Exzolt™
(fluralaner oral solution)

MERCK
Animal Health

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TREATMENT AND CONTROL FOR NORTHERN FOWL MITE INFESTATIONS IN CHICKENS
TECHNICAL MANUAL

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MERCK
Animal Health

BREAKTHROUGH
MITE CONTROL

Exzolt™, new from Merck Animal Health, eliminates over 99% of the northern fowl mites infestations, making it the most effective solution. Since Exzolt is applied through your drinking water system, no special training or equipment is needed.

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

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(fluralaner oral solution)

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(fluralaner oral solution)

SAFER, SIMPLER, MORE EFFECTIVE MITE CONTROL

Exzolt provides 99%+ mite elimination in three simple steps. First, deliver the initial administration through the hen's drinking water to eliminate live mites. Administration through the drinking water reduces stress caused by spraying and minimizes the potential of chemical exposure to human workers. Seven days later, apply the second administration to eradicate mite stages not susceptible to the first administration. Last, to maintain the fast and nearly complete elimination of mite populations that Exzolt provides, follow proper bio-security measures, such as:

- Avoid entrance of northern fowl mite with the allocation of new hen flocks
- Change outer clothing per house and use specific tools per each barn
- Clean and check northern fowl mite absence before being used in another house

1

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

2

SECOND ADMINISTRATION OF EXZOLT MAINTAINS THE EFFECTS OF TREATMENT TO KILL MITES AS THEY ENTER THE BLOOD-FEEDING STAGES

3

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.

NORTHERN FOWL MITES[1]

The northern fowl mite (*Ornithonyssus sylviarum*, Figure 1-1) poses a severe threat to poultry production. Fowl mites are the most common and damaging ectoparasite of poultry in the U.S.

Northern fowl mites can impact animal welfare and cause production losses. 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 MITES

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

NORTHERN FOWL MITE LIFE CYCLE

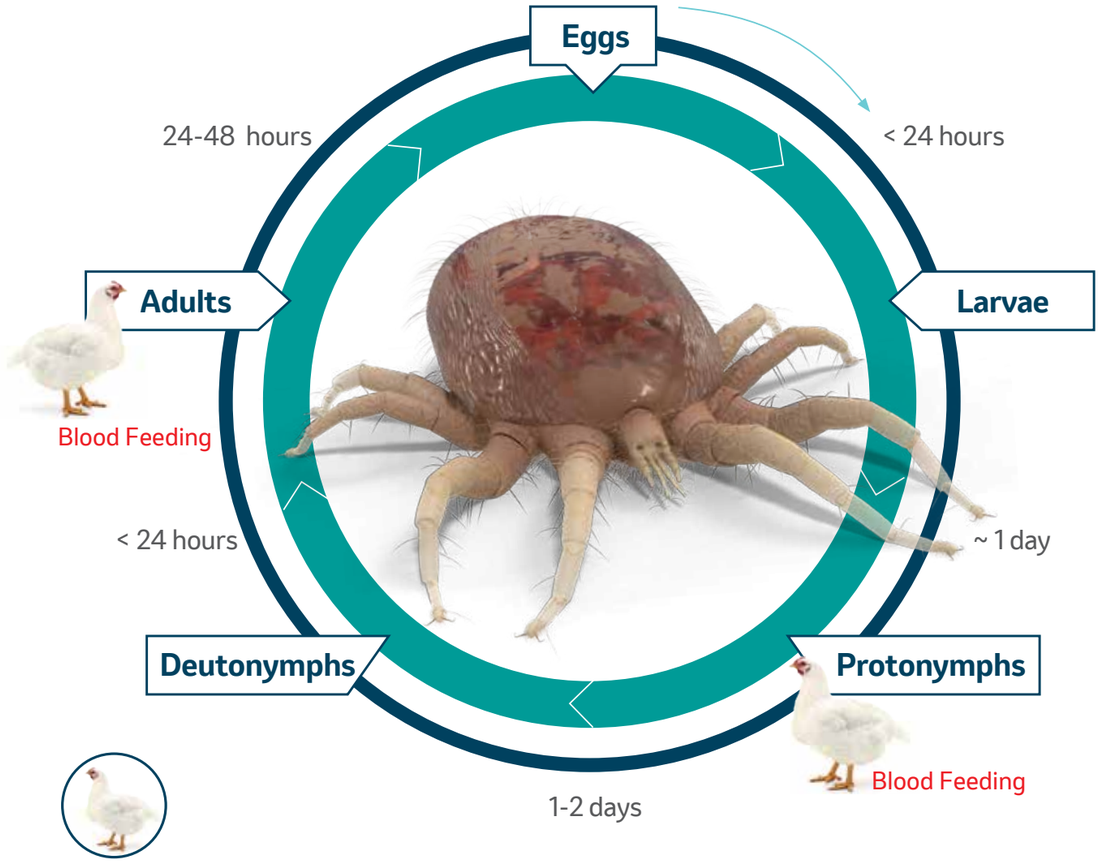


Figure 1-1: The life cycle of the northern fowl mite, *Ornithonyssus sylviarum*. Aside from the egg, fowl mites have 4 life-cycle stages: larva, protonymph, deutonymph and adult. Larvae hatch with 6 legs and do not feed. After the first molt, both nymphal stages and adults have 8 legs. Only protonymphs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

Figure 1-2: Photomicrograph of northern fowl mite adult female, *Ornithonyssus sylviarum* (100×).[1] (Used with permission.)

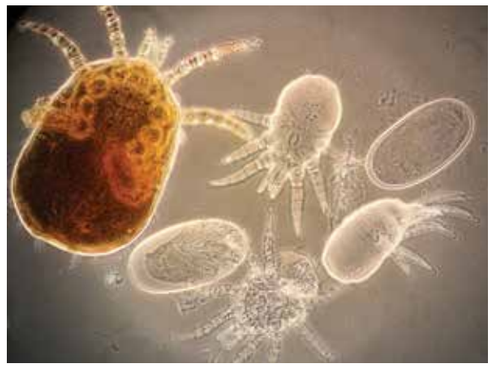


Figure 1-3: Photomicrograph of life stages of the northern fowl mite: egg (e), egg with developing larva (e-l), larvae (l), protonymph (p), and adult female.[1] (Used with permission.)

Figure 1-4: 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 an infestation on a new host.

The entire northern fowl mite (*O. sylviarum*) life cycle is completed on birds. Thus, chickens are infested with all northern fowl mite development stages (Figure 1-2), 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.

The direct northern fowl mite life cycle requires 5 to 12 days to complete (Figure 1-3). Because the life cycle can be as short as 5 days under optimal conditions, rapid growth of mite populations can occur. Northern 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. Northern 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 northern 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 1-4). 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, northern 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 1-5: 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.[2]

NORTHERN FOWL MITE IMPACT

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 northern 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.[3] 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.

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 1-5), equipment and poultry workers. In fact, visual 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.

Northern fowl mites to date have not been implicated as important pathogen vectors. 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).[1]

CONTROL ATTEMPTS

Successful integrated pest management programs for poultry should focus first on preventive 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 include chemical methods (e.g., pesticide sprays) and a variety of alternative strategies (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.[4]

Application of materials intended for northern fowl mite control need to be applied on the bird to be partially effective. On most large-scale production operations in the U.S., northern fowl mites are typically managed by spraying chemical pesticides at high pressure directly onto birds, with the spray directed upward toward the vent area. (Pesticides are most effective when applied to feathers in the vent region where mites gather.) However, the U.S. 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 northern 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 oral solution) solution for use in drinking water now offers an innovative approach for comprehensive management of northern fowl mites in chickens. As discussed in the following pages, 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.
- Northern fowl mites (*Ornithonyssus sylviarum*) are predominately found the U.S. and Latin America.
- Current methods for managing northern fowl 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.

