



Exzolt™

FLURALANER

TREATMENT FOR POULTRY MITE INFESTATIONS IN CHICKENS
TECHNICAL MANUAL

BREAKTHROUGH MITE CONTROL

Exzolt™, new from MSD Animal Health, wipes out over 99% of poultry mites (Poultry Red Mites and Northern Fowl Mites), making it the most impactful and effective solution for infestations. And because it's simply applied through your poultry drinking system, there's no special training or new equipment needed, meaning one person can treat all your birds faster than ever.

If you're serious about keeping your birds and your bottom line healthy, **visit Exzolt.com**

Exzolt™
FLURALANER

Exzolt.com



Exzolt™
FLURALANER

Exzolt 10 mg/ml solution for use in drinking water for chickens
Exzolt 10 mg/ml solution for use in drinking water for chickens
Exzolt 10 mg/ml solution for use in drinking water for chickens
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MSD



SAFER, SIMPLER, MORE EFFECTIVE

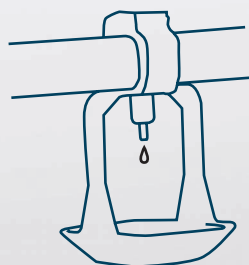
Exzolt provides 99%+ mite elimination in three simple steps. First, administer Exzolt to the hen's drinking water to eliminate live mites. Administration through the drinking water system avoids spraying and minimizes the potential of chemical exposure to the birds. Second, a second application to eradicate mite stages not susceptible to the first application.

To maintain the fast and nearly complete elimination of mites, implement the following bio-security measures, such as:

- ▶ Avoiding entrance of poultry mites with the allocation of new hen houses
- ▶ Changing outer clothing per house and use specific tools per each house
- ▶ Cleaning and checking for poultry mite absence before being used

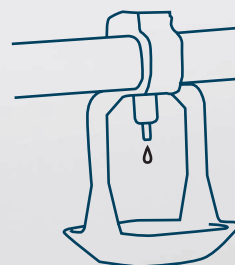
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FIRST APPLICATION OF EXZOLT
THROUGH DRINKING SYSTEM KILLS
BLOOD-FEEDING MITES



2

SECOND APPLICATION OF EXZOLT
EFFECTS OF TREATMENT TO KILL
ENTER THE BLOOD-FEEDING MITES



IVE MITE CONTROL

First, deliver the initial application through the drinking water reduces stress caused by human workers. Seven days later, apply the second application.

populations that Exzolt provides, follow proper

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MAINTAIN PROPER BIOSECURITY
MEASURES TO ELIMINATE
MITE REINFESTATION

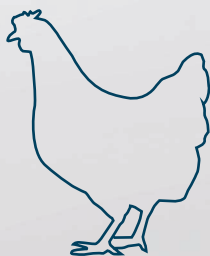


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SECTION ONE

POULTRY MITES





MITE PARASITES OF POULTRY

Though most health programs for chickens are focused on bacterial, viral, or coccidial threats, production birds are also vulnerable to parasites that can live on or in their bodies. Cage housing has greatly reduced the exposure of commercial layer flocks to internal nematode parasites and their intermediate carriers (although recent trends to cage free and free range production is causing some reversal of this trend), but the economic damage posed by external parasites has historically lagged far behind in technological advancement.

POULTRY RED MITES

Poultry Red Mites (*Dermanyssus gallinae*, Figure 1-1) are common skin parasites that are the most significant ectoparasite pest of laying hens worldwide and represent a severe threat to poultry production in Europe.¹ These extremely destructive nocturnal ectoparasites suck blood from host birds at night and then hide in the cracks, crevices, and litter of poultry houses when they are not feeding during the day.² Adult mites measure about 1 mm long, weigh about 76 µg (unfed) to 280 µg (after a blood meal), and are red in color after feeding, but they appear black, grey, or white without host blood in their system.³⁻⁶

POULTRY RED MITES

Kingdom ...*Animalia*
Phylum*Arthropoda*
Class*Arachnida*
Subclass.....*Acari*
Order*Mesostigmata*
Family*Dermanyssidae*
Genus.....*Dermanyssus*
Species.....*D. gallinae*

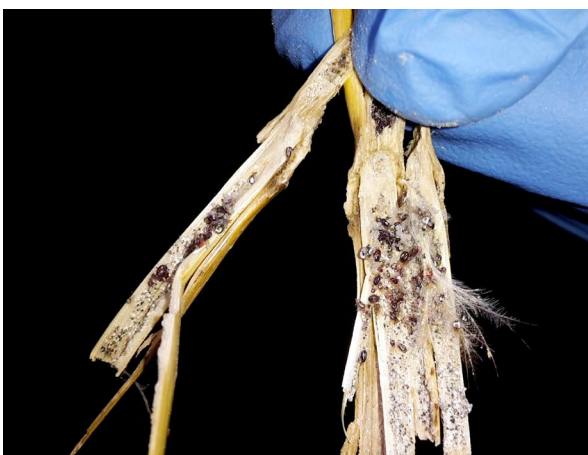


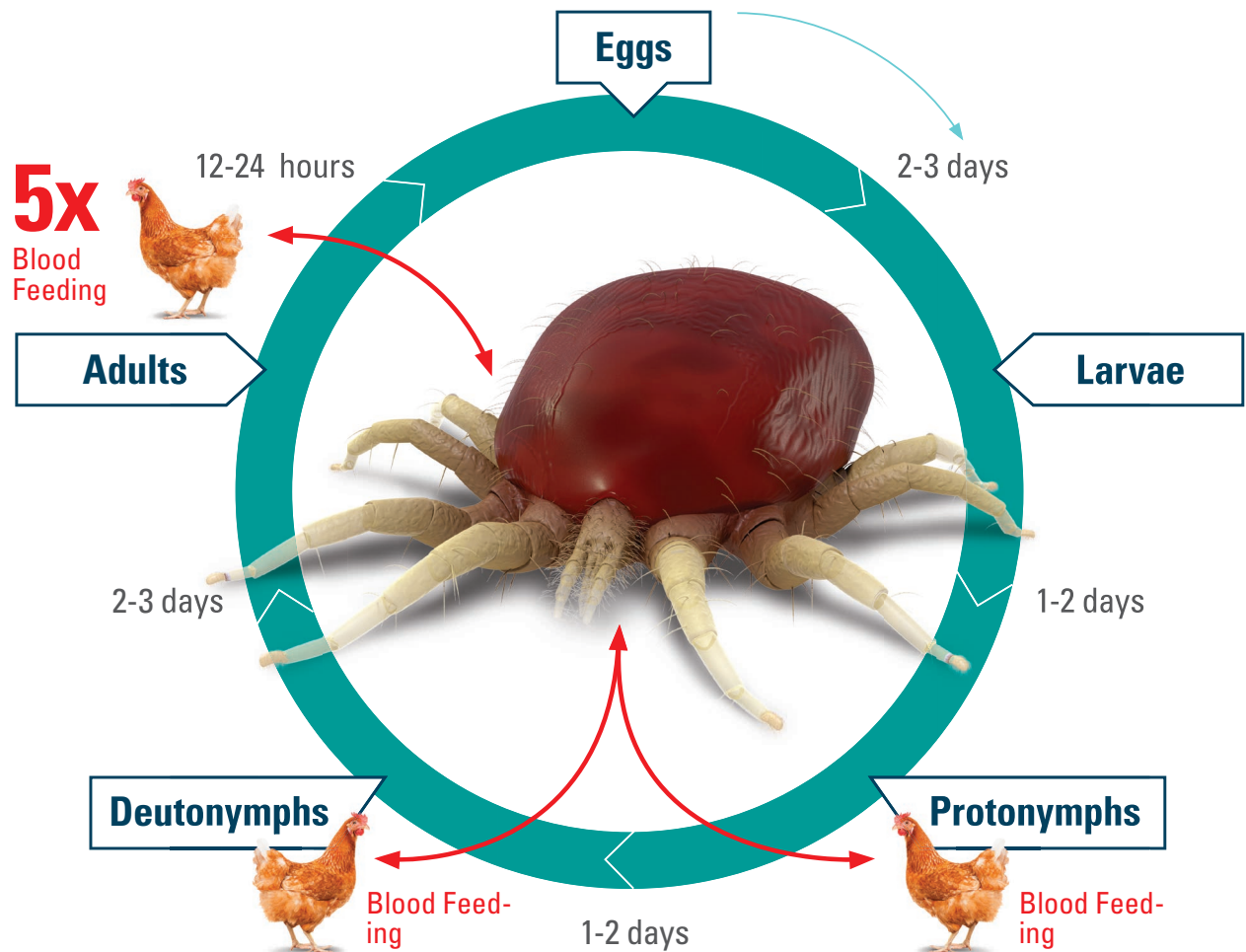
Figure 1-1: Poultry Red Mites on a piece of straw at a free-range farm..

HIGH PREVALENCE

Recent surveys have confirmed the extremely high and increasing prevalence of infestations with Poultry Red Mites in Europe. The average overall infestation rate of European layer houses has been estimated at 83%, with rates of 94% reported for Germany, Belgium, and the Netherlands, and 67% to 90% for France, Spain, Italy, Poland, Sweden, and the UK.⁷⁻⁹ Extrapolation of these estimates suggests that about 300 million hens in Europe are potentially suffering from mite infestations at any particular point in time. Poultry Red Mites are found in all production types, from backyard or organic farms to intensive, enriched cage or barn systems.¹⁰

Though only very limited information has been published in the scientific literature regarding red mite prevalence outside Europe, highly variable values have been reported for Africa, the Middle East, Asia, and Latin America. Poultry Red Mites are known to co-exist with Northern Fowl Mites in Latin America and Asia.

POULTRY RED MITE LIFE CYCLE



Based on Sparagano, et al.¹⁰

Figure 1-2: The life cycle of the Poultry Red Mite, *Dermanyssus gallinae*. Aside from the egg, the Poultry Red Mites have 4 life cycle stages: larva, protonymph, deutonymph, and adult. Larvae hatch with 6 legs and do not feed. After the first molt, both nymphal stages and adults have 8 legs. Protonymphs, deutonymphs, and adult females routinely feed on host blood, but males only occasionally feed.¹

PHYSIOLOGICAL DAMAGE

Infested chickens can develop anemia due to repeated mite bites, with laying hens possibly losing more than 3% of their blood volume every night.⁶ In extreme cases, *D. gallinae* infestation burdens may be so high that hens become severely anemic and die from blood loss alone.¹¹⁻¹² A 10-fold increase in hen mortality following severe infestation has been reported.¹¹

The direct mite life cycle can be as short as 7 days (Figure 1-2), which allows for rapid growth of mite populations. Heavy infestations of around 50,000 mites/hen in caged systems and up to 500,000 mites/hen in severe cases have been reported.² Typical mite density ranges between 25,000 and 50,000 mites/hen, levels that can induce aggressive feather-pecking and cannibalistic behavior, changes in feed and water intake, and decrease general condition.^{1,6,12,13} The most severe infestations occur in the warm season, from May to October.¹⁴

Mite bites are painful and induce skin irritation, contributing to high stress levels in infested birds. Increased self-grooming and head scratching both day and night (characteristic symptoms of anxiety that also contribute to decreased weight gain), have been observed in artificially infested hens.¹² Mite infestation has been associated with a 1.5-fold increase in corticosterone blood levels and a 22% decrease in β -globulin levels, indicative of somatic stress and immunosuppression. The adrenaline levels were also more than twice as high as in control animals, indicating psychogenic stress.¹⁷

In addition to experiencing anemia as well as higher rates of mortality and disease susceptibility, flocks infested with mites typically suffer eroded productivity as evidenced by:

- ▶ Decreased feed intake
- ▶ Decreased egg production
- ▶ Decreased egg quality (shell thinning, spotting)
- ▶ Decreased weight gain



Dermanyssus gallinae can also be involved in the transmission of numerous poultry pathogens, including zoonotic pathogens such as *Salmonella enteritidis* (responsible for one of the most widespread zoonoses worldwide).^{13,18-20} Mites have also been implicated as a vectors for numerous other poultry pathogens and avian influenza virus.^{10,21}

ECONOMIC DAMAGE

Economic losses from poultry mite infestations severely affect the productivity of the egg industry. Consequences of red mite infestation in a layer operation include primarily a negative impact on feed conversion ratio, a drop in egg production, an increase of downgraded eggs, and more susceptibility to poultry diseases.^{1,6,22} In 2017, Van Emous estimated that damage caused by mites in Europe is about € 231 million. This value is almost twice his 2005 estimation. This evaluation is based on a current total cost of red mite infestation of €0.60 per hen per year in the Netherlands.²³

A 2013-2014 FAO poultry census estimates the number of layer chickens in the 17 largest egg-producing countries in Europe to be 431 million.²⁴ Recently a large layer genetics supplier estimated that productivity losses can reach €0.57 per hen per year in case of moderate mite infestation (range €0.27 for low infestation to €2.50 for high infestation).²⁵ The estimation of the evolution of direct treatment costs is complex due to the changes in the acaricidal treatments arsenal available to the farmers for the past decades, as described later in this review. However, the same layer genetics supplier recently estimated the total cost of *D. gallinae* infestation to be up to €1 per laying hen per year, depending on the housing system, the infestation intensity, and the control methods used.²⁶ Based on the above recent poultry census data,²⁴ the updated average infestation prevalence (83%⁷), and recent per bird cost calculations,²⁶ we now estimate the current cost of red mite infestation for the egg industry in Europe to be about €200 million for productivity losses and up to about €360 million for overall costs.

POULTRY RED MITES AS VECTORS OF DISEASE

In addition to their effects on poultry production, Poultry Red Mites can spread an array of diseases of humans and other animals.^{1,8,19} Many bacterial and viral pathogens that affect both humans and animals and have been either isolated from red mites or had mite-vectoring transmission demonstrated in laboratory settings, including:

- ▶ *Salmonella gallinarum* and *enteritidis*
- ▶ *Pasteurella multocida*
- ▶ *Escherichia coli*
- ▶ *Listeria monocytogenes*
- ▶ *Mycoplasma synoviae*
- ▶ *Erysipelothrix rhusiopathiae*
- ▶ *Coxiella burnetti*
- ▶ *Nocardia brasiliensis*
- ▶ *Borrelia burgdorferi*
- ▶ Viral diseases such as Newcastle disease, St. Louis encephalitis, avian influenza, Eastern, Western, and Venezuelan equine encephalitis^{1,8,18,19,21,27,28}

HUMAN HEALTH CONCERNS

Poultry Red Mite pests are of growing concern in human medicine as *D. gallinae* infestation is increasingly responsible for human dermatological lesions (gamasoidosis), particularly in people living or working in close proximity to poultry.²⁸ In fact, red mites in poultry production buildings are recognized as a significant occupational hazard to poultry workers.^{29,30} A recent survey reported an increasing incidence of gamasoidosis worldwide and found that the disease is underdiagnosed.⁸ The survey showed that gamasoidosis severity is exacerbated by the persistence of mite infestation, treatment failures, and the potential transmission of zoonotic diseases by the mites (such as *Borrelia burgdorferi*, the cause of Lyme disease; *Babesia* spp.; *Bartonella* spp.). Dermatitis related to *D. gallinae* has become so substantial that workers in some countries have to be paid 3-times the basic rate to work with *D. gallinae*-infested birds.²²



Dermanyssus gallinae
Adult Poultry Red Mite

CONTROL ATTEMPTS

Current methods for managing red mites lack sufficient efficacy to keep infestations under control in many poultry farms.²⁶ The parasites are difficult to control since they can be transferred between flocks by crates, clothing, and wild birds, and they can be difficult to detect unless birds are examined at night when mites are feeding.³¹

Simply removing the host from an area will not eliminate the mites, as deutonymphs and adults are known to resist desiccation and live as long as 8 months without feeding.^{1,4} Furthermore, recent European legislation (2012) banning use of traditional cages for poultry birds (European Council Directive 1999/74/EC) has favored the use of housing systems incorporating more complex environments. While these animal welfare measures represent positive advancements for poultry husbandry, such housing systems appear to favor red mite proliferation and exacerbate infestation problems by providing mites more hiding possibilities, thus enabling them to more easily escape control measures.^{22,26,32,33}

Chemical control of mites is often attempted, involving treatment/spraying of the local environment including walls, floors, roosts, nest boxes, and birds. Synthetic acaricides such as organophosphates (dichlorvos, fenitrothion, chlorpyrifos, diazinon), carbamates (carbaryl, methomyl, propoxur), and pyrethroids (cyhalothrin) are the most common chemicals used against mite infestations, though their efficacy is questionable. Many conventional mite products have been withdrawn from European markets or banned in the past few years because they did not comply with European or national regulatory requirements with regard to human consumer and user safety.

Currently, the organophosphate phoxim is the only veterinary medicinal product registered in Europe for the treatment of *D. gallinae* infestations (since 2010). However, the product is not currently licensed in the largest layer industry countries such as Germany, Poland, Spain, and the UK, where the prevalence of *D. gallinae* infestations exceeds 80%. Furthermore, strict precautions pertaining to animal safety, user safety, and consumer safety must be observed when using phoxim:

- ▶ Although it may be applied in the presence of birds in an infested house, it should not be sprayed directly onto the birds, thus decreasing its ability to reach mite refuges in crevices and cracks close to the birds.
- ▶ The product is restricted only to those cases where its use is unavoidable because the infestation has become overwhelming.
- ▶ Users should wear special protective clothing during treatment and cannot re-enter the treated house for at least 12 hours after treatment.
- ▶ A 12-hour egg withdrawal period must be observed after treatment (makes product unsuitable for use in large, intensive layer farms).

Similar precautions apply for most other premise-spray/pesticide approaches to red mite control. Several acaricidal spray products are available in some European countries for the treatment of the poultry house and equipment (not birds), mainly for use during the unoccupied period between flocks. These products may contain pyrethroids (cypermethrin, permethrin, deltamethrin), carbamates (bendiocarb), abamectin, and spinosad. Most may only be applied in empty buildings; only a few (e.g., spinosad and cypermethrin) may be applied in the presence of birds in some countries. However, some of these products have no approved recommended egg withdrawal time, thus posing a potential human food-safety risk. In addition, currently marketed acaricidal products have only short residual activity,³⁴ which is a problem since *D. gallinae* mites may not encounter treated surfaces for several days post-application; and these products exert little or no activity on mite eggs, thus enabling regrowth of infestation burdens in treated houses. The application of pesticides to an entire poultry house also carries the risk of feed contamination, and treatments often need to be repeated at 2-week intervals. Thus, the use of chemical treatments involves the direct cost of the pesticides used plus labor costs, worker protection equipment, and the cost of maintaining the flock through the egg and meat withdrawal times following treatment.

Chemical treatments are also hampered by resistance development to multiple acaricides^{1,35} due to selection of resistant mites that survive due to uneven spraying, especially inside crevices and cracks resulting in exposure to sublethal concentrations.³⁶ Resistance to organophosphates has been reported in the literature for several decades³⁶ and is now widespread. For instance, 10 *D. gallinae* field isolates from German laying farms were all resistant to the pyrethroid cyfluthrin and 90% to the organophosphate phoxim.³⁷

Because of the many problems associated with use of conventional chemicals and pesticides for controlling Poultry Red Mites, several new alternative solutions have been proposed or developed in recent years, including essential oils, newer chemical pesticides (e.g., spinosad), predator mites, heat treatments, intermittent lighting programs, inert dusts (e.g., silica, diatomaceous earth), and even vaccines.²⁶

THE UNMET NEED FOR A NEW MITE CONTROL OPTION

In spite of many previous efforts, the development of more useful, effective, and innovative treatments to keep Poultry Red Mite infestations under control remains in great demand.²⁶ This unmet medical need has clearly been recognized by the scientific community, key opinion leader groups, the layer industry, and government organizations. The absence of an effective treatment is stated in most of the scientific papers dealing with poultry mites and has been officially recognized by the EU Commission, which created and funds the COST Action FA 1404 ("Improving current understanding and research for sustainable control of the Poultry Red Mite *Dermanyssus gallinae*," COREMI³²) to "rid laying hens of a common disease-spreading pest." This 4-year project started in late 2014 involves representatives from almost all EU countries, Turkey, and Israel.

NORTHERN FOWL MITES¹⁶

The Northern Fowl Mite (*Ornithonyssus sylviarum*, Figure 1-3) is another mite species that poses a severe threat to poultry production, particularly in North America, Brazil, China, and Australia. Fowl mites are the most common and damaging ecto-parasite of poultry in the US, though Poultry Red Mites (*D. gallinae*) can also afflict US flocks. Fowl mites also occur in Europe and may pose an emerging threat but are not comparable there to the damage caused by Poultry Red Mites (the reasons for this disparity are unclear).

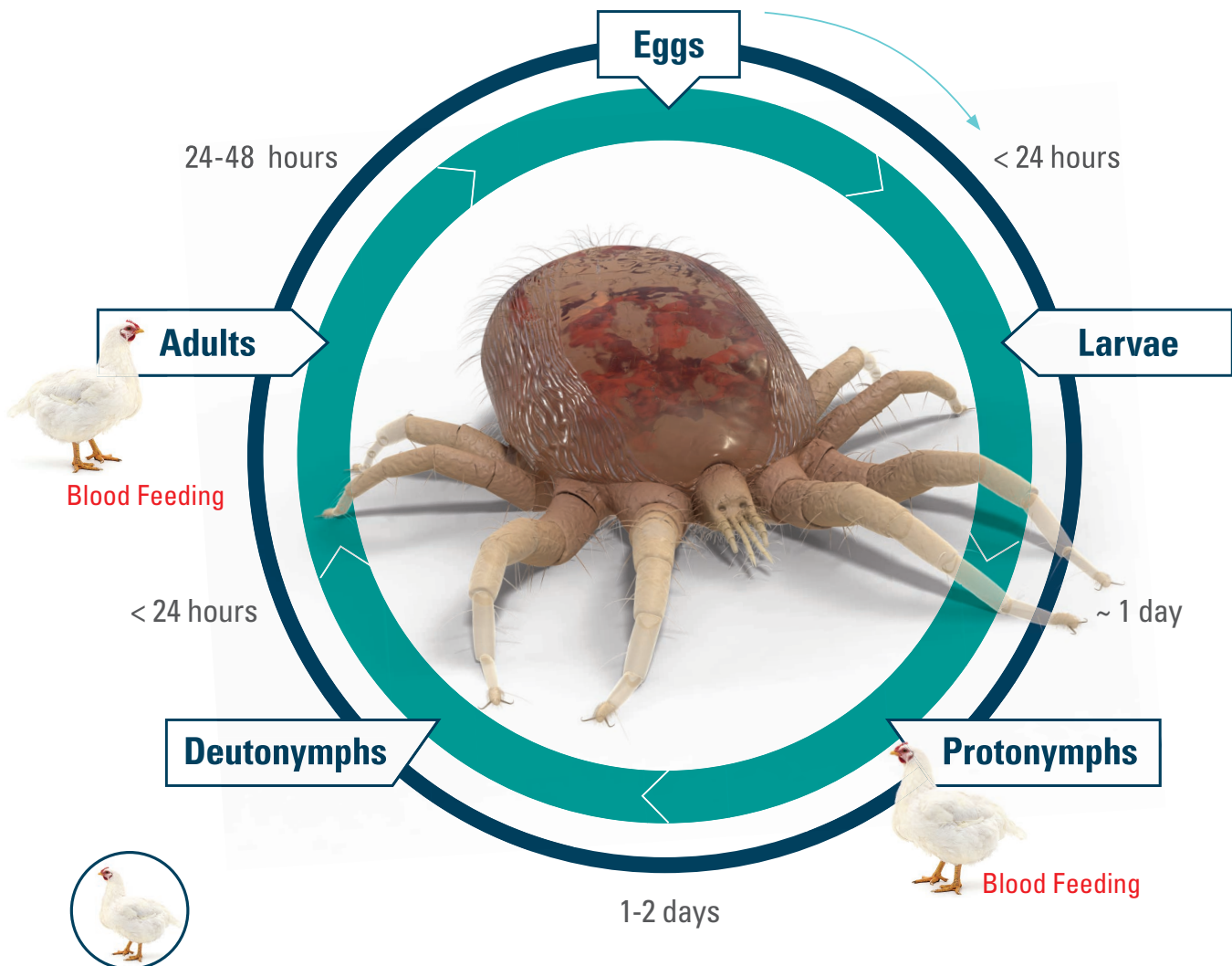
NORTHERN FOWL MITES

Kingdom ...*Animalia*
Phylum*Arthropoda*
Class*Arachnida*
Subclass.....*Acari*
Order*Mesostigmata*
Family *Macronyssidae*
Genus.....*Ornithonyssus*
Species.....*O. sylviarum*

Fowl mites are closely related to Poultry Red Mites and induce similar consequences in terms of animal welfare and production losses. Like Poultry Red Mites, *O. sylviarum* is considered a "dermanyssoid" mite but is placed in a different taxonomic family (Macronyssidae rather than the Dermanyssidae). Though originally a nest-parasite of wild birds, these obligate blood-

feeding mites have become common ectoparasites of domestic birds, causing direct damage to poultry and decreased economic output. Severe infestations and economic damage occur primarily in layer hens (chickens) or breeders (chickens or turkeys), which are raised for longer periods of time than other poultry types (such as broilers) and thus allow high mite populations to develop and be maintained.

NORTHERN FOWL MITE LIFE CYCLE



Entire Northern Fowl Mite life cycle exists on bird.

Figure 1-3: The life cycle of the Northern Fowl Mite, *Ornithonyssus sylviarum*. Aside from the egg, fowl mites have 4 life-cycle stages: larva, protonymph, deutonymph, and adult. Larvae hatch with 6 legs and do not feed. After the first molt, both nymphal stages and adults have 8 legs. Only protonymphs and adult females feed on host blood. All stages remain on the chicken (unlike daily exodus of red mites off the bird). Eggs mature to adults in as few as 5 days.



Figure 1-4: Photomicrograph of Northern Fowl Mite adult female, *Ornithonyssus sylviarum* (100 $\times$).¹⁶ (Used with permission.)



Figure 1-5: Photomicrograph of life stages of the Northern Fowl Mite: egg (e), egg with developing larva (e-l), larvae (l), protonymph (p), and adult female.¹⁶ (Used with permission.)

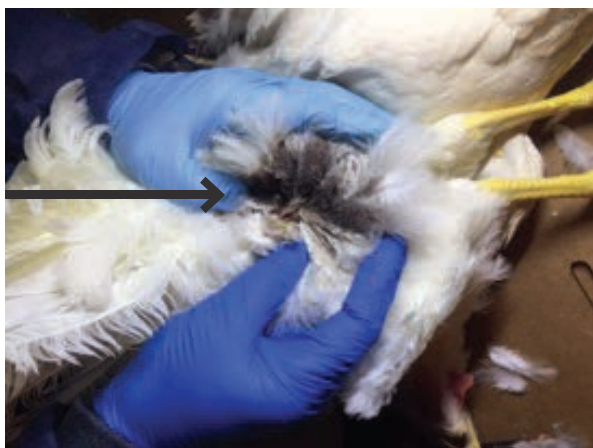


Figure 1-6: Northern Fowl Mites (primarily adults) blood-feeding at the base of an infested vent feather (arrow indicates area of concentrated mite feeding).

LIFE CYCLE, POPULATION DYNAMICS

Populations of Northern Fowl Mites are predominated by females (4:1 ratio), and even unfertilized eggs can produce male offspring available for future mating. Thus, introduction of a single unmated female, even if immature, may be able to trigger infestation of a new host.

A major difference between Northern Fowl Mites and Poultry Red Mites is that the entire *O. sylviarum* life cycle is completed on birds. Thus, chickens are infested with all *O. sylviarum* development stages (Figure 1-4), with the mites perpetually residing on host feathers (usually in the vent area) and migrating to the skin surface where only protonymphs and adults feed on blood (no daily exodus off the bird as with red mites). In addition, adult *O. sylviarum* are generally smaller (~0.6 mm) than *D. gallinae* adults (~1-1.5 mm).

The direct fowl mite life cycle requires 5 to 12 days to complete (Figure 1-5). Because the life cycle can be as short as 5 days under optimal conditions, rapid growth of mite populations can occur. Fowl mite eggs are laid primarily onto feathers of the vent region just anterior to the cloaca of an infested chicken, offering an optimum feather structure and microclimate favored by mites. Fowl mites are able to survive off a host for periods of 1 to 3 weeks. Protonymphs survive much better than adults, and survival of both stages improves at lower temperature and higher humidity.

Large numbers of fowl mites are readily visible on infested birds, with feathers typically appearing “dirty” because of the collection of mites, cast skins, eggs, and mite feces (Figure 1-6). Dense clusters of these components and matted feathers “glued” together by mite debris can create a semi-porous microhabitat that may be quite thick.

Northern Fowl Mites easily spread among caged hens and exhibit rapid and steady population growth when introduced to naïve chickens. On a flock basis, fowl mite infestations grow steadily for 4 to 10 weeks after introduction, while populations on individual birds usually peak 3 to 6 weeks post-introduction. Low mite levels then persist for the remainder of the bird’s life, and this decline in mite population is likely due to bird immune responses. However, older hens may experience moderate mite ‘relapses’ at intervals of about 5 to 6 weeks.



Figure 1-7: Adult Northern Fowl Mites on a chicken egg. Mites may be visible on eggs when populations are dense and may be used as a monitoring tool to gauge relative mite populations.

In severe cases, mite numbers can well exceed 50,000 and sometimes even 100,000 mites per bird. At such high infestation levels, mites can cause up to 6% blood loss per day per hen.³⁸ Mite populations vary substantially among birds of the same age and strain, with some of this variation under genetic control.

NORTHERN FOWL MITE IMPACTS

Blood-feeding by Northern Fowl Mites can cause skin irritation and inflammation (immune activation), anemia (up to 6% blood loss/day), and sometimes even death. Lesions associated with mite feeding have been identified as an animal welfare concern. The parasites are most damaging early in a new flock during the first egg-production cycle, when young hens are immunologically naïve to mites.

Resulting economic impacts due to fowl mite infestations include decreased egg production and feed conversion efficiency. One 2009 study reported profit reductions of \$0.07 to \$0.10/hen over only a 10-week peak mite-infestation period, which basically eroded all the expected profit margin at that time.³⁹ The damage appeared to depend on infestation level, as low rates (≤ 100 mites/bird) were not as economically damaging to egg production. Degradation of standard measures of egg quality (e.g., Haugh units, albumen height, egg weight, egg specific gravity) have also been documented over time as mite

populations increased. All of these negative impacts may also be related to mite-induced immune responses.

Like Poultry Red Mites, Northern Fowl Mites can also cause irritation to human workers. As mite populations expand and become very dense, mites may be seen on eggs (Figure 1-7), equipment, and poultry workers. In fact, visually monitoring of fowl mite numbers on eggs has been proposed as a relatively quick and easy monitoring method, though less exact than directly checking hens.

Unlike Poultry Red Mites, Northern Fowl Mites to date have not been implicated as important pathogen vectors (perhaps due to less research conducted compared to red mites). However, they may serve as a minor mechanical vector of western equine encephalitis and may play a role as a reservoir for poultry pathogens between flocks (especially if mites are ingested directly by birds while grooming).¹⁶

CONTROL ATTEMPTS

Successful integrated pest management programs for poultry should focus first on preventative measures and then, if infestation occurs, strive to keep the population below economic thresholds. Once Northern Fowl Mites are established in a facility they can be very difficult to eliminate and may become a predictable problem in subsequent flocks. Conventional control attempts are similar to those used for Poultry Red Mites, such as chemical methods (e.g., pesticide sprays) and a variety of alternative strategies discussed earlier (and other alternatives, like dust bags and encouragement of dust bathing and grooming behaviors). However, control efficiency is threatened by mite resistance to a shrinking arsenal of acaricidal compound classes, especially for the synthetic pyrethroids, but also against carbamates and organophosphates.⁴⁰

Applications of materials intended for Poultry Red Mite control are typically off-host (applied to mite resting areas, facility, etc.) while products for Northern Fowl Mites somehow need to be applied on the bird for effective control, thus complicating both practicality and host safety. On most large-scale production operations in the US, Northern Fowl Mites are typically managed by spraying chemical pesticides at high pressure directly

onto birds, with the spray directed upwards from underneath hanging 'battery' cages (pesticides are most effective when applied to feathers in the vent region where mites gather). However, the US industry is gradually transitioning away from using small wire battery cages for egg production and switching instead to larger 'enriched' cages or cage-free housing. Effective spraying of birds with pesticides is more difficult in these systems and thus severely limits some methods for mite control.

Clearly, an urgent need exists to develop new or alternative control methods for fowl mites because the number of available synthetic control agents is decreasing, concerns about pesticide

exposure are increasing, organic production is increasing (which disallows most pesticides), and poultry housing is changing.

THE NEW APPROACH

The development and approval of Exzolt (fluralaner) solution for use in drinking water now offers an innovative new approach for comprehensive management of Poultry Red Mites and Northern Fowl Mites in chickens. As discussed in the following pages, novel Exzolt provides a unique combination of potent efficacy, user convenience, and bird safety in a ready-to-use aqueous solution for water administration.

MITE PARASITES SUMMARY

- ▶ External mite parasites are a costly global problem for poultry production, with an estimated 83% average overall infestation rate for European layer houses.
- ▶ Poultry Red Mites (*Dermanyssus gallinae*) are the primary mite threat in Europe, while Northern Fowl Mites (*Ornithonyssus sylviarum*) predominate in other regions such as the US and Latin America.
- ▶ The approximate total cost of Poultry Red Mite infestations and overall costs totals €360 million in Europe, and the parasites can spread a diversity of bacterial and viral diseases of humans and other animals.
- ▶ Poultry Red Mites can spread a diversity of bacterial and viral diseases of humans and other animals.
- ▶ Current methods for managing Poultry Red Mites lack sufficient efficacy to keep infestations under control at many poultry farms, and often pose safety threats for both birds and humans.
- ▶ The development of more useful, effective, and innovative treatments to control mite infestations has been an unmet medical need recognized by the scientific community, key opinion leader groups, the layer industry, and governmental authorities.

SECTION TWO

PRODUCT PROFILE





PRODUCT PROFILE

Exzolt is a unique parasiticide for chickens that provides potent efficacy against Poultry Red Mites and Northern Fowl Mites via a convenient oral solution dosage form for administration in the drinking water. Fluralaner (carbamoyl-benzamide-phenyl-isoxazoline), the active ingredient of Exzolt, is a member of the novel antiparasitic compound class of isoxazoline-substituted benzamide derivatives. It is a racemic mixture of S- and R-enantiomers, with the S-enantiomer expressing activity (equal amounts of left- and right-handed enantiomers of the chiral molecule). Exzolt is a 1% fluralaner aqueous solution (10 mg/mL) intended for treatment and control of Poultry Red Mite (*D. gallinae*) and Northern Fowl Mite (*O. sylviarum*) infestation in chickens (pullets, breeders, and layers) when administered orally via drinking water twice (7 days apart) at a dose of 0.5 mg fluralaner per kg body weight (BW) (equivalent to 0.05 mL Exzolt/kg BW).

Fluralaner is a compound from a new chemical class and is the first isoxazoline approved for use in poultry. This development is notable because very few new pharmaceutical molecules have been licensed over the past few years for use in poultry in Europe. In fact, the high economic pressures and low margins inherent to egg and chicken meat production typically mean the poultry industry can “afford” only older compounds or agents widely used in the crop or premise-protection domain. In contrast, fluralaner has never been used in agriculture, and therefore represents a truly new and innovative treatment for poultry.

PHARMACOLOGY

After oral administration, fluralaner is absorbed rapidly from the medicated drinking water, reaching maximum plasma concentrations 36 hours after the first administration and 12 hours after the second one, following start of administration via medicated drinking water. The bioavailability is high, with approximately 91% of the dose absorbed following oral administration. Fluralaner is highly bound to protein. Fluralaner is widely distributed throughout the body, with the highest concentrations reported in the liver and skin/fat. No significant metabolites are observed in chickens, and fluralaner is mainly eliminated via the hepatic route. The apparent elimination half-life is approximately 5 days following oral administration.

MODE OF ACTION

Once ingested by a mite feeding on a treated chicken, fluralaner acts as a potent inhibitor of parts of the arthropod nervous system by acting antagonistically on ligand-gated chloride channels (GABA-receptor and glutamate-receptor). The compound has demonstrated high activity on various heterologous GABA-receptors (*Rhipicephalus microplus*, *Ctenocephalides felis*, *Drosophila melanogaster*), but shows no activity on the tested mammalian GABA-receptor.⁴¹ Research also showed that the fluralaner activity was not affected by the dieldrin resistance (RDL) using the RDL-GABA-receptor variants from *C. felis* and *D. melanogaster* (these RDL-GABA-receptor variants exhibit resistance against dieldrin and

partly against fipronil). In vitro bio-assays have shown that fluralaner is effective against parasites having proven field resistance, including organophosphates (tick, mite), pyrethroids (tick, mite), and carbamates (mite).⁴²

FAST, POTENT EFFICACY

Treatment of the host with Exzolt (instead of treating the host environment) is an innovative approach to targeting mite parasites that live on the premises and spend only 30 to 90 minutes/day on the host.⁶ Convenient treatment of poultry with Exzolt via the drinking water causes effective levels of the acaricide to be systemically distributed within all birds, ready to kill mites whenever parasites extract a blood meal from their hosts. Efficacy against *D. gallinae* begins within 4 hours after exposure of mites to treated chickens, and any mites feeding on treated chickens for at least 2 weeks after the first Exzolt administration will be killed. In addition, non-blood-feeding stages (eggs, larvae) are killed as soon as they develop into hematophagous forms, and egg production by female mites is stopped. Thus, the mite life cycle is disrupted due to the rapid onset of fluralaner activity, the very high mite-killing efficacy (duration of at least 2 mite life cycles), and the absence of egg production from female mites exposed to treated chickens. Exzolt also contributes toward long-term control of mite populations in a poultry house when used in conjunction with implementation of proper biosecurity measures.

Available data indicate that Exzolt is more effective than traditional spray products for rapidly and dramatically decreasing building infestation burdens. Furthermore, regrowth of mite populations takes longer than with spray treatments, indicating that more mites are eliminated by systemic treatment with Exzolt.

SAFE FOR CHICKENS AND PEOPLE

Fluralaner offers high selectivity for arthropods like poultry mites, and the formulated product has a large safety margin demonstrated in chickens. Unlike spraying, water-borne Exzolt is not stressful for hens and minimizes the potential for exposure of chemicals by human workers. Furthermore, Exzolt has a zero-day withdrawal period for eggs and is safe for layers, which is essential for large poultry units. Treatment compliance is also enhanced by convenient administration of the ready-to-use solution in drinking water, greatly reducing the workload for house workers compared to spraying (e.g., removing birds and/or eggs, repeat applications, quarantines, less safety equipment, application license requirements, fewer safety precautions, etc.).

BENEFITS OF EXZOLT

Dose rate: 0.5 mg fluralaner/kg BW administered twice, 7 days apart

POTENT EFFICACY:

- ▶ Induces a rapid and massive decrease in mite populations in a chicken house.
- ▶ Treats the bird, not just the environment.
- ▶ Fast start of kill (within 4 hours for Poultry Red Mite), with prolonged duration in birds spanning 2 mite life cycles.
- ▶ High bioavailability, systemic distribution in poultry.
- ▶ Active against mites resistant to classical acaricides.

WATER TREATMENT:

- ▶ Treats all birds, unlike hit-and-miss spraying.
- ▶ More convenient and cost-effective than sprays.
- ▶ Uniform dosing accuracy.
- ▶ 1 treatment—2 administrations, 1 week apart.
- ▶ Ready-to-use solution for simple dilution, with no sedimentation or clogging.
- ▶ Flexible; easy to tailor treatment to bird management programs (feeding, housing, etc.).
- ▶ Reduced labor compared to sprays.
- ▶ Re-sealable, multiple-entry container with an in-use shelf-life of 1 year.

SAFETY ASSURED:

- ▶ Zero egg withdrawal time for layers.
- ▶ Not stressful for hens, safe for breeders, no harm on egg production, hatchability, or progeny.
- ▶ Broad safety margin.
- ▶ Avoids exposure of house workers and birds to toxic chemical sprays.

NOVEL, REVOLUTIONARY EXZOLT

Exzolt provides poultry producers a revolutionary new option for managing Poultry Red Mites and Northern Fowl Mites that is effective, safe, convenient, and cost-effective, thus helping chickens maintain optimal productivity performance unhindered by profit-suppressing ectoparasites. Exzolt sets a new standard for management of mites and represents an excellent therapeutic component of comprehensive health programs aimed at optimizing overall performance of pullets, breeders, and layers.



EXZOLT LABEL INFORMATION

INGREDIENTS

Exzolt contains 10 mg fluralaner per mL of solution for use in drinking water for chickens. Excipient ingredients include α -tocopherol, diethylene glycol monoethyl ether, and polysorbate 80. It is a light yellow to dark yellow solution for use in drinking water.

INDICATIONS

Treatment and control of Poultry Red Mite (*Dermanyssus gallinae*) and Northern Fowl Mite (*Ornithonyssus sylviarum*) infestation in pullets, breeders, and layer hens. (Note: not approved for Northern Fowl Mites in the EU.)

DOSAGE AND ADMINISTRATION

For use in drinking water.

Dose: 0.5 mg fluralaner per kg BW (equivalent to 0.05 mL Exzolt) administered twice, 7 days apart. The complete course of treatment must be administered for a full therapeutic effect. If another course of treatment is indicated, the interval between two courses of treatment should be at least 3 months.

Determine the duration of time (between 4 and 24 hours) over which to administer the medicated water on the treatment day. This period of time must be long enough to allow all the birds to receive the required dose. Estimate how much water birds will consume during treatment based on the previous day's water consumption. The product should be added to a volume of water that the chickens will consume in one day. No other source of drinking water should be available during the medication period.

Calculate the volume of product needed based on the total weight of all birds in the house to be treated. To ensure administration of the correct dose, the body weight should be determined as accurately as possible and an accurate measuring device should be used for measuring the calculated volume of the product to be administered.

The required amount of product on each treatment day is calculated from the total BW (kg) of the entire group of chickens to be treated, using the following formula:

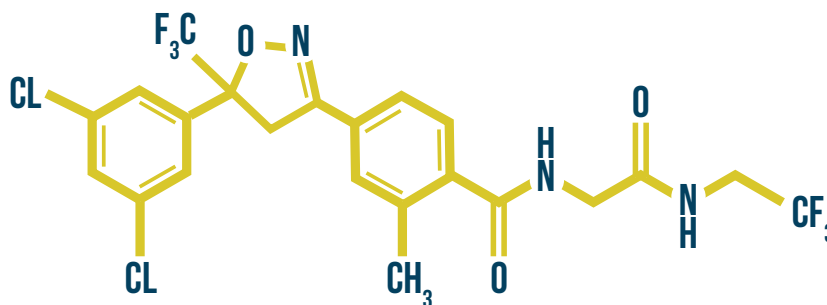
$$\text{volume of product (mL)/treatment day} = \text{Total estimated BW (kg) of chickens to be treated} \times 0.05 \text{ mL}$$

Therefore 500 mL of product treats 10,000 kg body weight (e.g., 5,000 chickens of 2 kg body weight each) per day of treatment administration.

Follow instructions on the full package leaflet to prepare medicated water.

FLURALANER CHEMICAL PROPERTIES

GENERIC NAME:	Fluralaner
CHEMICAL NAME:	(±)-4-[5(RS)-(3,5-dichlorophenyl)-5-(trifluoromethyl)-4,5-3-yl]-2-methyl-N-[2-oxo-2-(2,2,2-trifluoroethylamino) ethyl]benzamide
MOLECULAR FORMULA:	$C_{22}H_{17}Cl_2F_6N_3O_3$
MOLECULAR WEIGHT:	556.3
PHARMACOTHERAPEUTIC GROUP:	Ectoparasiticides for systemic use
ATCVET CODE:	QP53BE02



WITHDRAWAL PERIODS

Eggs: zero (0) days
Meat and offal: 13 days

breeding chickens were treated at 3-times the recommended dose for 2-times the recommended duration of treatment.

CONTRAINDICATIONS, ADVERSE REACTIONS, INTERACTIONS

None known.

In the absence of compatibility studies, Exzolt must not be mixed with other veterinary medicinal products.

ANIMAL PRECAUTIONS

Strict biosecurity measures at house and farm level should be implemented to prevent re-infestation of treated houses. To ensure long term control of the mite populations in a treated house, it is essential to treat any other infested poultry in houses in proximity to the treated one.

SAFETY, OVERDOSE

Safety has been demonstrated in layers and breeders, and product can be used during lay.

No adverse reactions/effects observed following treatment of 3-week old chickens, adult chickens, or laying hens (egg production) dosed at up to 5-times the recommended dose for 3-times the recommended duration of treatment. No adverse effects on reproductive performance when

USER PRECAUTIONS

The veterinary medical product may be slightly irritating to skin and/or eyes. Avoid contact with skin, eyes and mucous membranes. Do not eat, drink or smoke while handling the product. Wash hands and contacted skin with soap and water after use of the product. In case of eye contact, immediately rinse thoroughly with water. If the product is spilled, remove any affected clothes.

PACKAGING/STORAGE/EXPIRATION

Packaged in 1-L and 4-L bottles (HDPE) closed with an aluminum foil seal and a child-resistant polypropylene screw cap.

Do not store above 30° C.

Shelf life as packaged: 3 years

Shelf life after opening the immediate packaging:
12 months

Shelf life of medicated drinking water: 24 hours



PRODUCT PROFILE SUMMARY

- ▶ Exzolt (fluralaner) represents a completely new approach for comprehensive management of Poultry Red Mites and Northern Fowl Mites in chickens by systemically treating host birds (via drinking water) instead of treating the animal facilities.
- ▶ Fluralaner is a potent inhibitor of parts of the arthropod nervous system and acts antagonistically on ligand-gated chloride channels (GABA-receptor and glutamate-receptor).
- ▶ Treatment with Exzolt provides fast, convenient, and potent acaricidal efficacy, with proven safety for chickens and the users of the product.
- ▶ Exzolt is approved for treatment of Poultry Red Mite (*Dermanyssus gallinae*) and Northern Fowl Mite (*Ornithonyssus sylviarum*) infestation in pullets, breeders, and layers when provided in drinking water at 0.5 mg fluralaner/kg BW, administered twice 7 days apart.
- ▶ Exzolt has a zero-day withdrawal for chicken eggs and a 14-day withdrawal period for meat and offal, with no contraindications, known adverse effects, or known interactions.

NOTES

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SECTION THREE

PHARMACOLOGY AND ACTIVITY





PHARMACOLOGY AND ACTIVITY

Because orally administered fluralaner reaches target ectoparasites via the gastrointestinal tract and the bloodstream, a review of the pharmacologic behavior of fluralaner within chickens is relevant. Several studies documented the pharmacokinetics and pharmacodynamics/activity of fluralaner in support of product indications. As mentioned earlier, fluralaner is a racemic mixture of S- and R-enantiomers (equal amounts of left- and right-handed enantiomers of the chiral molecule), with the S-enantiomer expressing acaricidal activity.



Comprehensive studies examined the label recommendations and fluralaner pharmacokinetic profile of Exzolt.

PHARMACOKINETICS

Research investigated the pharmacokinetic profile of fluralaner in chickens after intravenous administration of 0.5 mg/kg BW and after oral administration by gavage at 0.25, 0.5, and 1 mg/kg BW (4 treatment groups, 45 laying hens/treatment).⁴³ Blood samples were collected pre-treatment and at 17 post-treatment time-points (5 minutes through 35 days). Fluralaner displayed a slow elimination profile (T_{1/2} of about 5 days after intravenous administration), dose linearity, and 91% oral bioavailability. Fluralaner is highly bound (~100%) to plasma proteins.⁴⁴

The pivotal pharmacokinetic study examined use of Exzolt according to label recommendations and assessment of the fluralaner pharmacokinetic profile after oral administration via drinking water.⁴⁴

STUDY DESIGN

- ▶ 55 healthy laying hens were treated with 2 single administrations of Exzolt (2-times 0.5 mg fluralaner/kg BW) at 7-day intervals.
- ▶ Blood samples were collected from hens pre-treatment and at 20 time-points from 6 hours until 21 days post-treatment (4 sample-times/hen).
- ▶ Plasma was analyzed for fluralaner concentration using a validated HPLC-MS/MS chiral method.

RESULTS

Fluralaner was quickly absorbed (Figure 3-1) and concentrations peaked at the 1.5-day time-point after the first administration and 0.5 day after the second administration, with C_{max} of 323.7 ng/mL and 355.1 ng/mL after the respective first and second administrations, indicating slight accumulation. Concentrations of fluralaner then decreased gradually. The AUC was 1297 ng*day/mL (day 0-7) and 2388 ng*day/mL (day 7-infinity). Notably, the 2-administration fluralaner plasma profile covered the time-period needed for completion of 2 consecutive mite life cycles, a fundamental concept used for selecting the dose regimen with the intent of imposing lengthy disruption of mite population dynamics.

CONCLUSIONS

Fluralaner oral bioavailability is high and drug was rapidly absorbed after oral administration in drinking water at 0.5 mg/kg BW twice at 7-day intervals, and slight accumulation occurred in blood, consistent with a rhythm of administration designed to maintain therapeutic efficacy between administrations. Appropriate levels were achieved and maintained in blood plasma for targeting of blood-feeding mites.

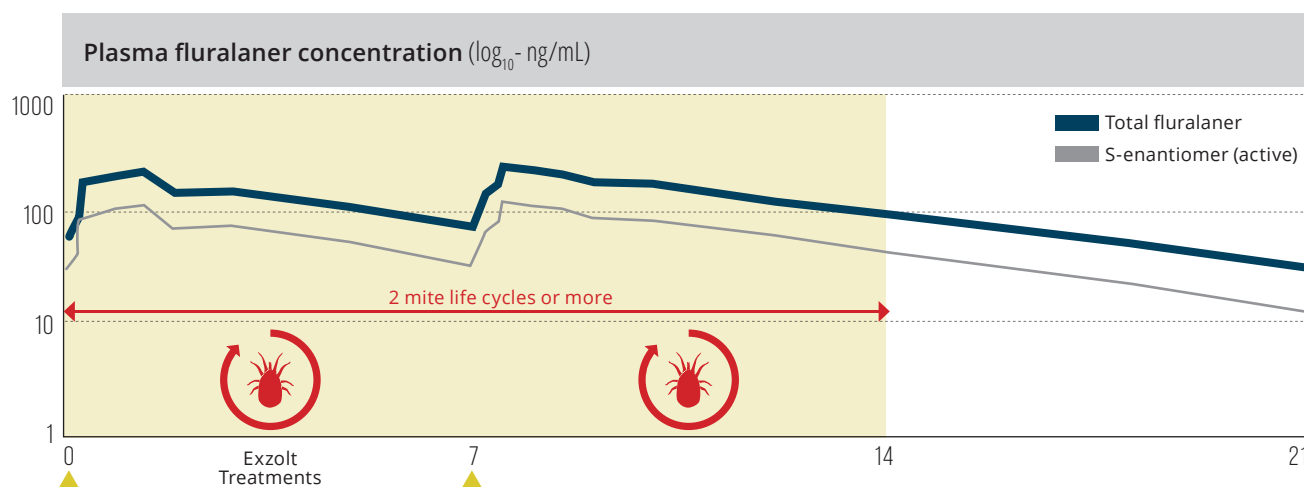


Figure 3-1: Fluralaner plasma concentration; time profile after Exzolt administration in chickens at 0.5 mg/kg BW in drinking water, twice at a 7-day interval.

PHARMACODYNAMICS AND ACTIVITY

In preliminary research regarding fluralaner pharmacodynamics and activity, fluralaner demonstrated a prominent feeding activity against *Dermanyssus gallinae* compared to a less potent contact activity, which is appropriate for an orally administered drug with a systemic mode of action (Figure 3-2).⁴³ Another study evaluated the susceptibility of nymph and adult mites to fluralaner, using an immersion test.⁴³ Fluralaner was highly active in vitro against both nymph ($LC_{90}=0.38$ ppm) and adult ($LC_{90}=0.52$ ppm) mite stages.

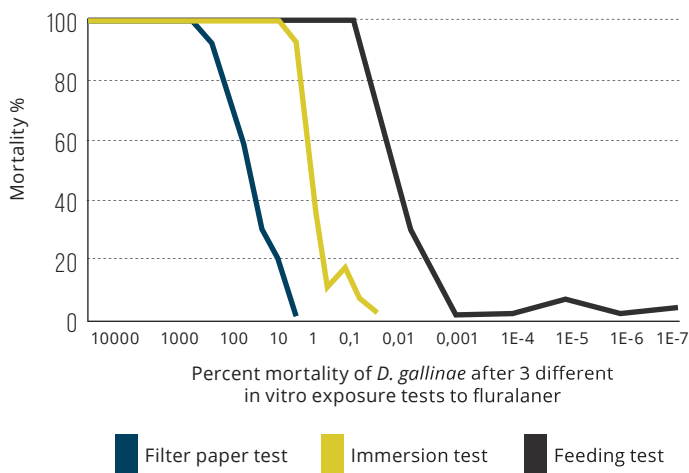


Figure 3-2: Percent mortality of *D. gallinae* after exposure to different fluralaner concentrations in 3 different tests.

The primary pharmacodynamic study of interest evaluated the speed of kill and the impact of treatment on reproductive capacity of mites after feeding on Exzolt-treated hens.⁴⁵

STUDY DESIGN

- ▶ 16 laying hens (~ 20 weeks of age) were randomly allocated to 2 groups of 8 hens housed in 2 separate pens. One group was treated on days 0 and 7 with Exzolt at the recommended dose of 0.5 mg fluralaner/kg BW while the second group remained unmedicated (controls).
- ▶ Six hens of each group were artificially infested with unfed *D. gallinae* (mainly adults) on multiple occasions between days 1 and 26. After each infestation, a sample of engorged mites were collected from each infested hen, incubated, and assessed for mortality after 4, 8, 12, and 24 hours.
- ▶ For the reproductive portion of the study, tubes containing mite eggs, surviving mites, or both were incubated for 8 additional days. Counts of dead adult male and female mites, mite eggs, larvae, and nymphs were performed daily.
- ▶ The primary reproduction parameter was the control of nymphal development (i.e., control of the capability to finally reproduce into nymphs as the first blood-sucking development stage). The conversion from each mite stage to the next was also determined, including the number of eggs laid per incubated female (fertility) and hatchability rate.



RESULTS

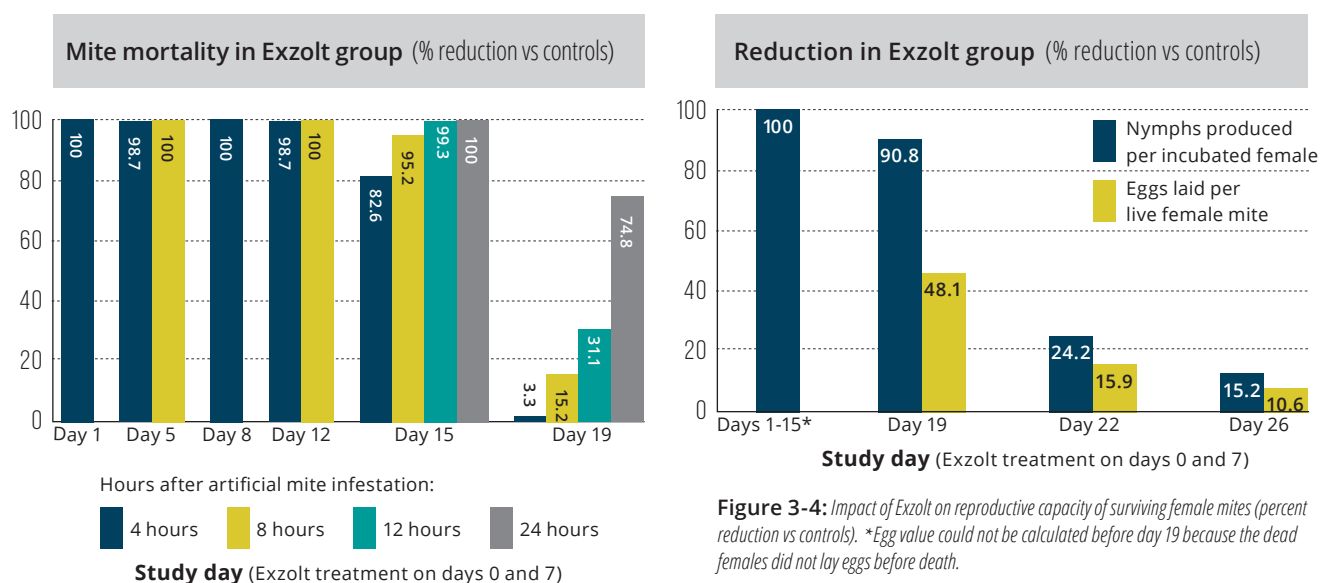
Outcomes for the speed-of-kill portion of the study (Figure 3-3) indicate that the mortality of mites infested on day 1 was 100% by 4 hours post-infestation while the mortality of mites infested on day 5 was 98.7% at 4 hours and 100% at 8 hours. After the second oral Exzolt treatment of hens on day 7, mortality of mites infested on day 8 was again 100% at 4 hours, and at day 12 near complete mite mortality (98.7-100%) was still observed within just 4 hours of mites feeding on treated hens. Notably, 100% mortality was observed within 24 hours for all mite infestations through day 15, without any oviposition. These impressive outcomes are related to the fact that mites feed on birds for a very short time (30-90 minutes), absorbing a high amount of fluralaner during this period. By day 19, mite mortality 24 hours post-infestation was 74.8% and the oviposition (eggs laid within 24 hours) was reduced 82.2% vs controls.

Nymph emergence (an indicator of reproductive success) was completely inhibited in treated hens through day 15, and reduction was still 90.8% at day 19 vs controls (Figure 3-4). Fertility of surviving females (control of eggs laid) was impaired mainly for mites infested on day 19 (48.1% reduction, Figure 3-4).

The oral administration of Exzolt was well tolerated in all hens, with no treatment-related adverse reactions observed in treated birds.

CONCLUSIONS

The treatment of hens with Exzolt led to profound and fast mite mortality for at least 15 days. Furthermore, the demonstrated fast onset of action resulted in an immediate interruption of the reproduction of the affected female mites prior to the lethal effect. Even 12 or 15 days after the second Exzolt treatment (days 19 and 22), the medication still demonstrated major impacts on the ability of surviving females to produce the next parasitic mite generation (nymph emergence). Ongoing mortality of incubated mites combined with a reduction of laid eggs are the main factors responsible for reduced reproduction of the mites (nymph emergence). Molting processes of the mite progeny were not notably impacted by fluralaner.



RESISTANCE

The development of resistance by mites to control agents is an ongoing concern, as with most all medicinal or chemical interventions used for animal health (e.g., antimicrobials, anthelmintics, insecticides, etc.). However, because fluralaner is a new agent recently introduced in veterinary medicine, resistance has not shown to be a concern and none has been detected. Isoxazolines like fluralaner act at previously unrecognized activity sites without cross-resistance to other chemotypes, and effects differ between insects and mammals so toxicity is selective.⁴⁶ In vitro bioassays have indicated that fluralaner offers efficacy against parasites with known field resistance to various chemical classes, including organophosphates (tick, mite), pyrethroids (tick, mite), and carbamates (mite).

During dose confirmation and field studies conducted in support of Exzolt approval, 12 Poultry Red Mite isolates were collected in 2014 and 2015 from 12 chicken houses in France, Germany, and Spain. The collected isolates were tested for in vitro acaricide sensitivity to fluralaner (11 isolates) and several other acaricides (not all tested for each isolate), with the following outcomes for various agents:⁴⁷

- ▶ Fluralaner: Isolates were highly susceptible to fluralaner with $LC_{90} < 15.63$ ppm.
- ▶ Phoxim: Demonstrated variable sensitivity as 6 of 11 isolates had LC_{90} greater than the recommended concentration for chicken houses (2,000 ppm). The average LC_{90} for the 11 recent isolates was 3 dilutions higher than an older isolate collected in 2011.
- ▶ Deltamethrin: 3 of 3 isolates tested had LC_{90} exceeding the recommended concentration for chicken houses (250 ppm).
- ▶ Propoxur: 2 of 2 isolates tested had $LC_{90} > 1,000$ ppm.
- ▶ Spinosad: 1 of 11 isolates tested had LC_{90} (> 4000 ppm) higher than the highest recommended concentration in the field (4000 ppm).

Additional in vitro research found that pyrethroid-resistant Northern Fowl Mites were also extremely sensitive to fluralaner.⁴⁸ The test isolate was resistant to pyrethroids (permethrin, deltamethrin) despite not being exposed to acaricides for at least 10 years. Still, the LC_{99} for fluralaner against adult fowl mites was achieved at only 8.09 ppm (study details presented in Efficacy section).

The Exzolt dosage regimen (2 administrations seven days apart) was designed to ensure effective treatment and elimination of all mite development stages from the flock, which limits resistance development. In addition, research⁴⁵ showed that female Poultry Red Mites dying after a blood meal from a fluralaner-treated hen were unable to lay any eggs, suggesting that the development of less sensitive mite stages from eggs is unlikely. Correct use of Exzolt according to label directions cannot be overemphasized, such as providing the full approved dose regimen (2 consecutive administrations 7 days apart), estimating bird weights as closely as possible, etc.



PHARMACOLOGY AND ACTIVITY SUMMARY

- ▶ After oral administration, fluralaner is rapidly absorbed, reaching maximum concentrations in plasma within 36 hours after first administration and within 12 hours after second administration following start of administration via medicated drinking water.
- ▶ The bioavailability is high, approximately 91% of the dose. Fluralaner is highly bound to protein. The apparent elimination half-life is approximately 5 days following oral administration.
- ▶ Fluralaner is a very potent acaricide, acting extremely fast (within 4 hours) and for at least 15 days when administered to chickens by the recommended Exzolt dosage regimen.
- ▶ Exzolt significantly impairs reproduction of *D. gallinae*. During the first 15 days post-treatment, all mites died within 24 hours without laying any eggs, and after 19 days, mite reproduction was still seriously limited due to a remaining killing effect on female mites combined with a reduction of their oviposition, thus limiting development of any less sensitive mite stages from eggs.
- ▶ No resistance for fluralaner has been detected in field isolates, and fluralaner was active in vitro against mite isolates that were resistant to other acaricides.
- ▶ The Exzolt treatment regimen (2 administrations 7 days apart) provides acaricidal activity for a duration sufficient to break 2 consecutive mite life stages, thus providing a complete elimination of mite population in the building and thus limiting development of resistance.

SECTION FOUR

SAFETY





SAFETY

A comprehensive safety program was conducted to demonstrate the safety of Exzolt for chickens. One study investigated safety in laying hens, the primary production class for Exzolt, while another study evaluated safety in young 3-week-old chicks. A reproductive safety study was also conducted, evaluating safety in both treated breeder birds and their eggs/progeny. Outcomes obtained in these studies support the safety of Exzolt in pullets, breeders, and layers, per label indications.

TARGET ANIMAL SAFETY – LAYING HENS

STUDY DESIGN

A pivotal study was designed to evaluate the safety of Exzolt treatment via drinking water when administered at 0 (control), 1-, 3-, or 5-times the recommended dose (0, 0.5, 1.5, or 2.5 mg fluralaner/kg BW, respectively) for 3-times the recommended frequency (6 treatments instead of 2), for a total overall dosage up to 15-times the recommended dose (Figure 4-1).⁴⁹

- ▶ 120 laying hens at peak egg production (~28 weeks of age; under high physiological stress related to intensive egg production) were randomly allocated to 4 treatment groups of 30 birds.
- ▶ The 3 treated groups were offered tap water medicated with Exzolt as described in Figure 4-1 on 6 occasions (days 0, 1, 2, 7, 8, 9) while control group birds received non-medicated water. (As a result, the total net doses of fluralaner administered were 3-, 9-, or 15-times the recommended total dose.)
- ▶ Clinical health was monitored throughout the 35-day study, and feed and water intake were recorded. Egg production was assessed daily for each treatment group, and eggs from 10 randomly pre-selected birds in each group were evaluated for typical quality parameters. Hematology and clinical chemistry parameters were measured pre-treatment and at multiple time-points.
- ▶ Eight birds per group were euthanized on days 10 and 28 for complete gross necropsy, and complete histopathological examination was performed on organs from the control and the highest dose (5X) group.
- ▶ Data collections were performed by personnel blinded to treatment.

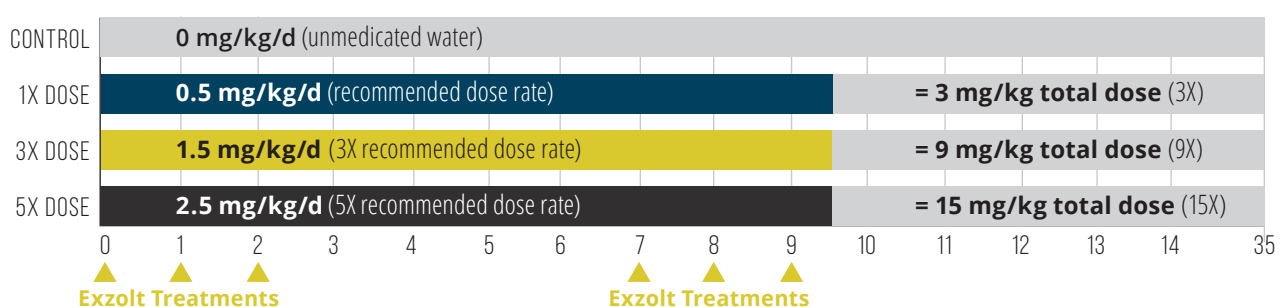


Figure 4-1: Experiment design of target animal safety study for laying hens.

RESULTS

Evaluations of the massive quantity of collected data revealed no clinically significant or relevant differences for any safety or toxicological parameters between the control group and all medicated groups, including birds treated at the 5X dose rate. Furthermore, no statistically significant differences between groups were detected for egg production, the number of abnormal eggs, or egg quality characteristics.

Water intake was similar between all treatment groups, indicating that normal palatability was maintained for drinking water medicated with Exzolt.

CONCLUSIONS

This target animal safety study demonstrated that Exzolt is well tolerated and normally palatable in laying hens, even when grossly overdosed at up to 5-times the recommended dose rate for 3-times the recommended frequency. Use of Exzolt at the recommended daily dose rate of 0.5 mg/kg BW twice at a 7-day interval clearly offers a wide margin of safety for laying hens and does not impact rates of water consumption.

TARGET ANIMAL SAFETY – 3-WEEK-OLD CHICKS

STUDY DESIGN

A similar pivotal study involving young chickens evaluated the safety Exzolt treatment via drinking water when administered at 0 (control), 1, 3, or 5-times the recommended dose (0, 0.5, 1.5, or 2.5 mg fluralaner/kg BW, respectively) for 3-times the recommended frequency (6 treatments instead of 2), for a total overall dosage up to 15-times the recommended dose (Figure 4-2).⁴³

- ▶ 320 broiler male and female chicks (3 weeks of age at start of dosing) were randomly allocated to 4 treatment groups of 80 birds.
- ▶ The 3 treated groups were offered tap water medicated with Exzolt as described in Figure 4-2 on 6 occasions at weekly intervals (days 0, 7, 14, 21, 28, 35) while control group birds received non-medicated water. (As a result, the total net doses of fluralaner administered were 3X, 9X, or 15X the recommended total dose.)
- ▶ Clinical health was monitored throughout the 54-day study, feed and water intakes were recorded, and pen body weights were obtained weekly. Hematology and clinical chemistry parameters were measured pre-treatment and at multiple time-points.
- ▶ Ten birds per group were euthanized on days 36 and 54 for complete gross necropsy, and complete histopathological examination was performed on organs from the control and the highest dose (5X) group.
- ▶ Data collections were performed by personnel blinded to treatment.

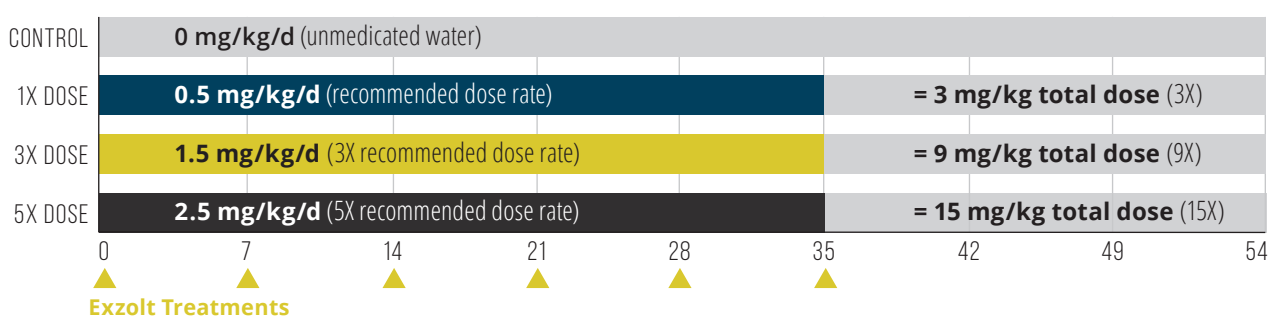


Figure 4-2: Experiment design of target animal safety study for 3-week-old chicks.

RESULTS

No clinically significant or relevant differences for any safety or toxicological parameters were detected between the control group and all medicated groups, including birds treated at the 5-times dose rate. The mortality rate per pen was similar for all groups, and treatments had no effect on BW or rates of feed consumption by chicks. Thus, use of Exzolt even at such extreme levels was not associated with any clinical, gross, or microscopic findings, productivity not measured.

Water intake was similar between all treatment groups, indicating that normal palatability was maintained for drinking water medicated with Exzolt.

CONCLUSIONS

This target animal safety study demonstrated that Exzolt is well tolerated and normally palatable in chicks and growing chickens, even when grossly overdosed at up to 5-times the recommended dose rate for 3-times the recommended frequency (6 occasions at 7-day intervals). Use of Exzolt at the recommended daily dose rate of 0.5 mg/kg BW twice at a 7-day interval clearly offers a wide margin of safety for growing chickens and does not impact water consumption. Consequently, there is no age restriction for Exzolt use in chickens.

REPRODUCTIVE SAFETY

STUDY DESIGN

A pivotal study assessed the reproductive safety of Exzolt when administered via drinking water to layer breeders (Bovans brown) at their time of peak egg production (31 weeks of age).⁵⁰

- ▶ Exzolt was purposefully overdosed at 1.5 mg/kg BW/day, a dose rate 3-times the recommended dose rate of 0.5 mg/kg (Figure 4-3). In addition, treatment was administered weekly for 4 consecutive weeks, 2-times the recommended frequency (2 treatments a week apart). As a result, the total net dose of fluralaner administered was 6 mg/kg BW, 6-times the recommended total dose.
- ▶ 432 layer breeders (384 females, 48 males) were randomly assigned to 16 pens holding 3 males and 24 females per pen. Birds in 8 pens received Exzolt treatment while birds in the other 8 pens served as non-medicated controls.
- ▶ The treatment period extended for at least 1 estrous cycle in females, complete egg formation, and spermatogenic cycle in the males. Thus, the treatment duration ensured that eggs collected for hatching during the last week of the treatment period were generated from ovum and sperm exposed to a dose in excess (3-times) of the recommended Exzolt dose.
- ▶ All eggs produced 1 week prior to the treatment period and during the last week of the treatment period were incubated. Chicks were evaluated at hatching, unhatched eggs were necropsied, and some hatchlings were monitored for 14 days to evaluate their viability. Some adult birds were necropsied at the end of the treatment period and histopathological evaluations were performed on the reproductive tract.

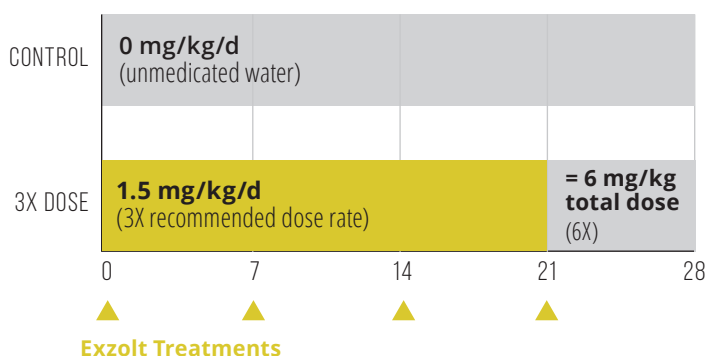


Figure 4-3: Experiment design of reproductive safety study for layer breeders.

RESULTS

No adverse impact of treatment with Exzolt was detected in regard to adult pen weight, egg production, egg weight, egg fertility, chick hatchability, chick viability, and day 14 chick weight. Furthermore, gross and microscopic examinations revealed no tissue alterations or changes in weights of reproductive organs attributable to Exzolt administration.

Water intake was similar between all treatment groups, indicating that normal palatability was maintained for drinking water medicated with Exzolt.

CONCLUSIONS

This reproductive safety study conducted in layer breeders demonstrated that Exzolt has no adverse effect on overall reproductive performance, even when overdosed at 3-times the recommended dose rate for 2-times the recommended frequency. Use of Exzolt at the recommended daily dose rate of 0.5 mg/kg BW twice at a 7-day interval clearly offers a wide margin of safety for breeder chickens and does not impact water consumption.

WITHDRAWAL PERIOD

Multiple radiolabeled metabolism studies and tissue-depletion studies were conducted to determine the tissue residue profile for Exzolt. As a result of this extensive body of research, the maximum residue limits (MRL) for fluralaner have been established in various countries and regions. Using appropriate statistical methodology, a withdrawal period of **14 days** after last administration of Exzolt was computed for meat and offal, but notably, **NO** withdrawal period (0 days) is required for eggs.

Eggs from layers medicated with Exzolt can be immediately consumed by humans after the first Exzolt administration. Use of Exzolt poses absolutely no loss of egg value or income, whether for human consumption or for hatch. This lack of an egg withdrawal period represents a tremendous advantage for Exzolt compared to competitive mite control strategies (e.g., phoxim, or Byemite®).

ENVIRONMENTAL SAFETY

Exzolt is a Veterinary Medicinal Product. As such, the environmental safety has been scrupulously evaluated as part of the approval process, according to the most recent EU and global regulatory guidelines. The evaluation followed a regulatory Risk Assessment process based on EU agricultural practices that considers all environmental compartments (soil, groundwater and surface water), all relevant exposure pathways, and all relevant organisms that are potentially at risk.

SAFETY SUMMARY

- ▶ Two target animal safety studies demonstrated that Exzolt was well tolerated and palatable in both very young birds as well as adult hens under high physiological stress related to intensive egg production, even when dosed at 5 times the recommended dose for 3 times the recommended duration of treatment (no adverse impacts on health, egg production, or growth performance).
- ▶ A reproductive safety study demonstrated that Exzolt is well tolerated in breeder chickens, even at 6-times the intended total dosage (no adverse effects on fertility, hatchability, chick viability, or overall reproductive performance).
- ▶ Use of Exzolt at the recommended daily dose rate of 0.5 mg/kg BW twice at a 7-day interval clearly offers a wide margin of safety for all classes of pullets, breeders, and layers and has no impact on water consumption.
- ▶ Eggs from hens treated with Exzolt are not harmful to consumers and **NO** withdrawal period is necessary before collection of eggs for human consumption (even on the day of administrations or between administration days). A withdrawal period of 14 days after last administration of Exzolt is required for human consumption of meat and offal.



NOTES

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BREAKTHROUGH WELFARE

Exzolt™
FLURALANER





SECTION FIVE

EFFICACY





EFFICACY

The recommended treatment regimen for Exzolt (0.5 mg/kg BW twice at a 7-day interval) was identified as a result of a comprehensive dose-determination program that evaluated multiple doses and durations using artificial infestations of Poultry Red Mites under experimental conditions. Clinical data demonstrating the efficacy of Exzolt against natural Poultry Red Mite infestations in layers, breeders, and pullets was subsequently generated from 4 studies:

- ▶ Three EU-based dose-confirmation studies were designed to confirm under field conditions the dose and frequency of treatment with Exzolt that was needed for efficacy against natural infections with *D. gallinae*. Because the earlier dose-determination studies showed that the frequency of treatment is also relevant for efficacy, the dose-confirmation studies focused on 2 treatments at a 7-day interval.
- ▶ An extensive 9-site field study assessed the efficacy of Exzolt against Poultry Red Mites for chickens reared under commercial production conditions.

Research also investigated the activity and efficacy of Exzolt against Northern Fowl Mites (*O. sylviarum*) when used at the same dose regimen as that used for Poultry Red Mites. An in vitro study confirmed the susceptibility of Northern Fowl Mites to fluralaner, and 1 dose-confirmation study, 1 field efficacy study, and 1 natural infestation efficacy study documented the efficacy and duration of the 2-dose Exzolt regimen against Northern Fowl Mite infestations in chickens.

EFFICACY VS. POULTRY RED MITES (*D. GALLINAE*)

TREATMENT REGIMEN RATIONALE

The objective of dose-determination studies was to select a treatment regimen (dose, number of administrations, and administration interval) that provides efficacy (killing activity) against Poultry Red Mites that feed on Exzolt-treated chickens. The duration of efficacy was intended to cover a period that is needed for the completion of 2 consecutive mite life cycles, an efficacy duration of at least 15 days. Mite development from egg to the first blood-

feeding stage (hematophagous protonymphs) normally occurs within 3 to 5 days (Figure 1-2), and a blood meal is required for further development of protonymphs to deutonymphs and adults, and also for production of mite eggs. Therefore, the target efficacy duration (~15 days) ensures that all non-blood-feeding mite development stages present at the time of first Exzolt administration will eventually ingest fluralaner once they develop into a blood-feeding stage.

DOSE-DETERMINATION RESEARCH

In preliminary research, a regimen dosing Exzolt at 0.5 mg/kg BW twice at a 7-day interval was identified as the most effective method for the treatment of induced red mite infestations on hens.⁴³ This regimen provided sufficient mite-killing efficacy against *D. gallinae* for at least 15 days and was thus able to span the targeted 2 mite life cycles. Other single-dose treatments, even those of higher drug concentration, did not provide persistent efficacy for 15 days.

A second study evaluated only 2-dose regimens where Exzolt was administered at a 7-day interval at 0, 0.25, 0.5, or 1.0 mg/kg BW to hens artificially challenged with red mites.⁴³



ANIMALS

32 laying hens
22 weeks of age

STUDY DESIGN

- ▶ A controlled, randomized dose-determination study. On day -12, hens were allocated to 4 treatment groups according to BW, 8 birds group.
- ▶ Exzolt treatments were administered via drinking water (first dose=day 0) as follows:
 - Group A: untreated control group
 - Group B: 0.25 mg fluralaner/kg BW/day twice 7 days apart
 - Group C: 0.5 mg fluralaner/kg BW/day twice 7 days apart
 - Group D: 1.0 mg fluralaner/kg BW/day twice 7 days apart
- ▶ Each hen was artificially infested with *D. gallinae* every 3 to 4 days until day 26.
- ▶ The numbers of dead, damaged, and live mites were counted 24 hours after each mite infestation to allow for determination of acaricidal effect.
- ▶ Mite mortality and inhibition were calculated for each treatment group at each time-point.

EFFICACY CRITERIA

Efficacy assessment was based on percent of mite mortality for treated birds as compared to the negative controls, and the duration of high efficacy.

RESULTS

All 3 Exzolt treatment groups experienced high mite mortality and inhibition (most 100%) for 1 to 5 days after each dose, while only 2.7% mite mortality was observed in controls. The low-dose group (B, 0.25 mg/kg) did not provide suitable efficacy for 15 days. In contrast, Group C birds (0.5 mg/kg) demonstrated consistent 100% efficacy against red mites, providing complete mortality for 15 days (Figure 5-1). The high-dose group (D, 1.0 mg/kg) did not show a significant additional benefit to justify the greater dose.

CONCLUSIONS

The Exzolt dosage regimen of 0.5 mg fluralaner/kg BW twice at a 7-day interval achieved 100% mite-killing efficacy for at least 15 days, thus meeting the target efficacy duration spanning 2 mite development cycles.

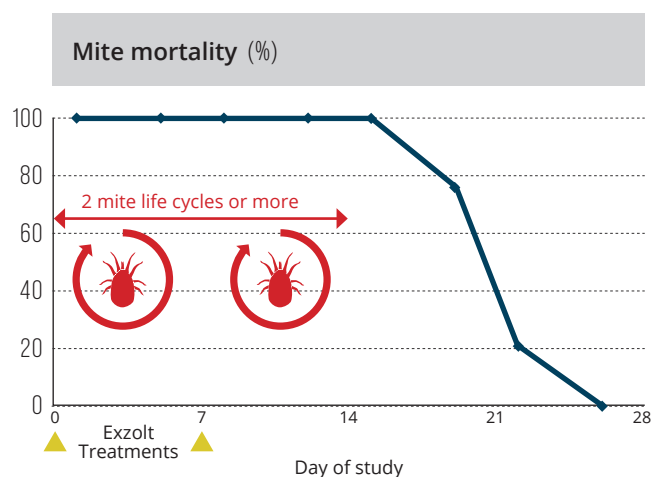


Figure 5-1: Mite mortality (*D. gallinae*; assessed 24 hours after each artificial infestation) experienced by hens administered Exzolt at 0.5 mg fluralaner/kg BW twice 7 days apart (Group C).

DOSE-CONFIRMATION STUDIES

Three studies were conducted to confirm the efficacy of the Exzolt dosage regimen found effective in previous dose-determination studies (0.5 mg fluralaner/kg BW in drinking water twice at a 7-day interval). The 3 studies, conducted under a common protocol at commercial laying farms of different sizes in Germany, France, and Spain, evaluated Exzolt dose efficacy for treating natural infestations of Poultry Red Mites (*D. gallinae*) (Table 5-1).⁴³ Unlike the previous dose-determination studies that directly evaluated efficacy by killing mites feeding on treated animals, these dose-confirmation studies assessed the effect of treatment on the reduction of the mite population in the house.



ANIMALS

Over 166,600 laying hens
46-58 weeks of age

STUDY DESIGN

- ▶ Three controlled, single-site dose-confirmation studies. Each study involved a qualifying commercial laying hen farm (in Germany, France, or Spain) with 2 comparable houses naturally infested with Poultry Red Mites (no recent acaricide treatment).
- ▶ The 2 houses at each of the 3 enrolled farms were as similar as possible with regard to size, breed, and age of chickens; feed/water distribution systems; and housing system (Table 5-1).
- ▶ Mite infestation levels of each house were assessed using mite traps, with farm eligibility confirmed if mite counts averaged > 100 in each house. Field isolates of study mites were confirmed to be susceptible to fluralaner using a contact in vitro test ($LC_{50} < 15.63$ ppm).
- ▶ On day 0 at each farm, birds in 1 house were treated with Exzolt (0.5 mg/kg BW/day twice 7 days apart) and the remaining house was left untreated (controls). Houses with the highest mite infestations were selected for Exzolt treatment, for animal welfare reasons. Treatment was administered either using a medication tank or a dosing pump.

	STUDY A	STUDY B	STUDY C
COUNTRY	Germany	France	Spain
PRODUCTION TYPE	Laying hens	Laying hens	Laying hens
HYBRID/BREEDS	Lohmann Brown	Hy-Line Brown	Lohmann LSL
NO. CHICKENS/HOUSE ON DAY 0	2715 (E) 2700 (C)	32,467 (E) 18,851 (C)	54,947 (E) 54,987 (C)
START MONTH	May	October	March
AGE ON DAY 0	46 wk	58 wk	53 wk (E) 54 wk (C)
STUDY DURATION	~ 4 months	~ 3 months	~ 6 months
MEAN INITIAL MITE COUNTS/ TRAP/HOUSE (BASELINE, ALL MITE STAGES AT DAY -1)	2250 (E) 751 (C)	1610 (E) 367 (C)	1313 (E) 1194 (C)
DRINKING WATER SYSTEM	Tank	Dosing pump	Dosing pump
HOUSING SYSTEM	Barn + free-range	Enriched cages	Enriched cages

E=Exzolt-treated house; C=control house

Table 5-1: Description of 3 Exzolt dose-confirmation studies.

- ▶ Additional mite traps were placed at the same locations on day -1 (baseline infestation level) and at multiple time-points thereafter. Traps were removed after 24 hours for mite differentiation and counting.
- ▶ Data were also collected regarding adverse events and the number of dead chickens, laid eggs, and downgraded eggs (study B) per house.
- ▶ Personnel involved in the assessment of efficacy and safety criteria were blinded to treatment group assignments.

EFFICACY CRITERIA

Efficacy assessment was based on the percentage reduction of mites (mobile stages, i.e., larvae, nymphs, adults) in the treated house at each post-treatment time-point compared to the control house. Production parameters were descriptively analyzed.

RESULTS

Study A was concluded after 4 months because mite counts decreased spontaneously in the control group. Study B was concluded at the end of the scheduled production cycle of the hens, after about 3 months. Study C extended about 6 months, until regrowth of the mite population in the treated house reached 50% of the baseline count. However, the birds in the control house received treatment with another product at week 11 (day 77) to ensure appropriate animal welfare. Analyses of water samples from all 3 studies confirmed that actual fluralaner concentrations were generally close to target levels.

The efficacy of Exzolt against all mite stages compared to controls is summarized in Figure 5-2. For all studies, efficacy resulting from the complete 2-dose regimen exceeded 95% by day 6 and 99% by day 14, achieving 100% at several time-points. Efficacy remained high through the end of the production cycle in study B (3 months) and for 17 and 19 weeks in studies A and C, respectively.

As expected, Exzolt efficacy against larval non-blood-feeding stages of mites (Figure 5-3) increased more slowly than efficacy against nymphs and/or adults. However, as larval stages progressed to later blood-feeding forms, Exzolt efficacy was more consistently detected, with more than 95% efficacy confirmed by day 6 (before the second administration). Thereafter, mite counts for larvae, nymphs, and adults were similar.

Favorable impacts of effective mite control were also observed for several production parameters (Figure 5-4). For each parameter, the difference between the Exzolt group and controls was calculated pre-treatment (week -1) and post-treatment (from first treatment through study conclusion). Mortality remained very low in both houses of each study. Overall, birds treated with Exzolt experienced an increase in the laying rate (0.9-1.9%) and a decrease in the percentage of downgraded eggs in study B (3.4% more normal eggs). These productivity benefits reflect the successful and prolonged relief of ectoparasite burdens by Exzolt, which thus allowed for better productivity in these field studies.

No adverse events related to the use of Exzolt were reported during the studies.

CONCLUSIONS

Dose-confirmation studies demonstrated that Exzolt consistently provided high efficacy against Poultry Red Mites when administered at 0.5 mg fluralaner/kg BW given twice 7 days apart via drinking water. Treated birds also demonstrated excellent tolerance of Exzolt, with no treatment-related adverse effects observed. Under commercial production conditions in these studies, the 2-dose Exzolt regimen successfully controlled mite infestations for extended periods far exceeding the 2 weeks of effective blood concentrations. This extended period of efficacy was due to the ability of Exzolt to interrupt mite life cycles and, thus, massively decrease mite populations of the houses.

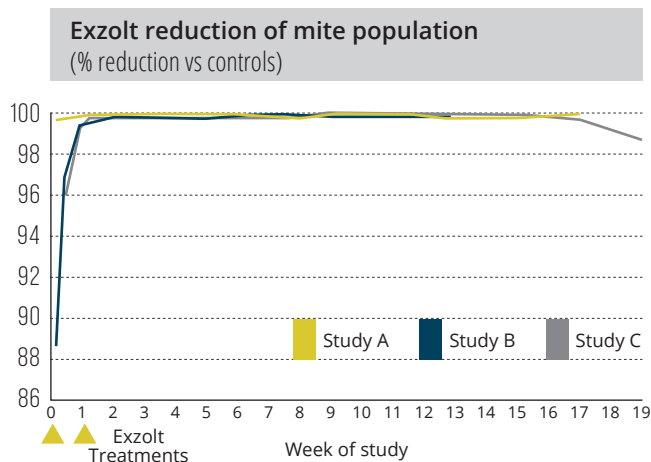


Figure 5-2: Percentage of Exzolt reduction of mite population vs controls in dose-confirmation studies (sum of larvae, nymphs, adults; weeks after first Exzolt administration).

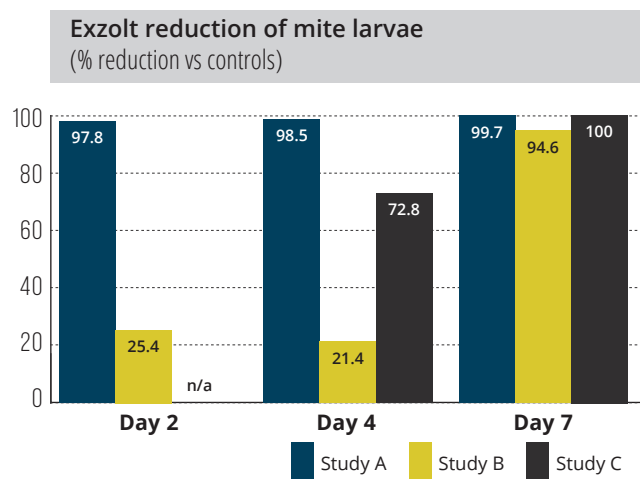


Figure 5-3: Percentage of Exzolt reduction of non-bloodsucking mite larvae 24 hours after trap placement (on days 1, 3, 6) vs controls in dose-confirmation studies (days after first Exzolt treatment).

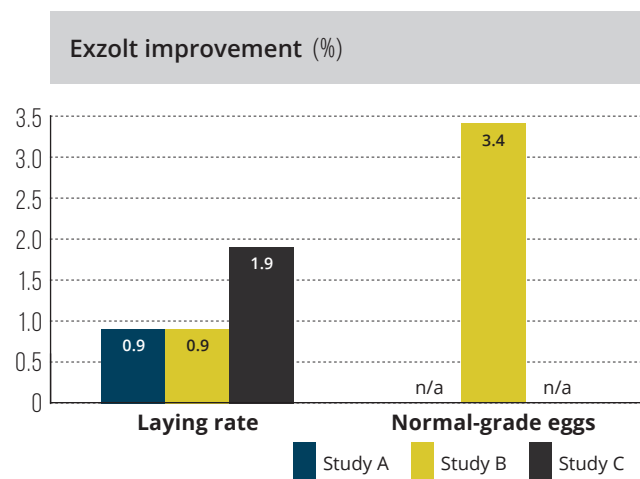


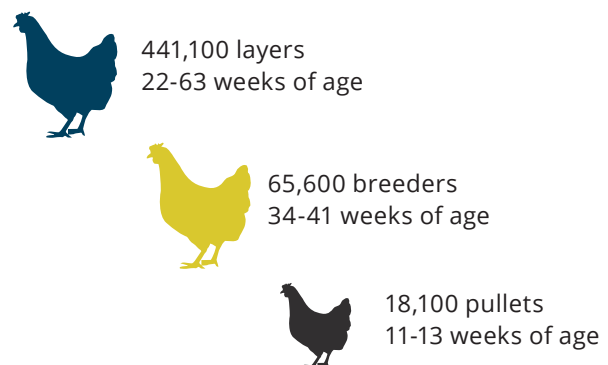
Figure 5-4: Productivity improvements of Exzolt group vs controls for average laying rate and percent of normal (non-downgraded) eggs over time; post-treatment change vs pre-treatment (treated house - control house).

FIELD EFFICACY STUDY

An extensive, multi-site field study was conducted in the Germany, France, and Spain to evaluate the efficacy of Exzolt (0.5 mg fluralaner/kg BW in drinking water twice at a 7-day interval) for treating natural infestations of Poultry Red Mites (*D. gallinae*) at a variety of commercial poultry farms.⁴³ The study was conducted at 9 sites involving 5 layer farms, 2 breeder farms, and 2 replacement chicken (pullet) farms (Table 5-2), using a protocol similar to the previous dose-confirmation studies. The farms covered all relevant chicken production types (layers, breeders, pullets), various housing systems (barn, barn with free-range, enriched cages), various drinking water systems (dosing pump, medication tank, gravity tank) and drinking water compositions, and different climatic conditions.

ANIMALS

Over 524,000 chickens:



STUDY DESIGN

- Field efficacy study at 9 sites, controlled and partially blinded. Each site was a qualifying commercial chicken farm (5 in Germany, 3 in France, 1 in Spain) with 2 comparable houses naturally infested with Poultry Red Mites (no recent acaricide treatment). Sites were enrolled between mid May and late July, at the peak of mite season.
- The 2 houses at each of the 9 sites (550-100,000 birds/house) were as similar as possible with regard to size, breed and age of chickens, feed/water distribution systems, housing system, climate control, and cleaning system (Table 5-2).

- ▶ Mite infestation levels of each house were assessed using mite traps. Field isolates of study mites were confirmed to be susceptible to fluralaner using a contact in vitro test ($LC_{90} < 15.63$ ppm for 7/9 isolates, and 15.63-31.25 ppm for the other 2 isolates).
- ▶ On day 0 at each farm, birds in 1 house were treated with Exzolt (0.5 mg/kg BW/day twice 7 days apart) and the remaining house was left untreated (controls). Houses with the highest mite infestations were usually selected for Exzolt treatment, for animal welfare reasons.
- ▶ Additional mite traps were placed at the same locations on day -1 (baseline infestation level) and at multiple time-points thereafter. Traps were removed after 24 hours for mite differentiation and counting.
- ▶ Data were also collected regarding the number of dead chickens and laid eggs, and 2 flocks were observed for adverse events. Rates of incubation, fertility, and hatchability were recorded for the 2 breeder farms.
- ▶ Personnel involved in the assessment of efficacy and safety criteria were blinded to treatment group assignments.
- ▶ The control house at 3 sites (L1, L3, L5) had to be treated for mites during the study due to an unacceptable mite burden (animal welfare). After treatment at site L3, mite counts for efficacy assessment were still available and > 95% through the end of the production cycle at day 238.

	SITE L1	SITE L2	SITE L3	SITE L4	SITE L5	SITE B1	SITE B2	SITE P1	SITE P2
COUNTRY	Germany	Germany	France	France	Spain	Germany	France	Germany	Germany
PRODUCTION TYPE	Layer	Layer	Layer	Layer	Layer	Breeder	Breeder	Pullet	Pullet
HYBRID/BREEDS	Dekalb White	ISA Brown	ISA Brown	Novo Brown	Lohmann Brown	Ross 308	Ross PM3J (hens) RJ344 (cocks)	Tetra Brown	Tetra Brown ^a
NO. CHICKENS/ HOUSE ON DAY 0	2000	550	56,000	62,000	100,000	24,000	8800	4600 (E) 7500 (C)	3000
AGE ON DAY 0	40 wk	54 wk (E) 63 wk (C)	40 wk	36 wk	22 wk (E) 27 wk (C)	35 wk (E) 34 wk (C)	41 wk	13 wk	11 wk
MEAN INITIAL MITE COUNTS/ TRAP/HOUSE (BASELINE, ALL MITE STAGES AT DAY -1)	568 (E) 409 (C)	2138 (E) 1673 (C)	2244 (E) 758 (C)	196 (E) 162 (C)	680 (E) 829 (C)	1148 (E) 521 (C)	8279 (E) 1841 (C)	430 (E) 997 (C)	1074 (E) 347 (C)
DRINKING WATER SYSTEM	Dosing pump	Gravity tank	Dosing pump	Dosing pump	Dosing pump	Dosing pump	Dosing pump	Medication tank	Medication tank
HOUSING SYSTEM	Barn	Barn + free-range	Enriched cages	Enriched cages	Enriched cages	Barn	Barn	Barn	Barn
HOUSE (TREATMENT GROUP) SEPARATION	Poor (same building; non-hermetic door)	Poor (same building; non-hermetic door)	Good (same building; corridor)	Good (same building; corridor)	Good (separate buildings)	Good (separate buildings)	Good (separate buildings)	Good (same building; corridor)	Good (same building; corridor)

E=Exzolt-treated house C=control house ^aalso Harco Black, Koenigsberger Blue, and Sussex

Table 5-2: Description of 9-site Exzolt field study.

EFFICACY CRITERIA

Efficacy assessment was based on the percentage reduction of mites (larvae, nymphs, adults) in the treated house at each post-treatment time-point compared to the control house. Production parameters were descriptively analyzed.

RESULTS

The efficacy of Exzolt (against all mite stages) compared to controls is summarized in Figure 5-5. The onset of efficacy was very fast, with an overall average mite reduction of 46.6% observed after the first night following the day of initial treatment (day 0; trap placed and mites collected for 24 hours). Efficacy was already 97.6% on day 3, achieved >97% by day 6, and persisted at that level for a lengthy period of time depending on the farm (100% efficacy at day 14 after the second Exzolt dose in 7/9 farms). A similar degree of fast and high onset of mite control efficacy was observed in all production types (layers, breeders, pullets) and housing systems.

The duration of mite population control following the 2-dose Exzolt regimen was assessed at the house level using a threshold of 90% reduction vs non-medicated controls (Figure 5-6). The duration of mite control was long and variable (2-8 months) and did not depend on the production type or the drinking water system, but was impacted by the duration of the remaining time until the end of the chicken production cycle and on the level of house separation of the compared groups. At 6 sites (L3, L4, B1, B2, P1, P2), no decrease of efficacy was observed until the end of the study (6 to 34 weeks; the 6-week efficacy of the pullet sites reflected the short remaining production cycle and shipment/removal of the birds). Three sites (L1, L2, L5) experienced a regrowth of the mite population in the treated group after 8 to 18 weeks.

Regarding house separation, the presence of 2 rooms close to each other under 1 roof, separated by a wall but connected by an incompletely hermetic door, allowed faster mite regrowth in the treated house (in about 2 months) at sites L1 and L2 than that observed at farms with appropriately separated houses or rooms. Under real field conditions, the 2 houses would be treated at the same time. In 4 of the 5 layer and breeder farms with an appropriate separation between the study groups (sites L3, L4, B1, B2), the mite population was controlled until the end of the production cycle (16-34 weeks).

Mortality was low at most of the farms and comparable between the study groups before and after treatment. However, the acaricidal efficacy of Exzolt and resulting relief from mite infestation positively impacted the laying rate at 6 of the 7 layer or breeder farms, with a relative increase ranging from 0.9% to 12.6% (Figure 5-7). The sole site with reduced post-treatment lay rate (B1) had a known history of reduced performance in the house selected for treatment relative to the house used as a control. In regard to breeder performance, increased fertility and hatchability rates (2.8% and 1.7%, respectively) were observed at the B2 site, but no improvements were detected at the other B1 breeder farm (site houses not fully comparable for these parameters).

No adverse events related to the use of Exzolt were reported during the study.

CONCLUSIONS

The field study confirmed the high efficacy and excellent safety of Exzolt when administered in the drinking water (0.5 mg fluralaner/kg BW twice 7 days apart). Under commercial production conditions at 9 diverse layer, breeder, or pullet farms, Exzolt provided fast and persistent reductions in mite counts and positively impacted production parameters (rates of lay, fertility, hatchability). The 2-dose Exzolt regimen successfully controlled natural mite infestations for extended periods, far exceeding the time of actual drug administration (e.g., up to 34 weeks, or nearly 8 months, at 1 layer farm).

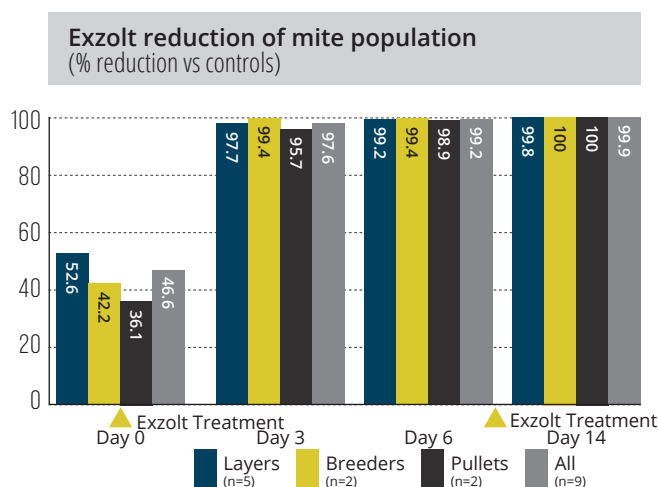


Figure 5-5: Percentage of Exzolt efficacy against mites vs controls at 9 sites of field efficacy study (larvae, nymphs, adults; days after first Exzolt treatment).

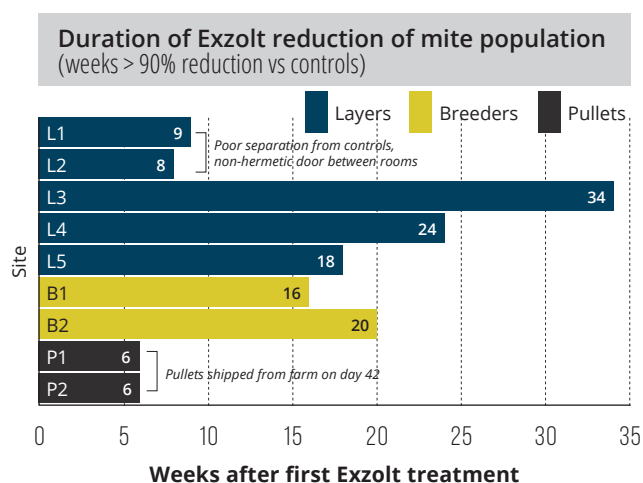


Figure 5-6: Duration (weeks) of mite population control provided by Exzolt at each study site (last sample date with mite reduction > 90% vs controls).

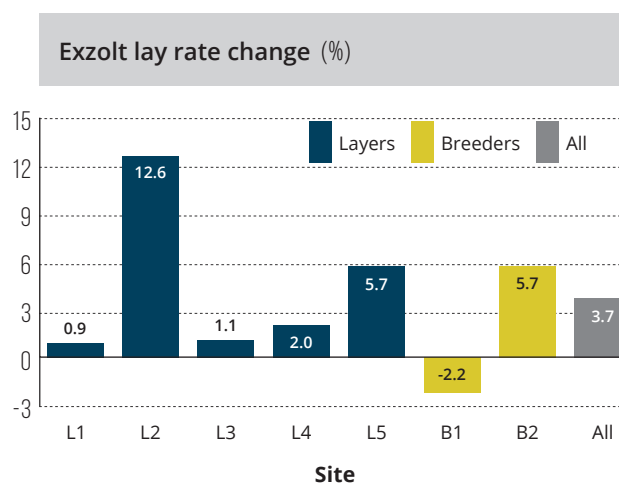


Figure 5-7: Lay rate improvements of Exzolt group vs controls; post-treatment change vs pre-treatment (treated house – control house). (At site L2, hens of the treated group were 9 weeks younger than those of the control group, likely accounting for some of the large difference at this site.)

EFFICACY VS. NORTHERN FOWL MITES (*O. SYLVIARUM*)

IN VITRO ACTIVITY

A study was conducted to determine the in vitro mortality of Northern Fowl Mites exposed to different acaricides (including fluralaner) and to assess impacts on mite egg production, hatch, and immature survival.⁴⁸

STUDY DESIGN

- ▶ 30 adult female fowl mites were placed in each of a series of filter paper packets treated with different concentrations of fluralaner, phoxim, spinosad, propoxur, deltamethrin, or permethrin.
- ▶ After 48 hours of continuous exposure, the packets were opened and mite mortality was assessed by counting and removal of dead and live mites, and eggs already deposited by the mites were counted.

- ▶ Packets were then re-sealed and held for an additional 72 hours after which emerged immature stages were counted to determine egg hatch or larval-protonymph survival.
- ▶ LC_{50} and LC_{99} values were calculated for adult mortality (probit analysis).

RESULTS

Mortality outcomes summarized in Table 5-3 indicated that the LC_{99} for fluralaner against adult mites was achieved at only 8.09 ppm. No other tested acaricides demonstrated this degree of mite susceptibility. Oviposition was reduced by fluralaner at concentrations higher than 100 ppm, while phoxim required 1500 ppm, spinosad 2000 ppm, deltamethrin 1000 ppm, and propoxur 100 ppm to reduce egg production. Permethrin had no effect on egg production. None of the acaricides reduced egg hatch or seriously impeded development to the nymph stage.

