

Innovative Formulation Of Paraffin And Silicon Oils For The Control Of Poultry Red Mite (*Dermanyssus gallinae*)- 3. Comparative Clinical Examination Of Efficacy With Active Matter SiO₂

A.Pavlicevic¹, R. Ratajac², I.Stojanov², I.Pavlovic³

¹(AVES MIT" DOO, Subotica-Bajmok, 24210 Bajmok, Cluster "Dermanyssus gallinae", Serbia)

²(Scientific veterinary institute „Novi Sad“, Novi Sad, Serbia)

³(Scientific veterinary institute of Serbia, Belgrade, Serbia)

ABSTRACT: Physical control, especially inert substances are an alternative to insecticides in the control of *Dermanyssus gallinae* (De Geer, 1778). The use of SiO₂ formulations is technically demanding. SiO₂ formulations have a number of limiting characteristics. However, if the formulation is selected appropriately and the facility hygienically prepared, the combined professional application of SiO₂ and facility rest period make for an efficient method of *D. gallinae* control. In this case, the efficacy of control can be achieved in the form of suppression for a one-year period (only with the preparatory treatment). In case of optimal conditions, the procedure of a proper combined application of SiO₂ formulations can lead to eradication of *D. gallinae* from production facilities. In contrast to this, the application of SiO₂ formulations in unconditioned situations, without facility rest period, as well as during exploitation, especially with high intensity infestation, is not sufficiently efficient and rational. The use of inert oils P 547/17 is technically simple, with the possibility of high operability. In comparison with SiO₂ formulation, P 547/17 has a number of better characteristics and is much more efficient on cages and equipment. On absorbent surfaces (floor and walls), a repeated application is required, as there is no possibility of creating a layer with a prolonged effect. The appropriate application of the P 547/17 formulation enables easier and faster highly efficient suppression, even eradication from production facilities, depending on the conditions being met. Not meeting the set conditions reduces the desired effect. The appropriate application of the selected inert substances in facility preparation for the housing of the flock affirms preventive veterinary medicine, with a highest degree of safety and efficacy.

KEYWORDS: *D. gallinae*, P 547/17, SiO₂, clinical examination.

I. INTRODUCTION

Synthetic neurotoxic compounds (acaricides, in the wider sense insecticides) have, to this day, remained the dominant control method for the most important ectoparasite of poultry, poultry red mite *Dermanyssus gallinae* (De Geer, 1778). Their efficacy has been evident, in some cases extraordinary, however was not alone capable of preventing the spreading of *Dermanyssosis*. To their, primarily unprofessional application, the development of resistance to insecticides [1,2,3]. has been added as well as not less favorable environmental conditions for *D. gallinae* control, which came about with the abandonment of conventional cage systems. In this way, the effort needed for *D. gallinae* control has increased, which has consequentially raised the toxicological risk (level) to which the consumers, poultry and the environment were exposed to. The situation is further made worse, as illegal and products not registered for such a purpose are often used [4]. The existing control of poultry red mite needs improvement [5,6,7]. The reason for this is an obvious safety risk, and at the same time, the need for an efficient and rational *D. gallinae* control. The alternative to insecticides is physical control. The physical control of poultry red mite can be conducted using: temperature, light and inert substance [8]. According to the achieved clinical impact, and especially in respect to the potential for improving *D. gallinae* control [9,10,11,12,13] we highlight inert substances.

II. AIM

Comparing the efficacy on poultry red mite (*Dermanyssus gallinae*) of the formulation of paraffin and silicon oils (P 547/17) and SiO₂ formulations in clinical conditions, and assessing the justification of their application.

III. MATERIALS AND METHODS

Clinical examinations have been conducted with the professional controlled application of the selected formulations and monthly monitoring, predominantly based on visual check-ups and early detection method in the period from 2012 to 2018 [10].

The examinations have been conducted in conventional and enriched cage systems.

IV. RESULTS

Table 1. Tabular display for example of application efficacy of the powder form of SiO₂

Number	Capacity of the facility, type of cage and housing	Method	Duration of suppression (months)	Finding (-/+) (months)
1	19000, Conventional, E	DE (no.2)	5	Vis.
2	50000, Conventional, E	DE (no.2)	4	Vis.
3	40000, Conventional, E	DE (no.2)	6	Vis.
4	25000, Conventional, E	DE (no.2)	7	Vis.
5	2500, Conventional, E	DE (no.2)	3	Vis.
6	50000, Conventional, E	DE (no.2)	5	Vis.
7	25000, Conventional, H	DE (no.2)	1	Vis.
8	25000, Conventional, H	DE (no.2)	1	Vis.
9	25000, Conventional, H	DE (no.2)	2	Vis.
10	25000, Conventional, H	DE (no.2)	3	Vis.

Key:(-) negative finding by early detection of *D. gallinae* method, (+) detected presence of *D. gallinae* by early detection mehtod; Vis. Visual checkup; H – treated housed facility; E – treated empty facility

Table 2. Tabular display for example of application of liquid form of SiO₂

Number	Capacity of the facility, type of cage and housing	Method	Duration of suppression (months)	Finding (-/+) (months)
1	14000, Conventional, E	Liquid, (no.9)	6	Vis.
2	35000, Conventional, E	Liquid, (no.9)	7	Vis.
3	42000, Conventional, E	Liquid, (no.9)	10	Vis.
4	70000, Enriched, E	Liquid, (no.9)	2	Vis.
5	14.000, Conventional, E	Liquid, (no.9)	6	Vis.
6	7.600, Conventional, E	Liquid, (no.9)	12	Vis.
7	18.000, Conventional, E	Liquid, (no.9)	3	Vis.
8	36.000, Conventional, H	Liquid, (no.9)	2	Vis.
9	35.000, Conventional, H	Liquid, (no.9)	2	Vis.
10	50.000, Conventional, H	Liquid, (no.9)	3	Vis.

Key: (-) negative finding by early detection of *D. gallinae* method; (+) detected presence of *D. gallinae* by early detection mehtod; Vis. Visual checkup; H – treated housed facility; E – treated empty facility

Table 3. Tabular display for example of combined application of SiO₂

Number	Capacity of the facility, type of cage and housing	Method	Duration of suppression (months)	Finding (-/+) (months)
1	5000, Conventional, E	Comb., no (2+9)	6	Vis.
2	22000, Conventional, E	Comb., no (2+9)	5	Vis.
3	2500, Conventional, E	Comb., no (2+9)	12	12 (-)
4	45000, Conventional, E	Comb., no (4+9)	12	10 (-)
5	25000, Conventional, E	Comb., no (4+9)	12	12 (-)
6	6 x 25000, Conventional, E	Comb., no (4+9)	12	12 (-)
7	18.0000, Conventional, E	Comb., no (4+9)	12	10 (+)
8	18.000, Conventional, E	Comb., no (4+9)	5	Vis.
9	2.500, Conventional, H	Comb., no (4+9)	3	Vis.
10	45000, Conventional, H	Comb., no (4+9)	2	Vis.

Key: (-) negative finding by early detection of *D. gallinae* method; (+) detected presence of *D. gallinae* by early detection method; Vis. Visual checkup; H – treated housed facility; E – treated empty facility

Table No. 4. Tabular display of example for application of P 547/17

Number	Capacity of the facility, type of cage and housing	Percentage in a water emulsion	Duration of suppression (months)	Finding (-/+) (months)
1	25000, Conventional, E	15	Pending	from 7. (+)
2	2000, Conventional, E	15	Pending	8 (-)
3	4500, Conventional, E	15	Pending	8 (-)
4	28000, Conventional, E	17, 19	3,5	Vis.
5	50.000, Conventional, E	20	Pending	6 (-)
6	50.000, Conventional, E	20	Pending	3 (-)
7	25000, Conventional, E	20	Pending	3 (-)
8	18000, Enriched, E	20	4	Vis.
9	200, Conventional, E	20	Pending	8 Vis. (+)
10	18000, Conventional, E	15 x 2	Pending	7 (-)

Key: (-) negative finding by early detection of *D. gallinae* method; (+) detected presence of *D. gallinae* by early detection method; Vis. Visual checkup; H – treated housed facility; E – treated empty facility

V. DISCUSSION

The impact of the physical and chemical making of SiO₂ formulation on biological efficacy of *D. gallinae* were examined by Shulz et al. (2014), and have apart from this also pointed out the importance of the application technology. The subject of our research is focused on the application and defining the most important characteristics which the use of inert oils in practice reveal.

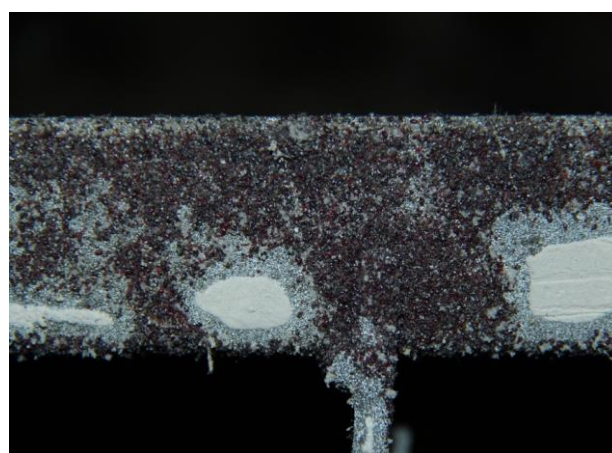
The efficacy of SiO₂ formulations have previously been established in laboratory conditions, samples 2, 4 and 9 [10,11,14].

Fig 1. Effect of natural diatomaceous earth (sample 2).

Based on the results of biological efficacy, we have conducted a selection of formulations for clinical examination. We have singled out characteristic examples



Fig 2. Detail from a cage without considerable effect of diatomaceous earth on *D. gallinae*.



The problem of the lack of a standardize method for laboratory and clinical examination of efficacy on *D. gallinae* burdens these examinations as well.

The varying of results with the same formulation has been a result of: discrepancies in the facility rest period duration, environmental and technological conditions, hygienic preparation of the facility, intensity and

extensity of the infestation, the infestation of the new flock and transport cages.

The characteristics of SiO₂ formulations are:

1. Application requires professional training and devices meant for powder form and liquid suspensions;
2. There is a problem in the distribution of the formulation at application. The powder form is applied well on equipment by electrostatic attraction. Even so, it cannot be technically applied to all the important spots on cages and equipment. Water suspensions have much better application possibilities, but their use with certain surfaces is irrational and technically unfeasible. This is why the complexness of the environment for SiO₂ formulations plays a very important role;
3. The powder form is easily removed, as is the liquid form while wet;
4. Their effect is exclusively through contact;
5. Laboratory examinations determine a different efficacy of formulations from inefficient to efficient [15,16,14]. Efficient formulations have all the necessary characteristics for the control of *D. gallinae*. Because of great variation of SiO₂ efficacy, it is recommended to make the selection according to the laboratory determined efficacy;
6. The lethal effect is slow, during which time gravid females lay eggs and therefore significantly diminish the control effect. These notions contradict the research of Maurer et al. [16] who have determined the cessation of the reproductive cycle;
7. They gave a good residual effect, the manifestation of which, however, is condition by the overall set of circumstances;
8. They exhibit a small lethal effect per surface unit, which together with the two previous characteristics, leads to insufficient efficacy at application in housed facilities. Especially in combination with a high infestation, filthy surfaces, removal of the layer and the laying of eggs of lethally exposed gravid females;
9. Behavioral adaptation [17]. is a highly pronounced mechanism, by which the infestation of *D. galliane* successfully adapts to the deficiencies of SiO₂ formulations (tabular display 1,2, 3/1, 2, 8-10);
10. The repellent effect additionally decreases efficacy and stimulates *D. gallinae* adaptation;
11. Freezing temperatures damages the water suspension layer in an empty facility and reduces its efficacy;
12. The humidity of the air is also important for efficacy of SiO₂ formulations [15].;
13. Impurities disallow contact and prevent the effect of SiO₂ formulations;
14. The formulation for preparation in a water suspension have a higher price than the powder form. Apart from the production process, the mites themselves create impurities, which are added to others, thus creating a favorable environment.

Fig.3. Behavioral adaptation of *D. gallinae* to the application of SiO₂ formulations.



Fig. 4. Detail of a cage, an example of an insufficient efficacy of the liquid formulation based on SiO₂ in a housed facility.



The examples from tabular displays 1/7-10; 2/8-10; 3/9, 10 point to the insufficient efficacy of SiO₂ based formulations if applied during flock exploitation. The efficacy is smaller the greater the intensity and extensity of *D. gallinae* infestation which is being treated.

Because of these characteristics, the formulations based on SiO₂ have thus far not been able to eliminate the dominant use of insecticides in the control of *D. gallinae*. There are advantages and disadvantages of the powder and liquid forms [18,19]. The many deficiencies of the powder and liquid forms of SiO₂ formulations can be overcome through their combined use [6,11,12]. This is achieved if the liquid form is applied on all primarily important places of the cages and equipment, especially those where the powder form cannot be distributed to, and where the persistent layer with the prolonged effect of the liquid form is well manifested. In places where the distribution of the liquid form is irrational, layers of natural diatomaceous earth are placed. Apart from this, important structures are completed with the powder form, so as to optimize the layer within the facility. An appropriate and detailed distribution of the SiO₂ layer in the facility with the liquid and powder forms is an important assumption, but is on its own still not sufficient for the success of control. The reason for this is hidden, inaccessible spots where *D. gallinae* specimens are not directly exposed to the formulations during application. This problem can be solved with the rest period of the facility. The importance of this rest period lies in the following:

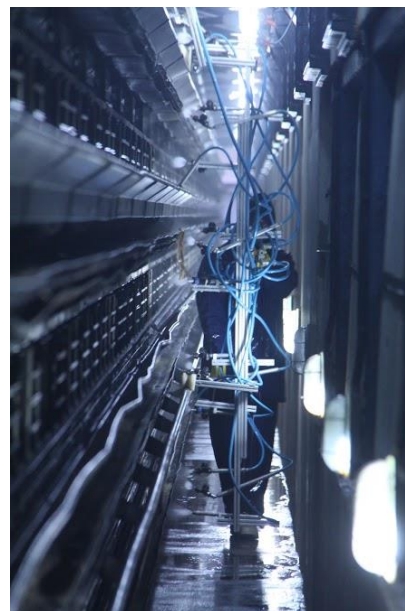
1. Change in the behavior of *D. gallinae* in an empty facility;
2. Stopping reproduction after the consumed blood is spent and
3. Avoiding deficiencies (distribution; slow effect with a limited lethal capacity) and bolstering the advantages of the SiO₂ formulations.

Another condition needs to be met. During facility rest period it is important that the temperature is kept within range of the physiological activity of *D. gallinae*. The higher the temperature, the greater its contribution is to the efficacy of the control measures conducted. During facility rest period *D. gallinae* has no dominant movement between hiding places and host. The movement is done in different directions, in search of a host, meaning that the mites are able to come into contact with SiO₂ formulations and become more exposed for elimination. In this way, the applied layer of the formulation based on SiO₂, in the conditions described, is able to achieve the desired results (tabular display 3/3-6). However, this is only the case with the *D. gallinae* infestation already present in the facility.

Fig. 5. An example of a prepared facility by combined method, application of the powder and liquid forms of the SiO₂ formulations.



Fig. 6. Application of the formulation of inert oils (P 547/17) in practical conditions



The housing of a new flock can lead to re-infestation with poultry and transport cages (tabular display 3/8). This is why biosafety measures are needed to prevent this risk. After powder, and then liquid SiO₂ formulations, the third generation of inert substances are water emulsions with inert oils (P 547/17). In comparison to SiO₂ formulations, we have established the following:

1. The application of P 547/17 is not as challenging in terms of training, nor requires a special type of applicator. Application devices for water solutions and liquid types of silicates can be use. The quality and position of the nozzle is adaptable. It can be applied manually, but is suitable for great applicability with machine applicators. It has not got such great training requirements. It does not require a special protective equipment, so working is easier for the staff which does the application. This minimizes the human error factor and optimizes the operability of conducting application;
2. The water emulsion has a better quality distribution in a facility and greater penetrative power. Because of a less detailed distribution and mode of action of P 547/17, the complexity of the facility has a lesser importance, and there is greater efficacy in comparison with SiO₂ even without facility rest period. However, the application is optimized with facility rest period;
3. The removal factor is somewhat smaller, but the importance of hygienic conditions is greater;
4. The effect is also exclusively through contact, which is helped by immobilization;
5. It has a faster and more efficient effect on the directly exposed;
6. Gravid females lay fewer eggs, which are directly exposed to the prolonged effect. Even with the subsequently exposed, in the second period immobilization is achieved;
7. The prolonged effect on nonabsorbent surfaces (cages and equipment) is highly efficient and lasting. It is the best recorded so far for *D. gallinae*, and used more than SiO₂. After housing, the prolonged effect continues, however the crucial impact on the level of the residual effect will be made by hygienic conditions. On absorbent surfaces (like floor and in some cases walls) there is no residual effect. This is why application has to be repeated on these surfaces, for mites which have subsequently arrived on the surface. The speed and efficiency also affect the need for a particular length of the facility rest period. Rest period is important with P 547/17 too, but to a somewhat smaller degree than with SiO₂;
8. It has a greater lethal effect per surface unit. No repellent effect has been recored, as was the case with SiO₂ formulation;
9. It does not create chemoresistance. The possibility of adaptive behavior is low;
10. We have not noticed the existence of a repellent effect during the distribution of the working emulsion in the facility, in contrast to the liquid and powder forms of SiO₂ formulations;
11. Freezing temperatures are not desirable, however we have not been able to assess the impact of low temperatures;
12. Air humidity has no significant impact on the efficacy of application;
13. The leftover layer of SiO₂ formulation disables the prolonged effect of P 547/17 (table 4/4, the residues of SiO₂ formulation on cages and equipment and impurities). Fat layers and impurities have a negative impact as they cannot be washed (table 4/4, fat layers and a short facility rest period). The light greasy layer of P 547/17 increases the dustiness of the cages and equipment. However, with some extra effort, good quality of hygienic conditions can be met in the environment. The lubrication can lead to belts slipping. In this case lime, diatomaceous earth or something similar is used to remove this problem;
14. It is nonabrasive for cages and equipment;
15. The formulation is nontoxic and the safety is further determined through the application method (in an empty facility before flock housing);
In contrast to SiO₂ formulations the water emulsion P 547/17 is easily applicable in the treatment of transport cages and secondhand cage and equipment. In this way, the preventive concept can be completed with biosafety measures.
Camarada et al. [20] have examined 20% neem ether oil (RP03™) azadirachtin in a housed facility. The procedure described (3 treatments over the course of 7 days) and suppression efficacy within two months. Such results are by far smaller than the achieved efficacy of P 547/17.
The possibility of *D. Gallinae* eradication with inert oils P 547/17 has not been proven yet. However, existing results in practice (table 4/2, 3, 10) and laboratory condition [14].point that this will be achieved.

VI. CONCLUSION

The appropriate application of the selected inert formulation in the preparation of the facility, with facility rest period and in temperature conditions for *D. gallinae* activity, even will all its deficiencies and limitations, allows a high level of suppression, and with an optimized procedure, also the eradication of *D. gallinae* from production facilities. In this way preventive veterinary medicine is affirmed, with the highest levels of safety and efficacy.

REFERENCES

1. A.Giangaspero, M. Marangi, S.Pati, M.A.Cafiero, C.Camarda and O.A.E. Sparagano, Investigating the Presence of Acaricide Residues in Laying Hens Naturally Infected by the Red Mite *Dermanyssus gallinae*. Proc. 12th Asean Food Conf. BITEC Bangna, Bangkok, Thailand, 2011, 223.
2. M.Marangi, V.Morelli, S.Pati, A.Camarda, and M.A.Cafiero, Acaricide Residues in Laying Hens Naturally Infested by Red Mite *Dermanyssus gallinae*. PLoS ONE 7(2), 2012, e31795. doi:10.1371/journal.pone.0031795.
3. A.Pavličević, I.Pavlović, N.Stajković, and P.Bratislav, Evidence for Resistance to Carbaryl in Poultry Red Mites from the Republic of Serbia and Montenegro. *Animal Science and Biotechnologies*, 49 (1), 2016,222-225.
4. A.Giangaspero, K.Bartley, M.Mul, E.Papadopoulous, L. Roy, D.Horvatek Tomic, and O. Sparagano, The anarchy in chemical control of *Dermanyssus gallinae*, and the need to establish specific EU regulations: the efforts of a COST Action // Abst.WVPA XXth Congress, Edinburgh, UK, 2017, 122.
5. J.Pritchard, T.Kuster, O.Sparagano, and F.Tomley, Understanding the biology and control of the poultry red mite *Dermanyssus gallinae*: a review. *Avian Pathology*, 44(3),2015, 143-153.
6. A.S. Flochlay, E. Thomas, and O. Sparagano, Poultry red mite (*Dermanyssus gallinae*) infestation: a broad impact parasitological disease that still remains a significant challenge for the egg-laying industry in Europe. *Parasites & Vectors* 10, 2017, 357, DOI 10.1186/s13071-017-2292-4.
7. A.Pavlicevic, R.Ratajac, I.Pavlović, D. Horvatek Tomić, and I.Stojanov, Approach to *Dermanyssus gallinae* eradication – use of inert compounds and integrated health protection of poultry inertnih jedinjenja i integralna zdravstvena zaštita živine. Abs. XX Symposium of epizootiologists and epidemiologists, with international participation, Vrnjacka Banja, 2018, 184-185
8. O.A.E.Sparagano, D.R. George, D.W.J. Harrington, and A.Giangaspero, Signification and Control of the Poultry Red Mite, *Dermanyssus gallinae*. *Annual Review of Entomology*, 59, 2014, 447-466.
9. A.Pavličević, A.Vasić, I. Pavlović, and JongUng Yoon, Detección temprana del ácaro rojo. *Albéitar (Esp)* 205, 2017, 24-26.
10. A.Pavličević, JongUng Yoon, I.Pavlović, M.Milanović, and T. Petrović, Kontrola *Dermanyssus gallinae* i program mera kontrole zaraznih bolesti – predlog integralne zdravstvene zaštite. Abs.XIX Simpozijum epizitologa i epidemiologa, Vršac, Srbija, 2017, 171-172.
11. A.Pavlicevic, JongUng Yoon, and I.Pavlovic I. Control of red poultry mite (*Dermanyssus gallinae*) by mechanical effect: the choice of current improvements of formulations, application and concept. Abst.3 rd COST Conference Oeiras, Portugal, 2017, 27.
12. A.Pavličević, R.Ratajac, M.Dotlić, I.Stojanov, and I.Pavlović, An innovative formulation of paraffin and silicone oils for the control of the red poultry mite (*Dermanyssus gallinae*) – examination of the efficiency under laboratory conditions. *Arhiv veterinarske medicine*, 10(2), 2017, 63-79.
13. A.Pavlicevic, JongUng Yoon, and I.Pavlović, The fipronil affair, pesticides in eggs - why it happened and can it be prevented from happening again. *Invention Journal of Research Technology in Engineering & Management* 2 (3), 2018, 30-35.
14. A.Pavlicevic, R.Ratajac, M.Dotlic, I.Stojanov, and I.Pavlovic, Innovative formulation of paraffin and silicone oils for control of poultry red mite (*Dermanyssus gallinae*) – 2. comparative laboratory efficacy examinations with active matter SiO₂. Abs.International Scientific Symposium "Animal breeding and pathology to day", Timisoara, 2018, 12.
15. O.Kilpinen, and T. Steenberg, Inert dusts and their effects on the poultry red mite (*Dermanyssus gallinae*). *Experimental and Apply Acarology*, 48, 2009, 51–62.
16. V. Maurer, and E. Perler, F.Heckendorn, In vitro efficacies of oils, silicas and plant preparations against the poultry red mite *Dermanyssus gallinae*. *Experimental and Apply Acarology*, 48, 2009, 31–41 DOI 10.1007/s10493-009-9254-2.
17. W. Ebeling, Sorptive dusts for pest control. *Annual Review of Entomology*, 16, 1971, 123–158.
18. J.Schulz, J.Berk, J.Suhl, L.Schrader, S.Kaufhold, I.Mewis, H.Mohammed Hafez, and C.Ulrichs, Characterization, mode of action, and efficacy of twelve silica-based acaricides against poultry red mite (*Dermanyssus gallinae*) in vitro. *Parasitol Research*, 113, 2014, 3167–3175, DOI 10.1007/s00436-014-3978-6
19. J.Schulz Maßnahmen zur Bekämpfung der Roten Vogelmilbe (*Dermanyssus gallinae*) in der ökologischen Legehennenhaltung. Inaugural-Dissertation zur Erlangung des Grades eines Doktors der Veterinärmedizin an der Freien Universität Berlin. Berlin, Germany Journal-Nr.: 3702
20. A. Camarda, N. Pugliese, A. Bevilacqua, E. Circella, L. Gradoni, D. George, O. Sparagano, and A.Giangaspero, Efficacy of a novel neem oil formulation (RP03™) to control the poultry red mite *Dermanyssus gallinae*. *Medical and Veterinary Entomology*, 2018, doi: 10.1111/mve.12296.