NOTES

SECTION TWO

PRODUCT PROFILE

PRODUCT PROFILE

Exzolt is a unique parasiticide for chickens that provides potent efficacy against 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 northern fowl mite (*O. sylviarum*) infestation in chickens laying hens and replacement chickens when administered orally via drinking water twice (7 days apart) at a dose of 0.5 mg fluralaner per kg (0.227 mg/lb) body weight (BW), equivalent to 0.05 mL of Exzolt/kg body weight (0.023 mL/lb).

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 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. The bioavailability is high, with approximately 91% of the dose absorbed following oral administration. 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 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.[5] Research also showed 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).[6]

FAST, POTENT EFFICACY

Treatment of the host with Exzolt (instead of treating the host environment) is an innovative approach to targeting northern fowl mite infestations.[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 begins soon 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 proper biosecurity measures.

Available data indicate 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.

BENEFITS OF EXZOLT

Dose rate: 0.5 mg fluralaner/kg (0.227 mg/lb) 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.
- Quick kill, with prolonged duration in birds spanning two 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-day egg withdrawl 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.

SAFE FOR CHICKENS AND PEOPLE

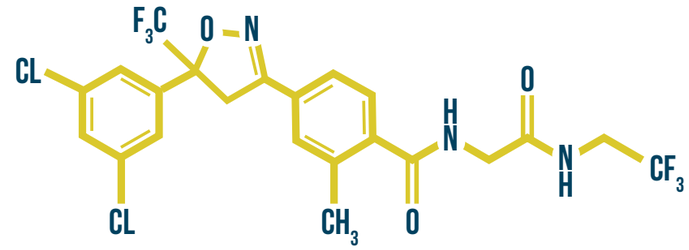
Fluralaner offers high selectivity for arthropods like poultry mites, and the formulated product has a large safety margin demonstrated in chickens. Unlike spraying, water-borne Exzolt is not stressful for hens and minimizes the potential for exposure of chemicals by human workers. Furthermore, Exzolt 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.).

NOVEL, REVOLUTIONARY EXZOLT

Exzolt provides poultry producers with a revolutionary new option for managing 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 northern fowl mites and represents an excellent therapeutic component of comprehensive health programs aimed at optimizing overall performance of pullets, breeders and layers.

FLURALANER CHEMICAL PROPERTIES

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



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

For the treatment and control of northern fowl mites (*Ornithonyssus sylviarum*) in laying hens and replacement chickens.

DOSAGE AND ADMINISTRATION

For use in drinking water.

Exzolt must be administered orally to chickens via the drinking water as 2 single doses spaced 7 days apart, with each dose consumed over a period of 6 to 24 hours. Each dose is 0.5 mg fluralaner/kg (0.227 mg/lb) body weight (BW), equivalent to 0.05 mL of Exzolt/kg body weight (0.023 mL/lb). The full course of therapy (2 single doses 7 days apart) must be administered for full therapeutic effect.

Determine the time period over which to administer the medicated water on the treatment day. This period of time must be a minimum of 6 hours and maximum of 24 hours and long enough to allow all birds to receive the required dose. **If the medicated water will contact rusty surfaces, it must be consumed within 8 hours of preparation.** Estimate how much water birds will usually consume during the selected treatment period based on the previous day's water consumption. Ensure that the amount of medicated drinking water offered will be consumed completely within the selected treatment period (between 6 and 24 hours). No other source of drinking water should be available during the medication period.

Calculate the volume of Exzolt 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 estimated as accurately as possible, and an accurate 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 or Lb) of the entire group of chickens to be treated, using the following formula:

Volume of Exzolt (mL) per treatment day = Total BW (kg) of birds to be treated x 0.05 mL/kg.

Or

Volume of Exzolt (mL) per treatment day = Total BW (lb) of birds to be treated x 0.023 mL/lb.

Therefore 500 mL of product treats 10,000 kg or 22,000 pounds BW (e.g., 5,000 chickens of 2 kg or 4.4 lb BW each) per day of treatment administration.

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

WITHDRAWAL PERIODS

Eggs: zero (0) days
Meat: 11 days

CONTRAINDICATIONS, ADVERSE REACTIONS, INTERACTIONS

None known.

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

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

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.

USER PRECAUTIONS

Protective gloves should be used. Care should be taken when handling the product to avoid skin and eye exposure, exposure of mucous membranes and accidental ingestion. Accidental exposure may cause skin and eye irritation. In case of eye contact, immediately rinse thoroughly with water. If wearing contact lenses, immediately rinse the eyes first, then remove contact lenses and continue to rinse the eyes thoroughly. Seek medical advice if symptoms occur. Wash hands and contacted skin with soap and water after use of the product. Remove contaminated clothes and launder with detergent.

PACKAGING/STORAGE/EXPIRATION

Exzolt is available in 1- and 4-liter HDPE plastic containers.

Store Exzolt at or below 86°F (30°C) and use within 6 months after first opening. Use the medicated water within 24 hours of preparation. **Use within 8 hours of preparation if the medicated water comes in contact with rusty surfaces.**

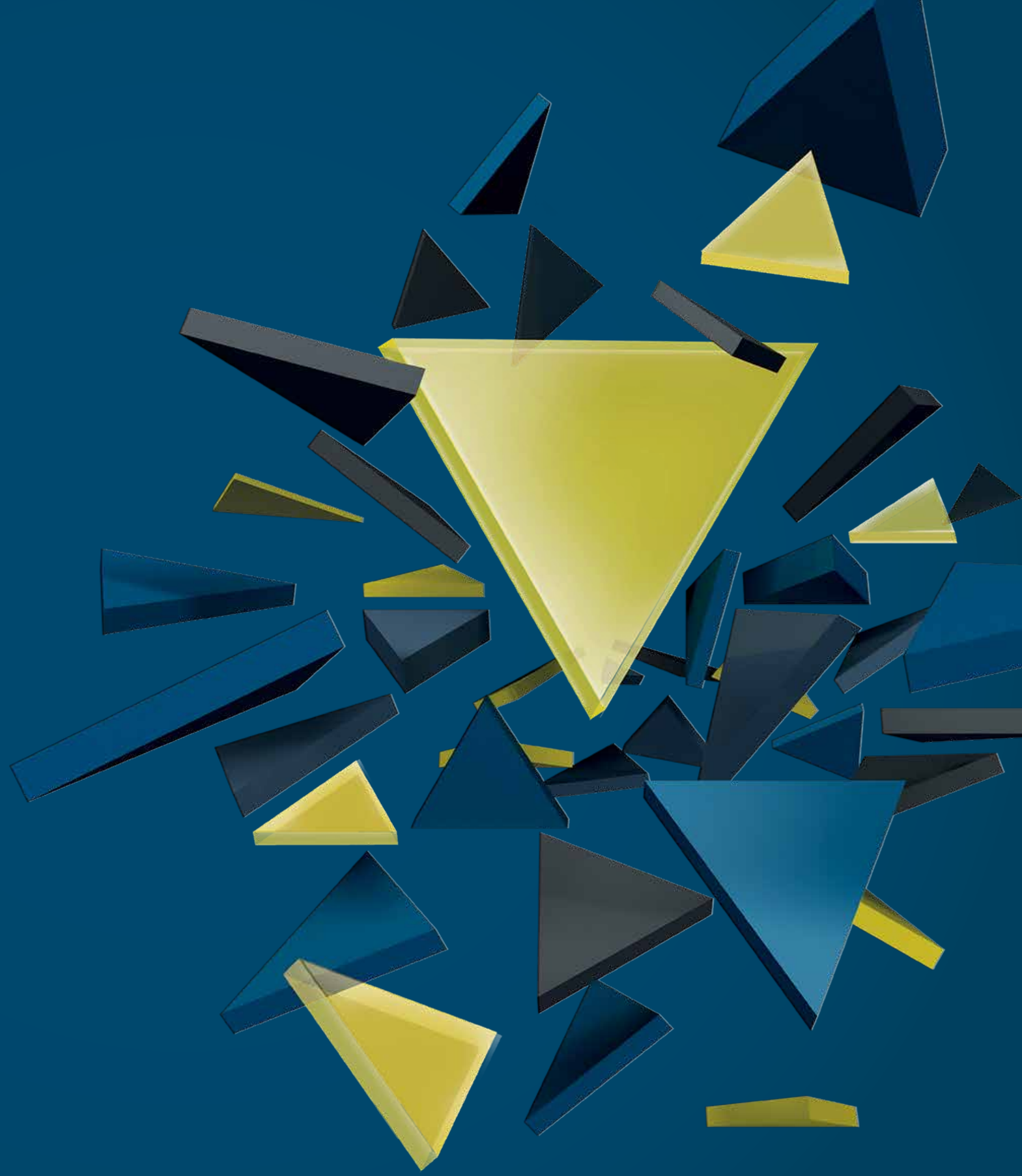
PRODUCT PROFILE SUMMARY

- Exzolt (fluralaner oral solution) represents a completely new approach for comprehensive management of 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 humans.
- Exzolt is indicated for the treatment and control of northern fowl mites (*Ornithonyssus sylviarum*) in laying hens and replacement chickens when provided in drinking water at 0.5 mg fluralaner/kg (0.227 mg/lb) BW, administered twice, 7 days apart.
- Exzolt has a zero-day egg withdrawal for chicken eggs and a 11-day withdrawal period for meat, with no contraindications, known adverse effects or known interactions.

NOTES

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).[6] Blood samples were collected pre-treatment and at 17 post-treatment time points (5 minutes through 35 days). Fluralaner displayed a slow elimination profile (T1/2 of about 5 days after intravenous administration), dose linearity and 91% oral bioavailability.

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.[6]

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 Cmax 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 the 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.

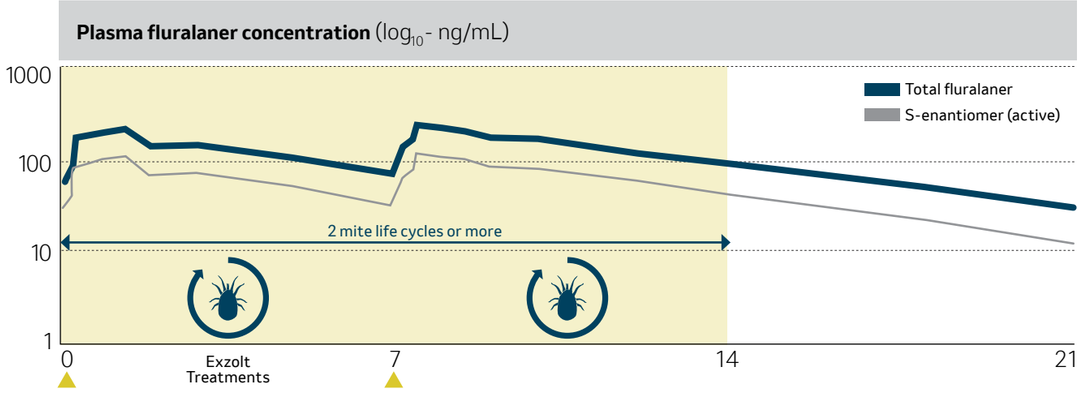


Figure 3-1: Fluralaner plasma concentration; time profile after Exzolt administration in chickens at 0.5 mg/kg BW in drinking water, twice at a 7-day interval.

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, 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.[7] 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).

Additional in vitro research found that pyrethroid-resistant northern fowl mites were extremely sensitive to fluralaner.[8] The test isolate was resistant to pyrethroids (permethrin, deltamethrin) despite not being exposed to acaricides for at least 10 years. Still, the LC99 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 7 days apart) was designed to ensure effective treatment and elimination of all mite development stages from the flock, which limits resistance development. 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. The apparent elimination half-life is approximately 5 days following oral administration.
- Fluralaner is a very potent acaricide, acting extremely fast and for at least 15 days when administered to chickens by the recommended Exzolt dosage regimen.
- 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.

NOTES

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. Two reproductive safety studies were also conducted, evaluating safety in both layer breeder chickens and broiler breeder chickens. Outcomes obtained in these studies support the safety of Exzolt in laying hens and replacement chickens, 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).[6,9]

- 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 the 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 (5 times) group.
- Data collections were performed by personnel blinded to treatment.

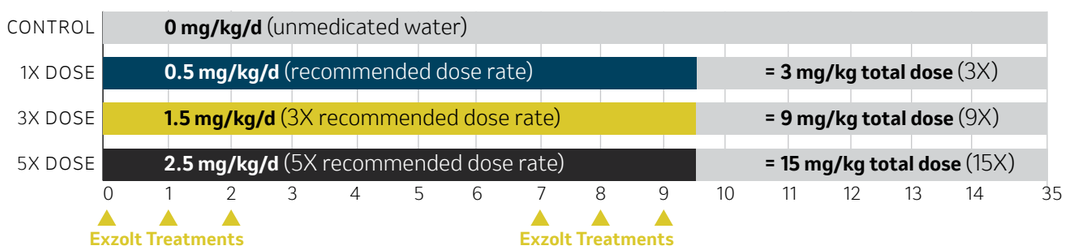


Figure 4-1: Experiment design of target animal safety study for laying hens.

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 5 times 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).[6]

- 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 3, 9 or 15 times 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.
- 10 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 (5 times) group.
- Data collections were performed by personnel blinded to treatment.

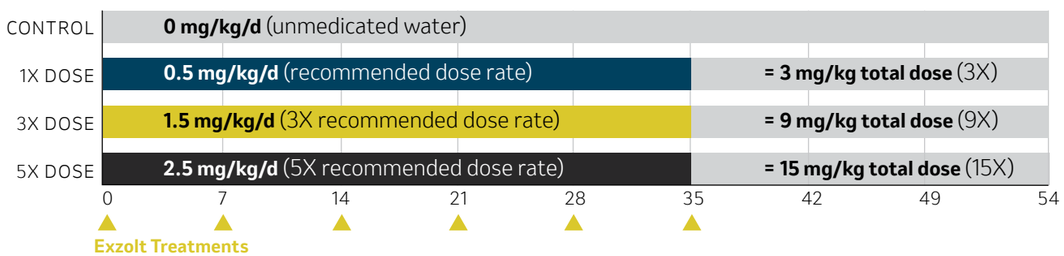


Figure 4-2: Experiment design of target animal safety study for 3-week-old chicks.

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 — LAYER BREEDER CHICKENS

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).[6,10]

- 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 of (3 times) 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.

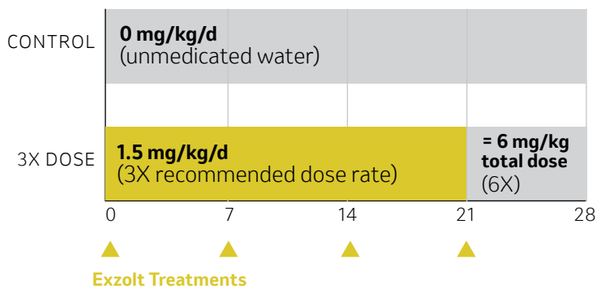


Figure 4-3: Experiment design of reproductive safety studies for layer breeders and broiler breeders.

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 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.

REPRODUCTIVE SAFETY — BROILER BREEDER CHICKENS

STUDY DESIGN

A study assessed the reproductive safety of Exzolt when administered via drinking water to broiler breeder chickens (Cobb 500 Fast Feathering Strain) at their time of approximate peak egg production (~33 weeks).[6] (The study design was nearly identical to the previous layer breeder study.)

- 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, double 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 broiler 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 nonmedicated controls.
- The treatment period extended for at least 1 estrous cycle in females, complete egg formation and a full 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.

RESULTS

No adverse impact of treatment with Exzolt was detected in regard to adult bird body weight, feed consumption, egg production, egg weights, fertility, hatchability, 14-day chick weights and 14-day chick viability. Furthermore, gross and microscopic examinations revealed no tissue alterations or changes in weights of reproductive organs attributable to Exzolt administration. Water intake was lower than controls in the overdosed Exzolt group, but the negligible difference was not considered clinically relevant compared to the day-to-day variation in water consumption within each treatment group.

CONCLUSIONS

This reproductive safety study conducted in broiler 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 broiler breeder chickens and does not substantially 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 11 days after last administration of Exzolt was computed for meat, 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 has been shown to pose no loss of egg value or income, whether for human consumption or for hatch.

SAFETY SUMMARY

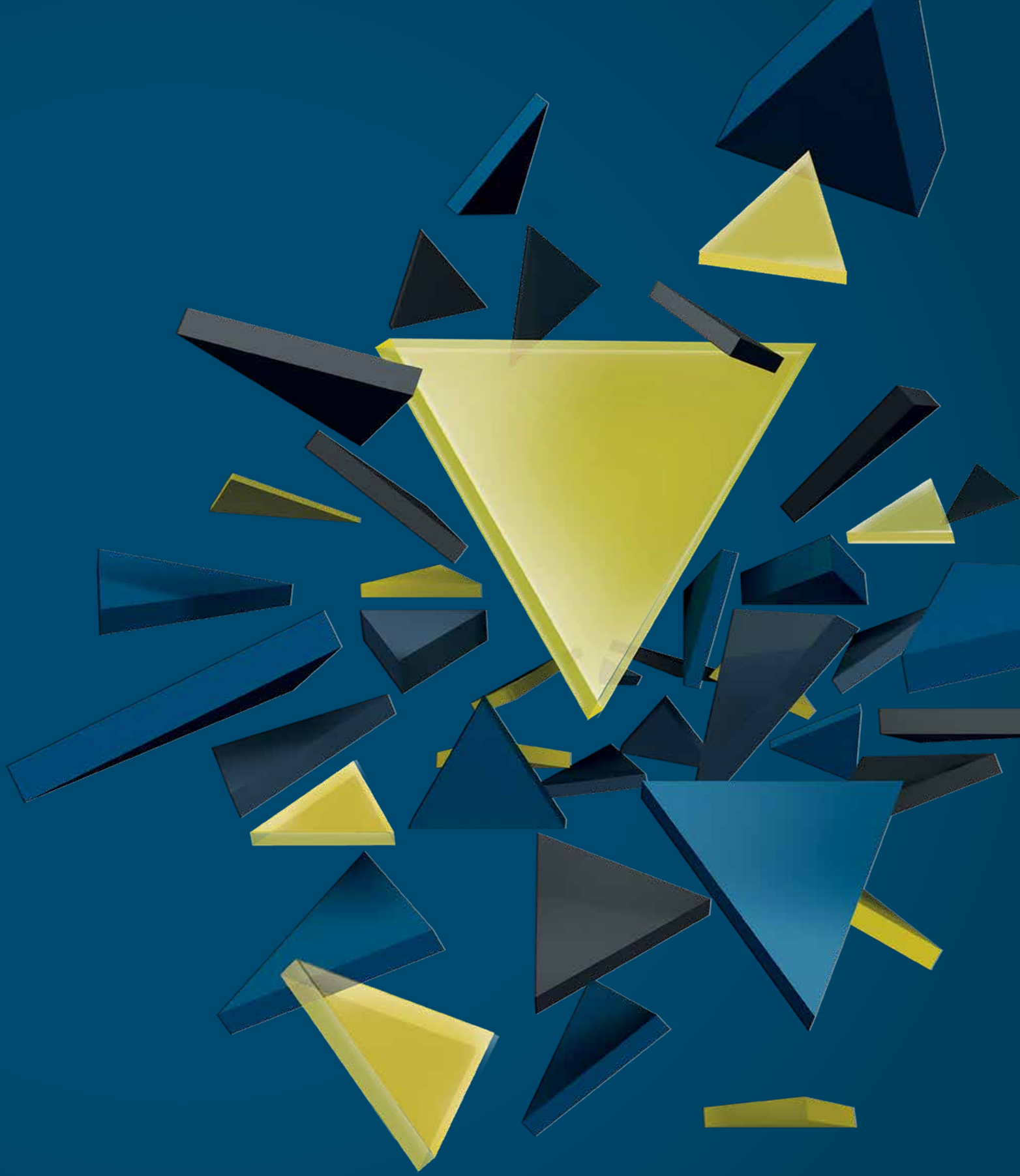
- Two target animal safety studies demonstrated that Exzolt was well tolerated and palatable in both very young birds as well as adult hens under high physiological stress related to intensive egg production, even when dosed at 5 times the recommended dose for 3 times the recommended duration of treatment (no adverse impacts on health, egg production or growth performance).
- Reproductive safety studies demonstrated that Exzolt is well tolerated in layer breeder and broiler 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 laying hens and replacement chickens 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 11 days after last administration of Exzolt is required for human consumption of meat.

BREAKTHROUGH
WELFARE

Exzolt™
(fluralaner oral solution)

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.

Research investigated the activity and efficacy of Exzolt against northern fowl mites (*O. sylviarum*). An in vitro study confirmed the susceptibility of northern fowl mites to fluralaner, and 2 dose-confirmation studies, 2 field-efficacy studies and one natural-infestation-efficacy study documented the efficacy and duration of the 2-dose Exzolt regimen against northern fowl mite infestations in chickens.

EFFICACY AGAINST 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.[8]

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, removal of dead and live mites and eggs already deposited by the mites were counted.
- Packets were then resealed and held for an additional 72 hours, after which immature stages were counted to determine egg hatch or larval-protonymph survival.
- LC50 and LC99 values were calculated for adult mortality (probit analysis).

RESULTS

Mortality outcomes summarized in Table 5-1 indicated the LC99 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

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) and 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.

ACARACIDE	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 5-1: In vitro susceptibility of adult northern fowl mites (*O. sylviarum*) to fluralaner and other acaricides.

DOSE CONFIRMATION STUDY – LAYING HENS

A study was conducted in the U.S. to confirm the effectiveness of the Exzolt dosage regimen (0.5 mg fluralaner/kg BW in drinking water twice at a 7-day interval) for treating infestations of northern fowl mites (*O. sylviarum*).[6] Conducted at a university research facility, the trial evaluated Exzolt dose efficacy in laying hens.

ANIMALS
128 laying hens
29 weeks of age

STUDY DESIGN

A controlled, single-site dose-confirmation study was performed. Birds were housed in a small-scale poultry house at a major western U.S. university. Hens were obtained as uninfested birds and, following a brief acclimation period, were infested with northern fowl mites.

Healthy birds with established mite infestation were ranked by mite counts on day -7 and then randomly assigned to 32 cages (4 birds/cage each) with 16 cages randomly assigned to each of 2 treatment groups (n=64 birds/group).

On day 0, birds in one 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.

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. The control group had an adequate infestation through day 14 but not on days 19 or 28.

EFFICACY CRITERIA

Efficacy assessment was based on the percent reduction of mites on treated birds compared to untreated controls.

RESULTS

The efficacy of Exzolt compared to controls is summarized in Figure 5-1. Early onset of action was noted as mite vent counts were significantly reduced 99.9% in the Exzolt group beginning at day 2 ($P < 0.0001$). Similarly, significant mite count reductions ($P < 0.0001$) were observed on subsequent sample days 8 and 14. No adverse events related to the use of Exzolt were observed.

Exzolt efficacy vs. northern fowl mites
(% reduction vs. controls)

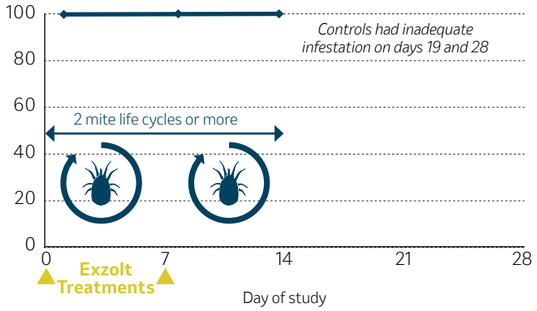


Figure 5-1: Efficacy of Exzolt (0.5 mg fluralaner/kg BW twice 7 days apart) for reducing counts of northern fowl mites (*O. sylviarum*) compared to controls in a dose-confirmation study for laying hens.

CONCLUSIONS

This dose-confirmation study verified that Exzolt provided high (100%) efficacy against northern fowl mite infestations of laying 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 the target efficacy duration spanning 2 development cycles for northern fowl mites.

DOSE CONFIRMATION STUDY – REPLACEMENT CHICKENS

A 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.[2,6]

ANIMALS

132 replacement chickens
14 weeks of age

STUDY DESIGN

- A controlled, single-site dose-confirmation study was performed. Birds were housed in a small-scale poultry house at a major California 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, and on day -6 were randomly assigned to 32 cages (4 birds/cage each), with 16 cages assigned to each of 2 treatment groups.
- On day 0, birds in one 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 for the 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 compared to untreated controls.

RESULTS

The efficacy of Exzolt compared to controls is summarized in Figure 5-2. Early onset of action was noted as mite vent counts were significantly reduced 99.3% in the Exzolt group beginning at day 2 ($P < 0.0001$). Similarly, significant 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.

Exzolt efficacy vs. northern fowl mites
(% reduction vs. controls)

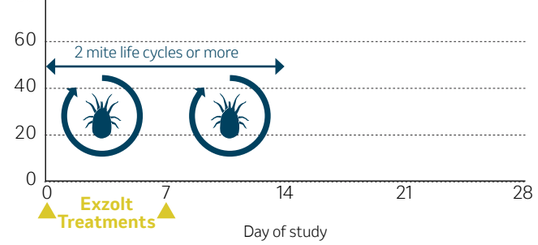


Figure 5-2: Efficacy of Exzolt (0.5 mg fluralaner/kg BW twice 7 days apart) for reducing counts of northern fowl mites (*O. sylviarum*) compared to controls in a dose-confirmation study for replacement chickens..

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 IN LAYERS – STUDY 1

A field study was conducted at a university facility in California to evaluate the efficacy of Exzolt (0.5 mg fluralaner/kg BW in drinking water twice at a 7-day interval) for treating infestations of northern fowl mites (*O. sylviarum*) in laying hens derived from a commercial poultry farm in California.[6]

ANIMALS

800 commercial White Leghorn chickens (Hy-Line)
23 weeks of age

STUDY DESIGN

- A controlled, single-site field efficacy study was performed. Commercial laying hens were obtained as uninfested birds and, following a brief acclimation period, were artificially 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 (cages/group).
- On day 0, birds in one 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 compared to untreated controls.

RESULTS

The efficacy of Exzolt compared to controls is summarized in Figure 5-3. Exzolt provided early onset of action as mite vent counts were significantly reduced 99.5% beginning at day 2. Mite count reductions of >99.9% 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.

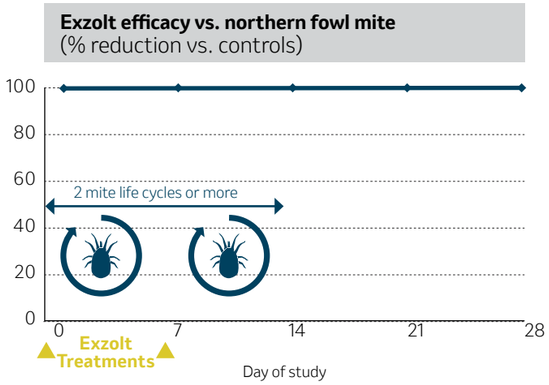


Figure 5-3: Efficacy of Exzolt (0.5 mg fluralaner/kg BW twice 7 days apart) for reducing counts of northern fowl mite (*O. sylviarum*) compared to controls in a U.S. field efficacy study in laying hens.

CONCLUSIONS

This U.S. field efficacy study demonstrated that Exzolt provided high (>99.9%) 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 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.

FIELD EFFICACY IN LAYERS – STUDY 2

A second field study was conducted at a major university in Pennsylvania to evaluate the efficacy of Exzolt (0.5 mg fluralaner/kg BW in drinking water twice at a 7-day interval) for treating infestations of northern fowl mites (*O. sylviarum*) in laying hens.[6]

ANIMALS

800 White Leghorn chickens (Hy-Line), 28 weeks of age

STUDY DESIGN

- A controlled, single-site natural infestation efficacy study was performed. Laying hens were obtained as uninfested birds and, following a brief acclimation period, were artificially infested with northern fowl mites.
- 800 healthy birds with established mite infestation were randomly placed into 400 cages (2/cage). On day -3, cages were randomly assigned to either of 2 treatment groups (200 cages/group).
- On day 0, birds in one group were treated with Exzolt (0.5 mg/kg BW/day twice 7 days apart) using a gravity tank system, while the other group remained untreated (controls). Exzolt dosage was based on total 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 compared to untreated controls.

RESULTS

The efficacy of Exzolt compared to controls is summarized in Figure 5-4. Exzolt provided early onset of action as mite vent counts were significantly reduced 99.6% beginning at day 2. Mite count reductions >99.9% 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.

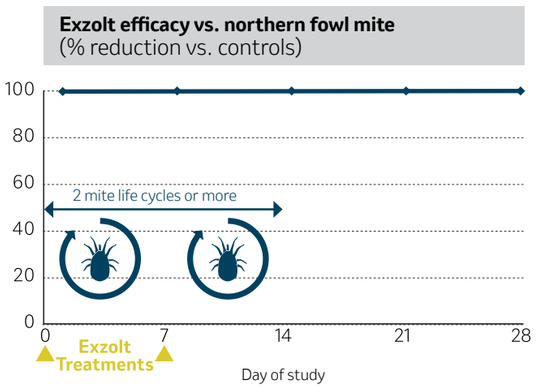


Figure 5-4: Efficacy of Exzolt (0.5 mg fluralaner/kg BW twice 7 days apart) for reducing counts of northern fowl mite (*O. sylviarum*) compared to controls in a U.S. field efficacy study in laying hens.

CONCLUSIONS

This U.S. field efficacy study demonstrated that Exzolt provided high (>99.9%) 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 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.

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.[12]

ANIMALS

60 commercial laying hens

STUDY DESIGN

- A controlled, single-site natural infestation efficacy study was performed. 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 one 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 compared to untreated controls.

RESULTS

The efficacy of Exzolt compared to controls is summarized in Figure 5-5. 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 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.

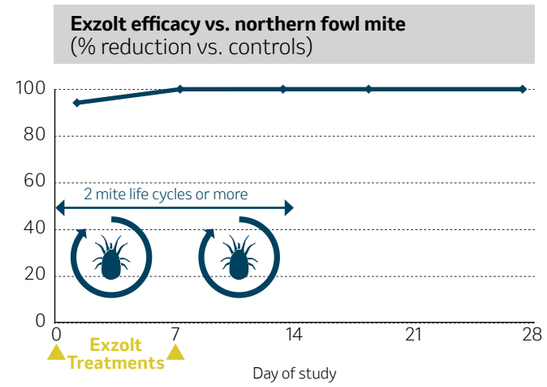


Figure 5-5: Efficacy of Exzolt (0.5 mg fluralaner/kg BW twice 7 days apart) for reducing counts of northern fowl mite (*O. sylviarum*) compared to controls in a Brazilian natural infestation efficacy study.

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.

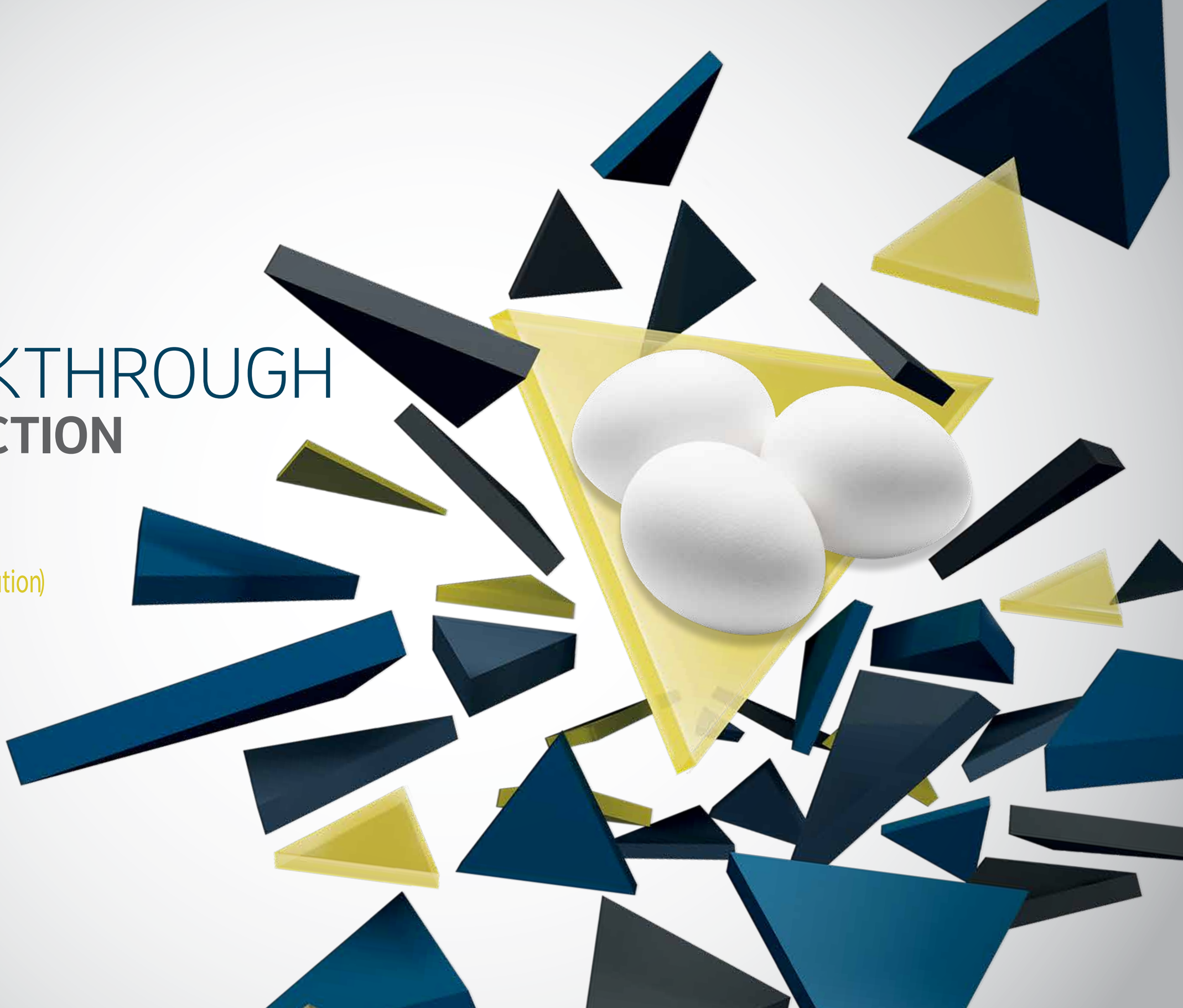
EFFICACY SUMMARY

- The 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.
- Northern fowl mites were found to be extremely sensitive to fluralaner in vitro. Two dose-confirmation studies verified Exzolt efficacy against northern fowl mite. Furthermore, 3 field efficacy studies demonstrated fast efficacy via Exzolt 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.
- Exzolt clearly offers potent, consistent, convenient and safe efficacy against northern fowl mite and thus helps prevent the significant adverse economic consequences of inadequately controlled mite infestation.

NOTES

BREAKTHROUGH
PRODUCTION

Exzolt™
(fluralaner oral solution)



SECTION SIX

USAGE GUIDELINES

USAGE GUIDELINES

DOSE CALCULATION

The dose for Exzolt is 0.5 mg fluralaner per kg (0.227 mg/lb) BW per day (equivalent to 0.05 mL Exzolt, or 0.023 mL/lb) administered twice, 7 days apart. The full course of therapy must be administered for full therapeutic effect.

Determine the time period over which to administer the medicated water on the treatment day. This period of time must be a minimum of 6 hours and maximum of 24 hours and long enough to allow all birds to receive the required dose. **If the medicated water will contact rusty surfaces, it must be consumed within 8 hours of preparation.** Estimate how much water birds will usually consume during the selected treatment period based on the previous day's water consumption.

Calculate the volume of Exzolt needed based on the total weight of all birds in the house to be treated. To ensure administration of the correct dose, body weight should be estimated as accurately as possible, and an accurate device should be used for measuring the calculated volume of product to be administered. The required amount of Exzolt on each treatment day should be calculated from the total body weight of the entire group of chickens to be treated:

Volume of Exzolt (mL) per treatment day = Total BW (kg) of birds to be treated x 0.05 mL/kg.

Or

Volume of Exzolt (mL) per treatment day = Total BW (lb) of birds to be treated x 0.023 mL/lb.

Examples:

TOTAL BODY WEIGHT OF BIRDS TO BE TREATED	VOLUME OF EXZOLT PER TREATMENT DAY (ML/DAY)
5,000 kg (11,023 lb)	0.25 L (250 mL)
10,000 kg (22,046 lb)	0.5 L (500 mL)
80,000 kg (176,370 lb)	4 L
320,000 kg (705,479 lb)	16 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 water is available to all nipple or bell drinkers.
- For each day of treatment, medicated water must be freshly prepared.
  - Add predetermined amount of fresh water into stock solution container. Add calculated amount of Exzolt to the fresh water in stock solution container. Rinse measuring device to ensure remaining Exzolt is added to stock solution. Stir stock solution until medicated water is homogeneous. Drop hose into stock solution and start proportioner (set at 1:128). For bulk medication tanks, follow the same sequence.
- Make sure the dosing pump is properly set to deliver the 1:128 ratio of medicated water during the predetermined treatment period (hours).
- Prior to introducing the medicated water, drain the drinker lines fully by opening the flush valve and checking end-line nipples to ensure no water remains. Prime the drinker lines with medicated water and confirm the medicated water has reached all end-line nipples.

CONSIDERATIONS FOR USE OF PROPORTIONER

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 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 oral solution)

SECTION SEVEN

SUMMARY

SUMMARY

Exzolt represents an innovative, completely new approach for comprehensive management of 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 soon after 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 (0.227 mg/lb) 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-day 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 11-day withdrawal for meat.
- High product quality and research-based technical support from Merck Animal Health.

SECTION EIGHT

REFERENCES

REFERENCES CITED

[1] Murillo AC, Mullens BA. A review of the biology, ecology, and control of the northern fowl mite, *Ornithonyssus sylviarum* (Acari: Macronyssidae). Vet Parasitol 2017; 246:30-37.

[2] DeLoach JR, DeVaney JA. Northern fowl mite ingests large quantities of blood from white leghorn hens. *J Med Entomol* 1981; 18:374-377.

[3] Mullens BA, Owen JP, Kuney DR, Szijj CE, Klingler KA. Temporal changes in distribution, prevalence and intensity of northern fowl mite (*Ornithonyssus sylviarum*) parasitism in commercial caged laying hens, with a comprehensive economic analysis of parasite impact. Vet Parasitol 2009; 160:116-133.

[4] Mullens BA, Velten RK, Hinkle NC, Kuney DR, Szijj CE. Acaricide resistance in northern fowl mite (*Ornithonyssus sylviarum*) populations on caged layer operations in southern California. Poultry Sci 2004; 83:365-374.

[5] Gassel M, Wolf C, Noack S, Williams H, Ilg T. The novel isoxazoline ectoparasiticide fluralaner: selective inhibition of arthropod γ-aminobutyric acid- and L-glutamate-gated chloride channels and insecticidal/acaricidal activity. *Insect Biochem Molec* Biol 2014; 45:111-124.

[6] Data on file. Merck Animal Health.

[7] Casida JE. Golden age of RyR and GABA-R diamide and isoxazoline insecticides: common genesis, serendipity, surprises, selectivity, and safety. *Chem Res Toxicol* 2015; 28:560-566.

[8] Mullens BA, Murillo AC, Zoller H, Heckeroth AR, Jirjis F, Flochlay-Sigognault A. Comparative in vitro evaluation of contact activity of fluralaner, spinosad, phoxim, propoxur, permethrin and deltamethrin against the northern fowl mite, *Ornithonyssus sylviarum*. Parasites & Vectors 2017; 10:358.

[9] Prohaczik A., Menge M, Huyghe. B, Flochlay-Sigognault A. and Le Traon G. Safety of fluralaner oral solution, a novel systemic antiparasitic treatment for chickens, in laying hens after oral administration via drinking water. *Parasit Vectors*. 2017; 8:363.

[10] Huyghe B, Le Traon G, Flochlay-Sigognault A. Safety of fluralaner oral solution, a novel systemic poultry red mite treatment, for chicken breeders' reproductive performances. *Parasit Vectors*. 2017; 10:540.

[11] Hinkle NC, Jirjis F, Szewczyk E, Sun F, Flochlay-Sigognault A. Efficacy and safety assessment of a water-soluble formulation of fluralaner for treatment of natural *Ornithonyssus sylviarum* infestations in laying hens. *Parasites & Vectors* 2018; 11:99.

[12] Jirjis F, Vettorato L, Chiummo R, Szewczyk E, Flochlay Sigognault A. Efficacy and safety of fluralaner administered orally against northern fowl mite (*Ornithonyssus sylviarum*) infestations in layers. *Amer Assoc Avian Path*, Indianapolis, July 2017.

NOTES

SECTION NINE

LABEL SUMMARY

Exzolt™
(fluralaner oral solution)

Exzolt is indicated for the treatment and control of northern fowl mites (*Ornithonyssus sylviarum*) in laying hens and replacement chickens.

ADVANTAGES*:

- Rapid and massive decrease in mite populations in a chicken house, with demonstrated 99%+ efficacy.*
- Full treatment by convenient application via drinking water 2 times, 7 days apart.
- Ready-to-use solution for simple dilution, with no sedimentation, clogging or spoilage.
- Zero-day egg withdrawal period, ideal for commercial layers.
- 11-day meat withdrawal after last administration of Exzolt.
- More effective, safe, targeted and convenient than mite sprays.
- Fast onset of acaricidal activity, with 99%+ reduction in northern fowl mite counts after just 2 days. Full treatment requires 2 administrations, 7 days apart, which spans 2 mite life cycles, disrupting mite population dynamics.

Exzolt is a fluranlaner oral solution for control of northern fowl mites available in 1 L and 4 L presentations.

IMPORTANT SAFETY INFORMATION:
EXZOLT (fluralaner oral solution)
Not for use in humans. Keep this and all drugs out of the reach of children. Accidental exposure may cause skin and eye irritation. Accidental ingestion may cause gastrointestinal disturbances and hypersensitivity reactions in humans. Chickens must not be slaughtered for human consumption for 11 days after the last treatment. No egg discard is required when used according to the labeling. For complete safety information and product dosing instructions, refer to the product label.

**Data on file, Merck Animal Health*

Exzolt™ (fluralaner oral solution)
10 mg of fluralaner/mL

Caution: Federal law restricts this drug to use by or on the order of a licensed veterinarian.

Description: Exzolt (fluralaner oral solution) is a concentrate for oral administration via drinking water containing fluralaner. Each mL of Exzolt contains 10 mg of fluralaner.

The chemical name of fluralaner is (±)-4-[5-(3,5-dichlorophenyl)-5-(trifluoromethyl)-4,5 dihydroisoxazol-3-yl]-2-methyl-N-[2-oxo-2-(2,2,2-trifluoroethylamino) ethyl] benzamide.

Indications for Use: Exzolt is indicated for the treatment and control of northern fowl mites (*Ornithonyssus sylviarum*) in laying hens and replacement chickens.

Dosage and Administration: Exzolt must be administered orally to chickens via the drinking water as 2 single doses spaced 7 days apart, with each dose consumed over a period of 6 to 24 hours. Each dose is 0.5 mg fluralaner/kg (0.227 mg/lb) body weight, equivalent to 0.05 mL of Exzolt/kg body weight (0.023 mL/lb).

General Mixing Directions: Determine the time period over which to administer the medicated water on the treatment day. This period of time must be a minimum of 6 hours and maximum of 24 hours and long enough to allow all birds to receive the required dose. **If the medicated water will contact rusty surfaces, it must be consumed within 8 hours of preparation.** Estimate how much water birds will usually consume during the selected treatment period based on the previous day's water consumption. Ensure the amount of medicated drinking water offered will be consumed completely within the selected treatment period (between 6 and 24 hours). No other source of drinking water should be available during the medication period.

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

The required amount of Exzolt on each treatment day is calculated from the total body weight of the entire group of chickens to be treated:

Volume of Exzolt (mL) per treatment day = Total body weight (kg) of birds to be treated x 0.05 mL/kg

Or

Volume of Exzolt (mL) per treatment day = Total body weight (lb) of birds to be treated x 0.023 mL/lb

Examples:

Total body weight of birds to be treated	Volume of Exzolt per treatment day
5000 kg (11,023 lb)	0.25 L (250 mL)
10,000 kg (22,046 lb)	0.5 L (500 mL)
15,000 kg (33,069 lb)	0.75 L (750 mL)
20,000 kg (44,092 lb)	1 L (1,000 mL)
80,000 kg (176,370 lb)	4 L
320,000 kg (705,479 lb)	16 L

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

- Check the water system to ensure it works properly and is free of leaks; also ensure 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 the product into a large medication tank or create a stock solution in a small container. The stock solution must be further diluted with drinking water and administered over time, using a proportioner or dosing pump. Always add product and water simultaneously in order to avoid foaming. It is important to rinse the measuring device used to measure the required product volume during the filling phase in order to ensure the complete dose is emptied into the medication tank or the stock solution and 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).
- Prior to introducing the medicated water, drain the drinker lines fully by opening the flush valve and checking end-line nipples to ensure no water remains.
- Prime the drinker lines with medicated water and confirm the medicated water has reached all end-line nipples.

Once the stock solution container is empty, rinse both the container and downstream water lines with unmedicated water (rinse water). Allow birds to consume the rinse water before reintroducing non-medicated water. The full course of therapy (2 single doses 7 days apart) must be administered for full therapeutic effect. Strict biosecurity measures at house and farm level should be implemented to prevent reinfestation 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.

Warnings

WITHDRAWAL PERIODS: Chickens must not be slaughtered for human consumption for 11 days after the last treatment. No egg discard is required when used according to the labeling.

User Safety Warnings: Not for use in humans. Keep this and all drugs out of the reach of children. Protective gloves should be used. Care should be taken when handling the product to avoid skin and eye exposure, exposure of mucous membranes and accidental ingestion. Accidental exposure may cause skin and eye irritation. In case of eye contact, immediately rinse thoroughly with water. If wearing contact lenses, immediately rinse the eyes first, then remove contact lenses and continue to rinse the eyes thoroughly. Seek medical advice if symptoms occur. Wash hands and contacted skin with soap and water after use of the product. Remove contaminated clothes and launder with detergent.

Accidental ingestion may cause gastrointestinal disturbances and hypersensitivity reactions in humans.

To obtain a copy of the Safety Data Sheet (SDS) or for technical assistance, call Merck Animal Health at 1-800-211-3573.

Contact Information: Contact Merck Animal Health at 1-800-521-5767 or sp-uspoultrycustomerserviceusa @merck.com. To report suspected adverse drug experiences, contact Livestock Technical Service at 1-800-211-3573. For additional information about reporting adverse drug experiences for animal drugs, contact FDA at 1-888-FDA-VETS or https://www.fda.gov/reportanimalae.

Clinical Pharmacology

Mechanism of Action: Fluralaner is for systemic use and belongs to the class of isoxazoline-substituted benzamide derivatives. Fluralaner acts as an inhibitor of the arthropod nervous system by antagonizing ligand-gated chloride channels (gamma-aminobutyric acid [GABA]-receptor and glutamate-receptor).

Pharmacokinetics: The pharmacokinetics of fluralaner were determined in 55 healthy laying hens treated with 2 single oral administrations of Exzolt (0.5 mg/kg twice) at 7-day intervals via drinking water. Concentrations of fluralaner peaked at 36 hours after the first administration and at 12 hours after the second administration. The maximum concentration (Cmax) of fluralaner was higher after the second dose (355 ng/mL) compared to after the first dose (323 ng/mL), suggesting slight accumulation. In a subsequent study, the oral bioavailability of fluralaner was determined in chickens after intravenous and oral administration (gavage) at a dose of 0.5 mg/kg. The oral bioavailability of fluralaner is 91%.

Target Animal Safety

Two margin of safety studies (growing broiler chickens and laying hens during peak egg production) and two reproductive safety studies (layer and broiler breeder chickens) were conducted.

The margin of safety study in broiler chickens was conducted in 320 3-week-old parent stock broiler chickens (Ross 308) at 1, 3 or 5 times the recommended dose of 0.5 mg/kg body weight for 3 times the recommended duration at 7-day intervals, and the margin of safety study in laying hens was conducted in 120 commercial laying hens (Novogen) at 1, 3 or 5 times the label dose for 3 consecutive days each, 7 days apart. No clinically relevant effects related to the administration of Exzolt were observed.

The reproductive safety study in layer chicken breeders was conducted in 432 (48 male and 384 female) commercial Bovans strain layer breeder chickens at 3 times the recommended dose on 4 occasions, 7 days apart and the reproductive safety study in broiler chicken breeders was conducted in 432 (48 male and 384 female) commercial Cobb 500 broiler breeder chickens at 3 times the recommended dose on 4 occasions, 7 days apart. No clinically relevant effects on reproductive safety parameters related to the administration of Exzolt were observed.

These studies support the safety of Exzolt in laying hens and replacement chickens when administered in drinking water as 2 single doses of 0.5 mg/kg body weight, 7 days apart.

Effectiveness

In a well-controlled dose confirmation study conducted in 128 commercial 14-week-old White Leghorn female replacement chickens (Hy-Line® strain), effectiveness against *Ornithonyssus sylviarum* was 100% on days 8, 14, 19 and 28 after the first treatment. No treatment-related adverse events were observed.

In a well-controlled dose confirmation study conducted in 128 commercial 29-week-old Brown Leghorn laying hens (Hy-Line strain), effectiveness against *Ornithonyssus sylviarum* was 100% on days 8 and 14 after the first treatment. No treatment-related adverse events were observed.

In a well-controlled field effectiveness study conducted in 800 commercial 23-week-old White Leghorn laying hens (Hy-Line strain), effectiveness against *Ornithonyssus sylviarum* exceeded 99.9% on days 8, 14, 19 and 28 after the first treatment. No treatment-related adverse events were observed.

In a well-controlled field effectiveness study conducted in 800 commercial 28-week-old White Leghorn (Hy-Line strain) laying hens, effectiveness against *Ornithonyssus sylviarum* exceeded 99.9% on days 8, 14, 19 and 28 after the first treatment. No treatment-related adverse events were observed.

In all 4 effectiveness studies, Exzolt achieved 99.3% or greater reduction in *Ornithonyssus sylviarum* mite counts on day 2 following the first dose administration.

How Supplied: Exzolt is available in 1- and 4-liter HDPE plastic containers.

Storage and Handling: Store Exzolt at or below 86°F (30°C) and use within 6 months after first opening. Use the medicated water within 24 hours of preparation. **Use within 8 hours of preparation if the medicated water comes in contact with rusty surfaces.**

Approved by FDA under NADA # 141-607

Distributed By: Intervet Inc. (d/b/a Merck Animal Health), 126 E. Lincoln Avenue, Rahway, NJ 07065

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