CONCLUSIONS

Northern Fowl Mites (*O. sylviarum*) appear to be similarly sensitive to fluralaner as Poultry Red Mites (*D. gallinae*). In this study, *O. sylviarum* mites were found to be extremely sensitive to fluralaner, even though the test isolate was resistant to pyrethroids (permethrin, deltamethrin) despite not being exposed to acaricides for at least 10 years. Reduction of egg production was obtained with fluralaner concentrations higher than lethal concentrations, but the drug had no further effects on reproduction in this study.

ACARIDE	LC ₅₀ (PPM)	LC ₉₉ (PPM)
FLURALANER	2.95	8.09
PHOXIM	420	750
SPINOSAD	1587	3123
PROPOXUR	86	181
DELTAMETHRIN	> 1000	> 1000
PERMETHRIN	> 1000	> 1000

Table 5-3: *In vitro* susceptibility of adult Northern Fowl Mites (*O. sylviarum*) to fluralaner and other acaricides.

DOSE CONFIRMATION STUDY – LAYERS

A study was conducted in the US to confirm that the Exzolt dosage regimen selected for *D. gallinae* in chickens (0.5 mg fluralaner/kg BW twice at a 7-day interval) was also relevant for treating infestations of Northern Fowl Mites (*O. sylviarum*).⁵¹ In this study, the efficacy of oral fluralaner was evaluated at 3 dosing regimens in layers infested with *O. sylviarum*.



ANIMALS

132 layer hens
32 weeks of age

STUDY DESIGN

- ▶ A controlled, randomized dose-confirmation study. Hens originating from an infested flock were distributed in 12 pens (11 birds/pen); 3 pens were then allocated according to vent mite-count densities to either of 4 treatment groups.
- ▶ Exzolt treatments were administered via drinking water (first dose=day 0) as follows:
 - Group A: untreated control group
 - Group B: 0.25 mg fluralaner/kg BW/day twice 7 days apart
 - Group C: 0.5 mg fluralaner/kg BW/day twice 7 days apart
 - Group D: 1.0 mg fluralaner/kg BW/day twice 7 days apart
- ▶ 6 hens in each pen were treated by gavage and 5 were left untreated to maintain an infestation challenge (permanent mite re-infestation). Pre-treatment mite counts ranged from 10 to 288/hen.
- ▶ Mite counts were performed using 2 feathers around the vent area taken from a randomly selected hen in each pen. Counts were conducted on days -1, 1, 2, 6, 8, 12, 15, 19, 22, and 26.

EFFICACY CRITERIA

- ▶ Efficacy assessment was based on the percent reduction of mites on treated hens vs. untreated controls.

RESULTS

All 3 Exzolt treatment groups experienced high efficacy (>90%) through day 19 or 22, and mite counts were significantly ($P \leq 0.013$) reduced vs controls through day 22. The efficacy of the recommended Exzolt dose appears in Figure 5-8.

CONCLUSIONS

The recommended Exzolt dosage regimen of 0.5 mg fluralaner/kg BW twice at a 7-day interval achieved 98% efficacy against Northern Fowl Mites for at least 15 days, covering at least 2 mite life cycles. Exzolt provided an efficacy profile against *O. sylviarum* similar to that provided for *D. gallinae*, even under a 'worst case' test scenario involving permanent re-infestation of treated birds due to the continuous presence of untreated mite-source hens in the same pen. (Under typical field conditions, all birds of a same flock would be treated, thus potentially allowing for total eradication of the mite population.)

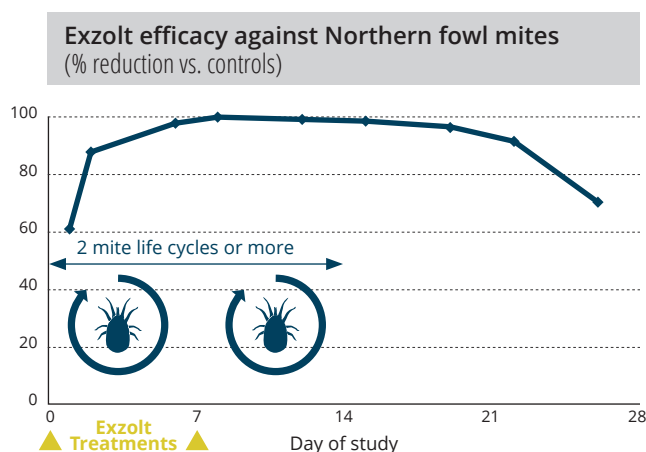


Figure 5-8: Efficacy of Exzolt (0.5 mg fluralaner/kg BW twice 7 days apart) for reducing counts of Northern Fowl Mites (*O. sylviarum*) vs. controls, under conditions with permanent re-infestation.

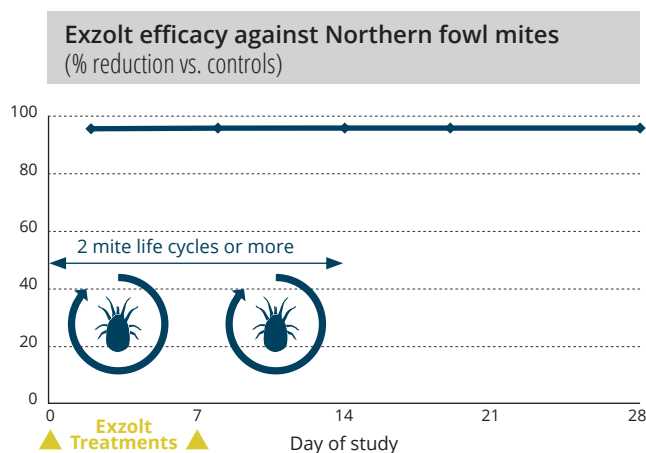


Figure 5-9: Efficacy of Exzolt (0.5 mg fluralaner/kg BW twice 7 days apart) for reducing counts of Northern Fowl Mites (*O. sylviarum*) vs. controls in a dose-confirmation study.

DOSE-CONFIRMATION STUDY – REPLACEMENT CHICKENS

Another US study was conducted to confirm the fowl mite efficacy of the Exzolt dosage regimen found effective in the previous dose-confirmation study (0.5 mg fluralaner/kg BW in drinking water twice at a 7-day interval). Conducted at a university research facility, the trial evaluated Exzolt dose efficacy for treating infestations of Northern Fowl Mites (*O. sylviarum*) in replacement chickens.³⁸



ANIMALS

132 layer hens
32 weeks of age

STUDY DESIGN

- ▶ A controlled, single-site dose-confirmation study. Birds were housed in a small-scale poultry house at a major Western US university. Birds were obtained as uninfested birds and following a brief acclimation period in the study cages were infested with Northern Fowl Mites.
- ▶ Healthy birds with established mite infestation were ranked by mite counts on day -8 and randomly assigned to 32 cages (4 birds/cage each) with 16 cages assigned to each of 2 treatment groups.
- ▶ On day 0, birds in 1 group were treated with Exzolt (0.5 mg/kg BW/day twice 7 days apart) via the drinking water while the other group remained untreated (controls). Exzolt dosage was based on cage bird weights determined on days -1 and 6.
- ▶ Mite vent counts were performed on days -7, 2, 8, 14, 19, and 28. Adequacy of infestation criteria set in the protocol specified that controls be infested with at least ≥ 25 mites per individual bird and average ≥ 50 mites per group. The criteria were met by most control birds, with average mite counts ranging from 127 to 226 between days 2 and 28.

EFFICACY CRITERIA

- ▶ Efficacy assessment was based on the percent reduction of mites on treated birds vs. untreated controls.

RESULTS

The efficacy of Exzolt compared to controls is summarized in Figure 5-9. Early onset of action was noted as mite vent counts were significantly reduced 99.6% in the Exzolt group beginning at day 2 ($P < 0.001$). Similarly, significant 100% mite count reductions ($P < 0.001$) were observed on every subsequent sample day, even at 4 weeks after initiation of treatment. No adverse events related to the use of Exzolt were observed.

CONCLUSIONS

This dose-confirmation study verified that Exzolt provided high (100%) efficacy against Northern Fowl Mite infestations of replacement chickens when administered at 0.5 mg fluralaner/kg BW given twice 7 days apart via drinking water. Treated birds also demonstrated excellent tolerance of Exzolt, with no treatment-related adverse effects observed. The Exzolt dose regimen successfully met (and far exceeded) the target efficacy duration spanning 2 development cycles for Northern Fowl Mites.

FIELD EFFICACY STUDY – US

A field study was conducted in a US university facility to evaluate the efficacy of Exzolt (0.5 mg fluralaner/kg BW in drinking water twice at a 7-day interval) for treating natural infestations of Northern Fowl Mites (*O. sylviarum*) in layer chickens derived from a commercial poultry farm in California.⁴³



ANIMALS

800 commercial White Leghorn chickens (Hy-Line W36), 16 weeks of age.

STUDY DESIGN

- ▶ A controlled, single-site field efficacy study. Commercial layer chickens were obtained as uninfested birds and following a brief acclimation period became naturally infested with Northern Fowl Mites.
- ▶ 800 healthy birds with established mite infestation were randomly placed into 400 cages (2/cage) on day -6. Cages were then randomly assigned to either of 2 treatment groups (400 cages/group).

- ▶ On day 0, birds in 1 group were treated with Exzolt (0.5 mg/kg BW/day twice 7 days apart) using a dosing pump system while the other group remained untreated (controls). Exzolt dosage was based on cage bird weights determined on day -1 and water consumption measured on days -4 and -3.
- ▶ Mite vent counts were performed on days -5, 2, 8, 14, 19, and 28. Adequacy of infestation criteria set in the protocol specified that at least 1 bird from 60 of 80 sampled control cages had a mite count ≥ 25 mites per individual bird. The criteria were met at each counting day.

EFFICACY CRITERIA

- ▶ Efficacy assessment was based on the percent reduction of mites on treated birds vs. untreated controls.

RESULTS

The efficacy of Exzolt compared to controls is summarized in Figure 5-10. Exzolt provided early onset of action as mite vent counts were significantly reduced 99.5% beginning at day 2. Mite count reductions of 100% were achieved on every subsequent sample day, even at 4 weeks after initiation of treatment. No adverse events related to the use of Exzolt were observed.

CONCLUSIONS

This US field efficacy study demonstrated that Exzolt provided high (100%) efficacy against Northern Fowl Mite infestations of commercial chickens when administered at 0.5 mg fluralaner/kg BW given twice 7 days apart via drinking water. Treated birds also showed excellent tolerance of Exzolt, with no treatment-related adverse effects observed. The Exzolt dose regimen successfully met (and far exceeded) the target efficacy duration spanning 2 development cycles for Northern Fowl Mites.

Exzolt efficacy against Northern Fowl Mites (% reduction vs. controls)

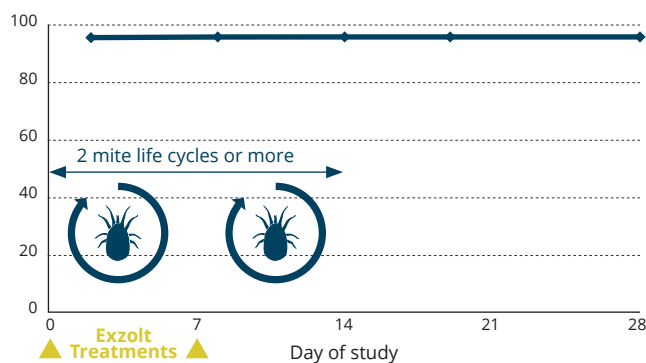


Figure 5-10: Efficacy of Exzolt (0.5 mg fluralaner/kg BW twice 7 days apart) for reducing counts of Northern Fowl Mites (*O. sylviarum*) vs. controls in a US field efficacy study.

NATURAL INFESTATION EFFICACY STUDY – BRAZIL

Another field study was conducted in Brazil to evaluate the efficacy of Exzolt (0.5 mg fluralaner/kg BW in drinking water twice at a 7-day interval) for treating natural infestations of Northern Fowl Mites (*O. sylviarum*) in laying hens from a commercial poultry farm.⁵²



ANIMALS

60 commercial laying hens.

STUDY DESIGN

- ▶ A controlled, single-site natural infestation efficacy study. Chickens were naturally infested with Northern Fowl Mites.
- ▶ 60 healthy birds with established mite infestation were randomly assigned to 2 treatment groups (30/group), with homogeneity of mite infestation confirmed on day -7. Groups were housed in different barns to avoid cross-infestation, with each barn holding 3 pens, 10 birds/pen.
- ▶ On day 0, birds in 1 group were treated with Exzolt (0.5 mg/kg BW/day twice 7 days apart) via the drinking water while the other group remained untreated (controls). Exzolt dosage was based on total bird weight per pen and average daily water consumption measured for each pen on days -3 to -2.

- ▶ Mite vent counts were performed on days 2, 8, 14, 19, and 28.

EFFICACY CRITERIA

- ▶ Efficacy assessment was based on the percent reduction of mites on treated birds vs. untreated controls.

RESULTS

The efficacy of Exzolt compared to controls is summarized in Figure 5-11. Early onset of action was noted as mite vent counts were significantly reduced 94% in the Exzolt group beginning at day 2 ($P < 0.01$). Furthermore, significant 100% mite count reductions ($P < 0.01$) were observed on every subsequent sample day, even at 4 weeks after initiation of treatment. No adverse events related to the use of Exzolt were observed.

CONCLUSIONS

This natural infestation efficacy study verified that Exzolt provided high (100%) efficacy against Northern Fowl Mite infestations of laying hens when administered at 0.5 mg fluralaner/kg BW given twice 7 days apart via drinking water. Treated birds also demonstrated excellent tolerance of Exzolt, with no treatment-related adverse effects observed. The Exzolt dose regimen successfully met (and far exceeded) the target efficacy duration spanning 2 development cycles for Northern Fowl Mites.

Exzolt efficacy against Northern Fowl Mites (% reduction vs. controls)

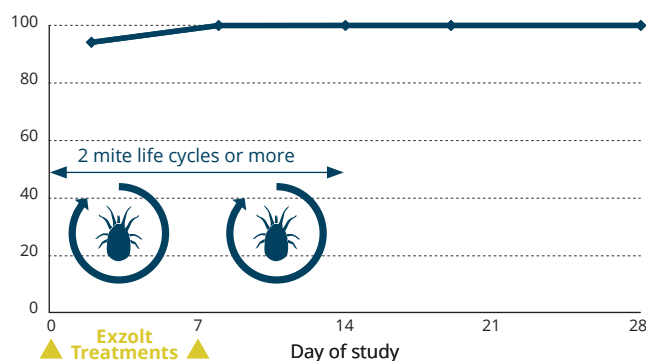


Figure 5-11: Efficacy of Exzolt (0.5 mg fluralaner/kg BW twice 7 days apart) for reducing counts of Northern Fowl Mites (*O. sylviarum*) vs. controls in a Brazilian natural infestation efficacy study.

EFFICACY SUMMARY

- ▶ The Poultry Red Mite efficacy of Exzolt administered in the drinking water of chickens at the recommended dosage regimen of 0.5 mg fluralaner/kg BW twice 7 days apart was evaluated in multiple studies, including a dose-determination study using artificial Poultry Red Mite infestation as well as several dose-confirmation studies and a 9-site field-efficacy study conducted under natural infestation conditions.
- ▶ Total or near-total elimination of Poultry Red Mite populations in the treated houses was quickly achieved in all layer, breeder, and/or pullet houses treated.
- ▶ This effect was maintained for a duration largely exceeding the duration of effective blood concentrations required to kill mites feeding on the treated birds.
- ▶ Northern Fowl Mites were found to be extremely sensitive to fluralaner in vitro. Two dose-confirmation studies verified Exzolt efficacy against fowl mites using the same dose regimen as for Poultry Red Mites. Furthermore, 2 field efficacy studies demonstrated fast Exzolt efficacy in reducing Northern Fowl Mite infestations within 2 days of treatment initiation, and 100% efficacy that extended for at least 4 weeks after initiation of treatment.
- ▶ In several studies, positive impacts on productivity (e.g., laying rate, fertility, etc.) were observed for treated birds, likely due to the ability of Exzolt to massively decrease the mite population of the house.
- ▶ Exzolt clearly offers potent, consistent, convenient, and safe efficacy against Poultry Red Mites and Northern Fowl Mites and thus helps prevent the significant adverse economic consequences of inadequately controlled mite infestation.

