

Efficacy of afoxolaner plus milbemycin oxime and afoxolaner alone as treatment for sarcoptic mange in naturally infested dogs

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Abstract

Sarcoptic mange is a pruritic, contagious, ectoparasitic skin disease that affects mammals, including the domestic dog. The objective of this study was to evaluate and compare the efficacy of afoxolaner plus milbemycin oxime (NexGard Spectra) and afoxolaner alone (NexGard) as treatments for sarcoptic mange in naturally infested dogs. A total of 142 dogs naturally infested with *Sarcoptes scabiei* was evaluated. The dogs were diagnosed by microscopic examinations of skin scrapings. The dogs were divided into 2 groups: 96 dogs were treated with a combined dosage of 2.50 to 5.36 mg/kg body weight (BW) of afoxolaner and 0.50 to 1.07 mg/kg BW of milbemycin oxime and 46 dogs were treated with 2.50 mg/kg BW of afoxolaner alone. The presence or absence of pruritus and lesions were evaluated using an analogous scale on days 7, 14, 21, 28, and 56 after receiving the treatment. Data obtained were analyzed by Student's *t*-test ($P \leq 0.05$). The single oral treatment of afoxolaner plus milbemycin oxime resulted in a significant reduction in pruritus of 87.4% at 28 d after treatment ($P \leq 0.05$). Resolution of the lesions after treatment was variable, with a significant decrease ($P \leq 0.05$) observed within the first 14 d, although this parameter continued to improve until the end of the study on day 28, when a decrease of 96% was observed. By the end of the study, a single dose of either the afoxolaner alone or the afoxolaner combined with milbemycin oxime was effective in significantly reducing the signs associated with sarcoptic mange during a 56-day evaluation period.

Résumé

La gale sarcoptique est une maladie cutanée pruritique et contagieuse causée par un ectoparasite qui affecte les mammifères, incluant le chien domestique. L'objectif de la présente étude était d'évaluer et de comparer l'efficacité d'afoxolaner plus oxime de milbemycine (NexGard Spectra) et l'afoxolaner seul (NexGard) comme traitement pour la gale sarcoptique chez des chiens naturellement infestés. Un total de 142 chiens naturellement infestés avec *Sarcoptes scabiei* furent évalués. Les chiens étaient diagnostiqués par examen microscopique de grattages cutanés. Les chiens furent divisés en deux groupes : 96 chiens furent traités avec un dosage combiné de 2,50 à 5,36 mg/kg de poids corporel (BW) d'afoxolaner et de 0,50 à 1,07 mg/kg BW d'oxime de milbemycine et 46 chiens furent traités avec 2,50 mg/kg BW d'afoxolaner seul. La présence ou l'absence de prurit et de lésions furent évaluées en utilisant une échelle analogue aux jours 7, 14, 21, 28 et 56 après avoir reçu le traitement. Les données obtenues furent analysées à l'aide d'un test de *t* de Student ($P \leq 0,05$). Le traitement unique avec de l'afoxolaner plus oxime de milbemycine a résulté en une réduction significative du prurit de 87,4 % au jour 28 après le traitement ($P \leq 0,05$). Une résolution des lésions après le traitement était variable, avec une diminution significative ($P \leq 0,05$) étant observée au cours des 14 premiers jours, bien que ce paramètre continua de s'améliorer jusqu'à la fin de l'étude au jour 28, alors qu'une diminution de 96 % fut observée. À la fin de cette étude, une dose unique de soit de l'afoxolaner seul ou une combinaison afoxolaner-oxime de milbemycine était efficace à réduire de manière significative les signes associés avec la galle sarcoptique durant une période d'évaluation de 56 jours.

(Traduit par Docteur Serge Messier)

Introduction

Sarcoptic mange in animals, which is caused by the *Sarcoptes scabiei* mite (1), is still a problem in most parts of the world (2). Although mites are largely a host-specific species in their natural range, there are reports of *S. scabiei* infesting diverse hosts, including more than 100 species of mammals, such as dogs, cats, rabbits,

ovines, bovines, raccoons, and humans (2,3). *Sarcoptes scabiei* is a permanent obligate ectoparasite that lives and reproduces in the epidermis (4), most notably in sparse hair regions.

Infestations of *S. scabiei* var. *canis*, which causes sarcoptic mange in dogs, are not seasonal and have no age, breed, or sexual prevalence (5). However, animals enduring poor conditions, such as stress, overpopulation, poor nutrition, or immunosuppression, seem to

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be most susceptible to the disease (6). Sarcoptic mange is a pruritic disease with lesions that usually begin in less dense regions of the integument, such as the periocular skin, ear margins, and elbows (7). It can manifest itself *via* alopecia, scales, scabs, papules and, less often, lichenification and melanoderma (8). Skin damage can occur due to self-trauma secondary to pruritus (9), which allows mites to spread to an increasing proportion of the epidermis (7). Secondary skin infections are also common (9).

Diagnosis is based on clinical history, with a sudden onset of intense itching in one or more localized areas (10). Diagnosis is definitive with microscopic examination of skin scrapings revealing mites, eggs, and the remains of feces (11). As the sensitivity of this test has been shown to be less than 50% (12), the response to treatment is also considered a diagnostic method (13).

Therapeutic options for controlling sarcoptic mange consist of topical products such as selamectin (8) or moxidectin with imidacloprid (14). Medicated baths are usually part of the therapy to control scabies as they reduce the presence of scabs and dead mites and shorten treatment times (8). