sato *et al.*, 1993; Ryono *et al.*, 2001; Tatami *et al.*, 2001).

Sexual chemical communication between mites has been extensively investigated in several species, but not in PRM. Thus, it would be interesting to fill this knowledge gap on chemical communication in PRM. The potential synergic effects between the sex pheromone and aggregation pheromone could potentially be identified, which could be used to control PRM.

### Host kairomones

Kairomones are molecules emitted by individuals mediating interspecific interactions that are generally harmful to the emitters (Pageat, 2005). This section addresses kairomones that are attractive to some mites, including *D. gallinae* (Table 2). PRM aim to localize potential hosts to feed. The uropygial gland products of chicken might have potential kairomones in the host location process (Pageat, 2005; Koenraadt and Dicke, 2010). The identification of some of the compounds released by these products revealed the presence of alcohols, alkanes and alkenes (described in Table 2). However, these compounds were directly identified from the chicken gland, and it was not confirmed that these compounds are volatiles that could be detected over a distance, as in olfactometer experiments (Roy *et al.*, 2018). Following behavioural experiments in olfactometers, Koenraadt and Dicke (2010) observed that aged feathers of chicken (found on the floor of poultry houses and with potential microbial activity) were more attractive

to PRM than fresh feathers (feathers cut from live hens). Mites responded to small numbers of feathers (10 feathers in a 2.5 L glass jar), indicating that they can detect very low amounts of these kairomones; however, no precise measurement was made (Koenraadt and Dicke, 2010). Carbon dioxide (CO<sub>2</sub>) can also be considered as a kairomone for PRM. A gradual increase to CO<sub>2</sub> levels induces host-seeking behaviour in mites, whereas a quick increase induces mites to freeze. This attitude is probably linked to the potential presence of predators (Kilpinen, 2005). Finally, temperature is a powerful stimulus that activates mites and, coupled with vibrations, might contribute to host detection and localization (Kilpinen, 2001, 2005).

Following host localization, PRM are attracted by several kairomones produced by the host (Roy *et al.*, 2018). Several compounds were identified from living hens by SPME, followed by GC/MS analyses. Their attracting effect to PRM was confirmed with tube olfactometer experiments. A synergic effect of four molecules characterizing gallinaceous bird scent [(E)-non-2-enal, nonanoic acid, octanal and oct-1-en-3-ol] was highlighted by olfactometry (Roy *et al.*, 2018). More generally, the attracting capacity of VOCs from the skin and feather extracts of hens (composition not available in the paper) was identified in a study where it was necessary to attract PRM to feed on synthetic membranes (Harrington *et al.*, 2010). However, it is not clear whether these molecules are involved in attracting *D. gallinae* to the host (*i.e.* detection of skin VOCs) or whether they serve as are contact cues for skin recognition. Several studies have investigated kairomones produced by hens; however, knowledge remains limited about the potential attracting role of VOCs in

the environment of hens, with future studies being required on this subject.

Few studies have investigated the kairomones of other mite species. The methodology exploited with other species is similar to that used with PRM. After analysing the body extracts of male and female adult houseflies *Musca domestica* and after feeding experiments, N-phenyl-N-glucoside and N-phenyl-N-mannoside were identified as two kairomones that attracted the phoretic mite, *Macrocheles muscaedomesticae* (Wicht *et al.*, 1971; Achiano and Giliomee, 2006 cited in ref of the review of Carr and Roe, 2015). In addition, female predator mites *Phytoseiulus macropilis* were attracted by unidentified kairomones from *T. urticae* in Y-tube olfactometer experiments (Amin *et al.*, 2010). However, three kairomones (methyl palmitate, ethyl palmitate and methyl linolenate) were identified in *Varroa jacobsoni* and *Varroa destructor*, which are two species of honey bees parasites, following behavioural experiments with four-armed olfactometers and mite wind tunnels (Le Conte *et al.*, 1989; Calderone and Lin, 2001). The use of a wind tunnel allows the long-range effect of different compounds to be tested. The use of this methodology on PRM could extend current knowledge on the potential attracting properties of pheromones and kairomones.

### Repulsive substances

Olfactive repellents can be divided into two categories: (1) true repellents (also called expellents) or spatial repellents, and (2) odour masking compounds (or attraction inhibitors) (Deletre *et al.*, 2016). A true repellent induces a shift in orientation from an odour source without direct contact. In comparison, odour masking compounds induce the attractiveness of the host to decline, or difficulties to localize the host using odour cues (Deletre *et al.*, 2016).

Several experiments were conducted to test the repellent properties of substances studied on PRM, including contact experiments (i.e. Petri dishes with filter paper saturated with the compound) and olfactive experiments using olfactometers (Nechita *et al.*, 2015; Masoumi *et al.*, 2016). Globally, it is interesting to evaluate the repellent effect of a substance to: (1) test a potential negative repellent effect of a toxic substance (e.g. acaricide) (Lundh *et al.*, 2005) and (2) confirm the repulsive effect of a substance that could be used as a pure repellent (Nechita *et al.*, 2015). The different compounds tested on PRM are summarized in Table 3, showing the level of repulsive effect.

### Essential oils

The repulsive and/or acaricidal efficiency of essential oils has been extensively studied. For instance, Lundh *et al.* (2005) showed that neem oil, *Azadirachta indica* (Jussieu) (concentrations of 15 to 20%), has a good acaricide effect associated with a low repellent effect under laboratory conditions. In another study, Camarda *et al.* (2018) tested the effectiveness of this oil under field conditions. The authors showed that a 20% neem oil formulation (RP03<sup>TM</sup>) caused the mite population in poultry farms to decline by 94.65 to 99.80% after three nebulizations of the product over one week (Camarda *et al.*, 2018). Other essential oils are also effective, particularly essential oils from the family of *Lamiaceae* (Martinov) such as *Origanum* (L.), *Satureja* (L.), *Thymbra* (L.) and *Thymus* (L.) species. These species are characterized by a major compound, carvacrol, which is a good acaricide candidate with a toxic effect on *D. gallinae*. It has no repellent effect at 1% concentration; however, above this concentration, repellence occurs (Kirimer *et al.*, 1995; Barimani *et al.*, 2016).

Out of several essential oils tested for their pure repellent properties during *in vitro* contact bioassays, thyme and lavender oils

had the highest repellent activity with, respectively, 80 and 40% of the oil-treated surface area being avoided by PRM (Nechita *et al.*, 2015; Pritchard *et al.*, 2016). Nevertheless, in this type of experiment, it is not possible to distinguish whether mites are repelled by olfactory or contact cues, with complementary studies in olfactometers being required (Deletre *et al.*, 2016). The association of thymol (i.e. the major compound of thyme essential oil) and carvacrol was evaluated in olfactometer bioassays for potential synergic repellent effects (Masoumi *et al.*, 2016). The study, unfortunately, revealed that the addition of thymol to carvacrol decreased the repellent effect of carvacrol. Other studies analysed the different compounds present in a given essential oil by GC-MS experiments, and tested the repellent effect of the oil on PRM by Y-tube olfactometer experiments (Tabari *et al.*, 2017; Santana *et al.*, 2018). The essential oil of *Artemisia sieberi* (Besser) [rich in  $\alpha$ -thujone (31.5%),  $\beta$ -thujone (11.92%), camphor (12.3%) and 1,8-cineole (10.09%)] and the essential oil of *Myrcia oblongata* (DC) [rich in caryophyllene oxide (22.03%) and trans-verbenol (11.94%)] showed a significative repellent effect to *D. gallinae*. Nevertheless, the effect of each of their constituent compounds was not evaluated (Tabari *et al.*, 2017; Santana *et al.*, 2018). Finally, a study by Lee *et al.* (2019) obtained interesting results, in which two essential oils (cinnamon bark and clove bud oils) and some of their constitutive compounds were tested for their repellent abilities. The two oils and their compounds were repellent to PRM, except for cinnamyl acetate from cinnamon bark oil, and for two compounds from clove bud oil (eugenol and eugenol acetate), which even showed an attractant effect, following 120 min of application. The concentration of the active compound over time might be an issue, as many essential oils exhibit high repellent activity against *D. gallinae* over the short-term due to their high volatility (George *et al.*, 2008, 2009). A dose effect exists for several molecules, including thyme oil, which could be used as a repellent or as an acaricide, depending on the concentration. When used as an acaricide, essential oils have a short-term effect; however, it is possible to couple them with other biological control methods, such as entomopathogenic fungus. When these two methods are coupled, the short-term acaricide effect of essential oils is complemented with the delayed, but long-term effect, of the fungus. For example, an entomopathogenic fungus [*Beauveria bassiana* (Balsamo-Crivelli) Vuillemin] in combination with essential oils [*Eucalyptus globulus* (La Billardière), *Eucalyptus citriodora* (Hook), *Thymus vulgaris* (Linné), and *Eugenia caryophyllata* (Thunb.)] was tested to create a new way to control *D. gallinae* (Immediato *et al.*, 2016). Following toxicity tests, it was found that the combination of *B. bassiana* and *E. globulus* at 0.2% might be the best combination for an acaricide effect against *D. gallinae*.