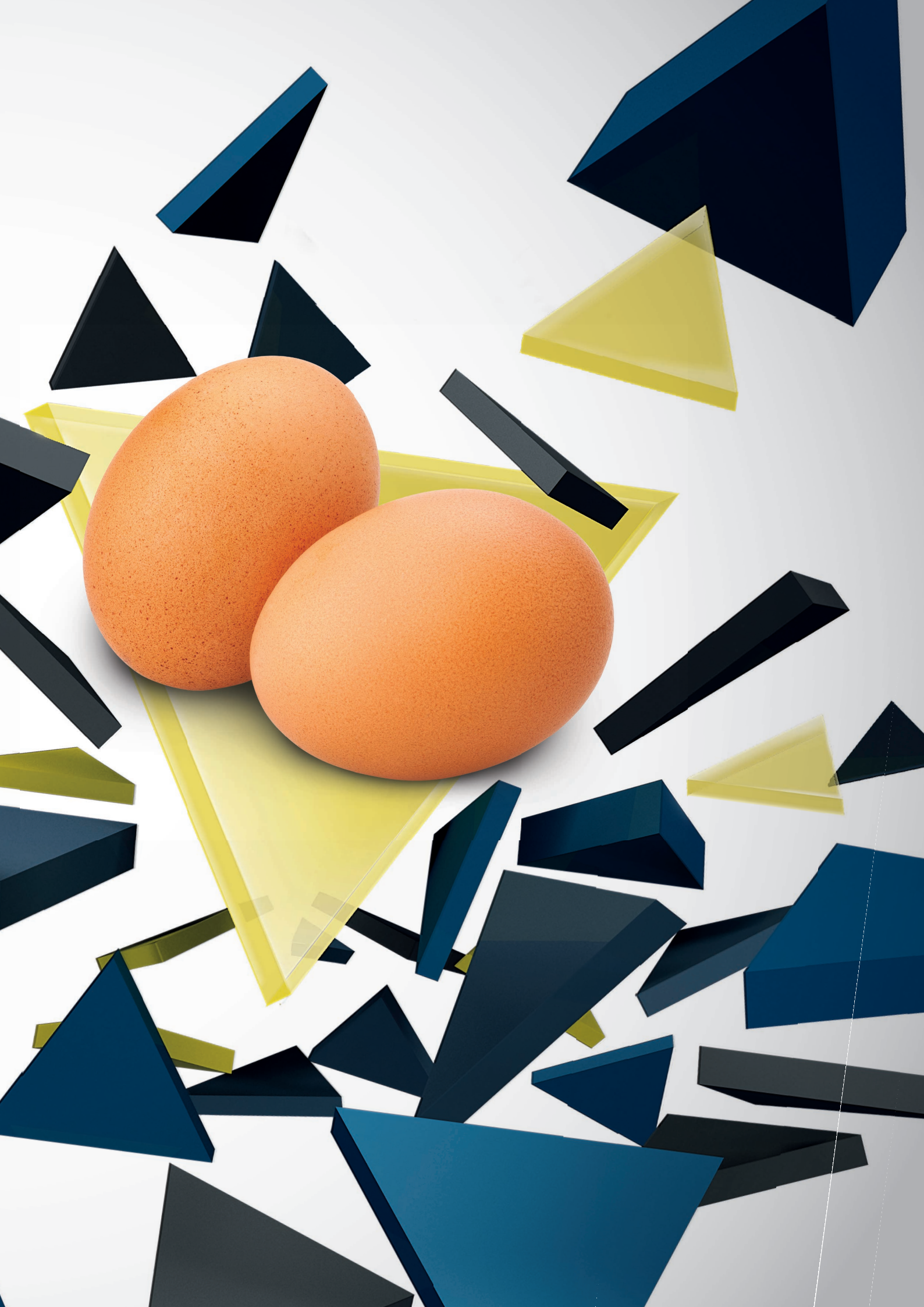
NOTES

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BREAKTHROUGH PRODUCTION

Exzolt™
FLURALANER





SECTION SIX

USAGE GUIDELINES





USAGE GUIDELINES

DOSE CALCULATION

The dose for Exzolt is 0.5 mg fluralaner per kg BW per day (equivalent to 0.05 mL Exzolt) administered twice, 7 days apart. The full course of therapy must be administered for full therapeutic effect.

First, determine the time period (in hours) needed to administer the product on the treatment day. This period must allow all birds to receive the required dose. Estimate how much water birds will consume during treatment based on their water consumption during the same period the day before start of treatment.

Next, calculate the needed volume of Exzolt, based on the total weight of the bird group to be treated. To ensure administration of the correct dose, body weight should be determined as accurately as possible and a sufficiently accurate measuring device should be used. The required amount of Exzolt on each treatment day should be calculated from the total body weight (kg) of the entire group of chickens to be treated, using the following formula:

volume of product (mL)/treatment day = Total body weight (kg) of chickens to be treated × 0.05 mL

Examples:

TOTAL BODY WEIGHT OF CHICKENS TO BE TREATED	AMOUNT OF EXZOLT PER TREATMENT DAY (ML/DAY)
40,000 kg	2000 mL (2 L)
160,000 kg	8000 mL (8 L)

(1 L of Exzolt fully treats 10,000 kg body weight across the 2 doses – e.g., 5000 chickens weighing 2 kg body weight each will use a total of 1 L of Exzolt, or 0.5 L per treatment day × 2 treatment days.)

PREPARING STOCK SOLUTION AND DOSING BIRDS

The instructions below need to be followed in the order described to prepare the medicated water.

- ▶ Check the water system to ensure it works properly and is free of leaks; also ensure that water is available to all nipple or bell drinkers.
- ▶ For each day of treatment, medicated water must be freshly prepared.
 - Mix the required volume of Exzolt into a large medication tank or with additional water to create a stock solution to be dosed by proportioner or dosing pump. Always add product and water simultaneously in order to avoid foaming. It is important to rinse the container used to measure the required product volume during the filling phase in order to ensure that the complete dose is emptied into the medication tank or the stock solution and that no residues remain in the measuring device. Stir the stock solution or the content of the medication tank gently until the medicated water is homogeneous, and connect the medication tank or the proportioner or dosing pump to the drinking water system.
- ▶ Make sure the dosing pump is properly set to deliver the medicated water during the predetermined treatment period (hours per day).
- ▶ Prime the drinker lines with medicated water and check to see when medicated water has reached the end of the line. This procedure should be repeated on each day of administration.

CONSIDERATIONS FOR USE OF DOSING PUMPS

Dosing pumps offer very interesting possibilities in therapeutics, allowing prompt action in targeting the treatment of a room, a pen, or a batch of animals, and the flexibility to alter the medication, or even adjust the dose, during a course of treatment. The use of dosing pumps for administering medications requires adherence to Good Practice standards to maximize the accuracy of dosing, which reduces the overuse of medications, limits the risk of developing resistance, and eases conformity with withdrawal times and maximum residue limits in meat, offal, and eggs.

The distribution of drugs via drinking water using dosing pumps requires a homogeneous active ingredient which dissolves or mixes well, reliable equipment, and a well-regulated water flow at the level of the drinkers. It is absolutely essential to check the flow of water through the drinkers, the cleanliness of the tank, the state of the dosing pump, and the valves governing the distribution of the water to each room or pen. Regular servicing of pumps is crucial for the accuracy of dosing. This also means systematic cleaning and rinsing of the equipment after each use and a yearly maintenance program.



Adhere to Good Practice standards when administering Exzolt through dosage pumps.

BREAKTHROUGH APPLICATION

Exzolt™
FLURALANER





SECTION SEVEN

SUMMARY





SUMMARY

Exzolt represents an innovative, completely new approach for comprehensive management of Poultry Red Mites and Northern Fowl Mites in chickens, providing systemic acaricidal activity within the bird instead of relying on external contact with a pesticide. Only Exzolt offers a unique combination of features and benefits that, together, distinguish the product as a major advance for optimizing the health and productivity of layers, breeders, and pullets threatened by external mite parasites.

- ▶ Contains fluralaner, a potent yet safe acaricide from a new chemical class, the first isoxazoline approved for use in poultry.
- ▶ Induces a rapid and massive decrease in mite populations in a chicken house, often with more than 99% efficacy.
- ▶ Fast kill starts within hours of administration, and the 2 administrations a week apart span 2 mite life cycles, thus disrupting mite population dynamics.
- ▶ Convenient treatment in the drinking water at a low dose of 0.5 mg/kg BW/day, repeated 1 week later, allowing uniform and accurate dosing compared to other control methods.
- ▶ Ready-to-use aqueous solution (1% fluralaner, 10 mg/mL) for simple dilution, with no sedimentation, clogging, or spoilage.
- ▶ Treats the bird, not just the surroundings. Much more effective, safe, targeted, and convenient than mite sprays.
- ▶ Highly active against mite strains resistant to classical acaricides.
- ▶ Ideal for layers due to *zero egg withdrawal* period.
- ▶ No adverse impacts on egg production, hatchability, or chick survival of breeders (in fact, positive impacts often observed).
- ▶ Safe and well tolerated in all classes of chickens, with a very wide margin of safety.
- ▶ Avoids exposure of house workers and birds to toxic sprays.
- ▶ Short 13-day withdrawal for meat or offal.
- ▶ Fulfills a critical unmet need for overall animal health and welfare in several regions including Latin America, Africa, Middle East and Asia etc.
- ▶ High product quality and research-based technical support from MSD Animal Health.



SECTION EIGHT

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SECTION NINE

LABEL SUMMARY





SUMMARY OF PRODUCT CHARACTERISTICS (NON EU SPC)

1. NAME OF THE VETERINARY MEDICINAL PRODUCT

Exzolt 10 mg/ml solution for use in drinking water for chickens

2. QUALITATIVE AND QUANTITATIVE COMPOSITION

Active substance:

Each ml contains 10 mg fluralaner.

For a full list of excipients, see section 6.1.

3. PHARMACEUTICAL FORM

Light yellow to yellowish-brown solution for use in drinking water.

The colour of Exzolt may change slightly over time due to the presence of the antioxidant (α -tocopherol).

4. CLINICAL PARTICULARS

4.1 TARGET SPECIES

Chickens.

4.2 INDICATIONS FOR USE, SPECIFYING THE TARGET SPECIES

Treatment and control of Poultry Red Mite (*Dermanyssus gallinae*) or Northern Fowl Mite (*Ornithonyssus sylviarum*) infestation in pullets, breeders and layer hens.

4.3 CONTRAINDICATIONS

None.

4.4 SPECIAL WARNINGS

Care should be taken to avoid the following practices because they increase the risk of development of resistance and could ultimately result in ineffective therapy:

- Too frequent and repeated use of acaricides from the same class, over an extended period of time.
- Underdosing, which may be due to underestimation of body weight, misadministration of the product, or lack of calibration of the volume measuring device.

4.5 SPECIAL PRECAUTIONS FOR USE

Special precautions for use in animals

Strict biosecurity measures at house and farm level should be implemented to prevent re-infestation of treated houses. To ensure long term control of the mite populations in a treated house, it is essential to treat any other infested poultry house in proximity to the treated one.

Special precautions to be taken by the person administering the veterinary medicinal product to animals

The veterinary medical product may be slightly irritating to skin and/or eyes.

Avoid contact with skin, eyes and mucous membranes.

Do not eat, drink or smoke while handling the product.

Wash hands and contacted skin with soap and water after use of the product.

In case of eye contact, immediately rinse thoroughly with water.

Remove clothes affected by the spillage.

4.6 ADVERSE REACTIONS (FREQUENCY AND SERIOUSNESS)

None known.

4.7 USE DURING PREGNANCY, LACTATION OR LAY

The safety of the veterinary medicinal product has been demonstrated in layers and breeders. The product can be used during lay.

In clinical field studies in layer hens, it was shown that treatment of mite infestation was usually associated with an increased laying rate.

4.8 INTERACTION WITH OTHER MEDICINAL PRODUCTS AND OTHER FORMS OF INTERACTION

None known. In clinical field trials, no interaction was observed with accompanying routine treatment such as vaccination, deworming or supplementing with nutritional additives.

4.9 AMOUNTS TO BE ADMINISTERED AND ADMINISTRATION ROUTE

For use in drinking water.

The dose is 0.5 mg fluralaner per kg body weight (equivalent to 0.05 ml Exzolt) administered twice, 7 days apart. The full course of therapy must be administered for a full therapeutic effect.

Determine the time period (between 3 and 24 hours) to administer the treatment on each treatment day. This period of time must allow all birds to receive the required dose. Estimate how much water birds will consume during treatment based on the previous day's water consumption.

Calculate the volume of Exzolt needed based on the total weight of the bird group to be treated. To ensure administration of the correct dose, body weight should be determined as accurately as possible and an accurate measuring device should be used for measuring the calculated volume of the product to be administered.

The required volume of product for each treatment day is calculated from the total body weight (kg) of the entire group of chickens to be treated:

ml product/treatment day = Total body weight (kg) of chickens to be treated x 0.05 ml

500 ml of Exzolt treats 10,000 kg body weight (e.g., 5,000 chickens of 2 kg body weight each) per day of treatment administration.

The instructions below need to be followed, in the order described, to prepare the medicated water:

- Check the water system to ensure it works properly and is free of leaks; also ensure that water is available to all nipple or bell drinkers.
- For each day of treatment, medicated water must be freshly prepared.
 - Mix the required volume of Exzolt into a large medication tank or with additional water to create a stock solution. The solution should be dosed by proportioner or dosing pump. Always add product and water

simultaneously in order to avoid foaming. It is important to rinse the container used to measure the required product volume during the filling phase in order to ensure that the complete dose is emptied into the medication tank or the stock solution and that no residues remain in the measuring device. Stir the stock solution or the content of the medication tank gently until the medicated water is homogeneous. Connect the medication tank or the proportioner or dosing pump to the drinking water system.

- Make sure the dosing pump is properly set to deliver the medicated water during the predetermined treatment period (hours per day).
- Prime the drinker lines with medicated water and check to see when medicated water has reached the end of the line. This procedure should be repeated on each day of administration.

After each treatment administration, fill the stock solution container with clean (unmedicated) water to rinse the water lines.

4.10 OVERDOSE (SYMPTOMS, EMERGENCY PROCEDURES, ANTIDOTES), IF NECESSARY

No adverse reactions were observed following the treatment of 3-week old and adult chickens dosed with up to 5 times the recommended dose for 3 times the recommended duration of treatment.

No negative effects on egg production was observed when layer hens were treated with up to 5 times the recommended dose for 3 times the recommended duration of treatment.

There were no adverse effects on reproductive performance when breeding chickens were treated at 3 times the recommended dose for 2 times the recommended duration of treatment.

4.11 WITHDRAWAL PERIOD(S)

Meat and offal: 13 days.

Eggs: zero days.

5. PHARMACOLOGICAL PROPERTIES

Pharmacotherapeutic group: Ectoparasiticides for systemic use.

ATCvet code: QP53BE02.

5.1 PHARMACODYNAMIC PROPERTIES

Fluralaner is an acaricide and an insecticide. It is efficacious against poultry mites, including *Dermanyssus gallinae* (Poultry Red Mite) and *Ornithonyssus sylviarum* (Northern Fowl Mites). Fluralaner has a high potency against mites, mostly by exposure via feeding, i.e. it is systemically active on target parasites.

Fluralaner is a potent inhibitor of parts of the arthropod nervous system by acting antagonistically on ligand-gated chloride channels (GABA-receptor and glutamate-receptor). In molecular on-target studies on insect gamma-aminobutyric acid (GABA) receptors of flea and fly, fluralaner is not affected by dieldrin resistance.

The onset of activity against *Dermanyssus gallinae* is within four hours of mites starting to feed on treated chickens.

All mites feeding on treated chickens are killed for at least two weeks after the first Exzolt administration. Fluralaner stops the production of eggs by female mites.

The mite life cycle is broken due to the rapid onset of activity of fluralaner, to the very high efficacy in killing mites feeding on treated chickens for a duration of at least two mite life cycles, and to the absence of egg production from female mites exposed to treated chickens.

Exzolt contributes towards the long term control of the mite populations in the poultry house. In clinical field studies, it was shown that treatment quickly eliminated *Dermanyssus gallinae* populations in treated houses, and that parasite population regrowth was controlled when proper biosecurity measures were implemented.

In vitro bio-assays show that fluralaner is effective against parasites having proven field resistance, including amidines (tick), organophosphates (tick,

mite), cyclodienes (tick, flea, fly), macrocyclic lactones (sea lice), phenylpyrazoles (tick, flea), benzophenyl ureas (tick), pyrethroids (tick, mite) and carbamates (mite).

5.2 PHARMACOKINETIC PARTICULARS

After oral administration, fluralaner is absorbed rapidly, reaching maximum plasma concentrations 36 hours after the first dosing and 12 hours after the second dosing following start of administration via medicated drinking water. The bioavailability is high, with approximately 90% of the dose absorbed following oral administration. Fluralaner is highly bound to protein. Fluralaner is widely distributed throughout the body, with the highest concentrations reported in the liver and skin/fat. No significant metabolites are observed in chickens, and fluralaner is mainly eliminated via the hepatic route. The apparent elimination half-life is approximately 4 days following oral administration.

6. PHARMACEUTICAL PARTICULARS

6.1 LIST OF EXCIPIENTS

α-tocopherol

Diethylene glycol monoethyl ether

Polysorbate 80

6.2 INCOMPATIBILITIES

In the absence of compatibility studies, this veterinary medicinal product must not be mixed with other veterinary medicinal products.

6.3 SHELF LIFE

Shelf life of the veterinary medicinal product as packaged for sale: 3 years

Shelf life after first opening the immediate packaging: 1 year

Shelf life of the medicated drinking water: 24 hours

6.4. SPECIAL PRECAUTIONS FOR STORAGE

Do not store above 30°C.

6.5 NATURE AND COMPOSITION OF IMMEDIATE PACKAGING

HDPE bottle closed with an aluminum foil seal and a child-resistant polypropylene screw cap.

Pack sizes: bottles of 1 litre or 4 litres.

Not all pack sizes may be marketed.

6.6 SPECIAL PRECAUTIONS FOR THE DISPOSAL OF UNUSED VETERINARY MEDICINAL PRODUCT OR WASTE MATERIALS DERIVED FROM THE USE OF SUCH PRODUCTS

Exzolt should not enter water courses as this may be dangerous for aquatic invertebrates.

Any unused veterinary medicinal product or waste materials derived from such veterinary medicinal products should be disposed of in accordance with local requirements.

NOTES

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TREATMENT FOR POULTRY MITE INFESTATIONS IN CHICKENS

TECHNICAL MANUAL