### Plant extracts

Most studies on plant extracts have focused on the acaricide properties of the extracts, and are not evaluated in this review, because repellent properties are not discussed. Only two studies evaluated the repellent properties of two plant extracts. Rajabpour *et al.* (2018) showed that aquatic and ethanolic extracts of *Conocarpus erectus* are repellent for PRM using contact experiments. Nevertheless, the repellent effect described here was unclear, as it was not possible to distinguish the contact and olfactive repellent effects. In another study, the acaricidal and repellent activities of a methanolic extract and fractions from the rhizome of *Cnidium officinale* were tested to control *D. gallinae* (Kim *et al.*, 2018). Kim *et al.* (2018) showed that the methanolic extract of *C. officinale* has a 91.3% repellent activity on PRM, while (Z)-ligustilide, which is an isolated compound from *C. officinale*

**Table 3.** Chemical compounds with a high repellent effect on *D. gallinae*

Tested substances	Species	Experiments	Effect	References
2-Methoxycinnamaldehyde	<i>Dermanyssus gallinae</i>	Y-tube olfactometers	High repellent effect	Lee <i>et al.</i> (2019)
$\alpha$ -Thujone-rich <i>Artemisia sieberi</i> Besser essential oil		Y-tube olfactometers		Tabari <i>et al.</i> (2017)
$\beta$ -Caryophyllene		Y-tube olfactometers		Lee <i>et al.</i> (2019)
Carvacrol		Y-tube olfactometers		Barimani <i>et al.</i> (2016); Masoumi <i>et al.</i> (2016)
Cinnamaldehyde		Y-tube olfactometers		Lee <i>et al.</i> (2019)
Cinnamon bark oil		Y-tube olfactometers		Lee <i>et al.</i> (2019)
Clove bud oil		Y-tube olfactometers		Lee <i>et al.</i> (2019)
Coumarin		Y-tube olfactometers		Lee <i>et al.</i> (2019)
<i>Cnidium officinale</i> Makino methanolic extract		Y-tube olfactometers Fumigation bioassays Contact bioassays Spray bioassays		Kim <i>et al.</i> (2018)
<i>Conocarpus erectus</i> L. extract		Filter paper bioassays		Rajabpour <i>et al.</i> (2018)
Lavender oil		Filter paper contact bioassays Pasteur pipette bioassays		Nechita <i>et al.</i> (2015)
<i>Myrcia oblongata</i> DC. leaves oil 10%		Y-tube olfactometers Fumigation bioassays		Santana <i>et al.</i> (2018)
Neem oil		Field assays Cardboard traps Filter paper in Petri dishes bioassays		Lundh <i>et al.</i> (2005); Camarda <i>et al.</i> (2018)
Thyme oil		Petri dishes bioassays Y-tube olfactometers Filter paper contact bioassays Pasteur pipette bioassays		George <i>et al.</i> (2009); Nechita <i>et al.</i> (2015); Masoumi <i>et al.</i> (2016); Pritchard <i>et al.</i> (2016)
(Z)-ligustilide	Y-tube olfactometers Fumigation bioassays Contact bioassays Spray bioassays	Kim <i>et al.</i> (2018)		

roots, showed a repellent response rate equal to 100% using a T-tube olfactometer.

### Application of VOCs in field assays

Here, current applications of VOCs to control PRM are described along with potential other strategies from other Acari species relevant for PRM control. The introduction of repulsive substances to hen feed could make the emission of repellent VOCs against the PRM possible (El Adouzi *et al.*, 2019). For instance, Nor-Mite<sup>®</sup> (mostly composed of >2% eugenol) was added to hen feed, and the VOCs emitted by hens were analysed. GC-MS analysis showed that Nor-Mite<sup>®</sup> fed hens released several VOCs, including eugenol. Following olfactometer experiments, the repellent effect of eugenol and other VOCs from Nor-Mite<sup>®</sup> fed hens was confirmed. In addition, the authors suggested that other molecules present in Nor-Mite<sup>®</sup>, such as trans-caryophyllene and geraniol, might contribute to this repellent effect, with possible additive/synergic effects (not tested in this paper) (El Adouzi *et al.*, 2019).

Several devices have been tested to control other Acari species. For example, one design using attracting VOCs is an 'attract and kill' trap has been applied to others species of Acari. A system based on the encapsulation of an attracting pheromone coupled to an acaricide was developed for the brown dog tick

*Rhipicephalus sanguineus* (Latreille 1806) by Dhivya *et al.* (2014). The capsule was made of a natural polymer (*i.e.* chitosan). The species assembly pheromone was chosen as an attractant, and was coupled with deltamethrin as acaricide. Laboratory and field tests showed a reduction of infestation, as the ticks were discovered dead around the traps. Another study using aggregation-attachment pheromone together with deltamethrin to attract and kill *Amblyomma variegatum* (Fabricius 1794) (the tropical bont tick) demonstrated an overall tick control efficiency of 92% (Kelly *et al.*, 2014). Similar studies could be conducted for *D. gallinae* by coupling the aggregation pheromone with a natural acaricide (*e.g.* an essential oil) or entomopathogenic fungi.

### Conclusions

PRM is a world economic problem for the poultry industry, and the development of solutions with low risk on human health should be promoted to control their populations, especially in hen egg laying farms. The development of sustainable pest control based on an understanding of chemical ecology is being increasingly implemented, and should be applied to PRM. New insights on pheromonal communication of PRM are fundamental to create efficient control strategies.

Knowledge remains more limited about chemical communication in mites including PRM compared to ticks, mainly due to their smaller size and, consequently, the lower emission of VOCs. In the last decade, the identification of attractants in mites has increased, along with more information on their associated effects on mite behaviour. Nevertheless, a large knowledge gap exists on VOCs that attract PRM that should be addressed in the future.

The lack of knowledge, especially on PRM pheromones, could be explained by the difficulties encountered to maintain an artificial breeding of PRM (Kirkwood, 1971; Zeman, 1988; Bruneau *et al.*, 2001; Harrington *et al.*, 2010). This hurdle prevents the availability of fed mites reducing the implementation of pheromones studies. For the development of further PRM pheromone studies, an efficient artificial or semi-artificial breeding system must be developed to facilitate the supply of fed individual as well as to allow a control of several parameters (e.g. time since the last meal, female fertilization or not *etc.*). Among the current possible alternative, the use of a semi-artificial laboratory breeding on alive chicken (*i.e.* a plastic bag filled with PRM in contact with the chicken) seems to be a promising solution as it provides a high number of individuals in a short period of time (Nunn *et al.*, 2019).

Concerning PRM's kairomones, several studies are currently available with no assays of these molecules in field conditions (Pageat, 2005; Koenraadt and Dicke, 2010; Roy *et al.*, 2018). Different implementations are still to be needed to consider these molecules as an efficient field attractant such as the emission source apposition or the concentration of the synthetic molecule. Moreover, the interference of kairomones from hosts when synthesized kairomones are applied in layer farms would be difficult to avoid and should decrease the efficacy of the product.

In contrast to the limited number of studies on attracting VOCs, many studies exist on repulsive VOCs to PRM. This phenomenon might be explained by the fact that repelling PRM outside poultry farms is the first solution that comes to mind in terms of control strategies. Essential oils and plant extracts seem to represent promising repellents, despite their short-term efficiency. Several designs using VOCs on PRM have been elaborated and tested under field conditions. Further studies are needed to optimize these strategies, or to create new ones based on 'attract and kill' or 'push-pull' principles.

Finally, although mites and ticks are two separated groups in the Acari taxon, similarities exist in how they communicate. Therefore, researchers investigating the chemical communication of mites should refer to studies on chemical communication in ticks, for which there is extensive published literature.

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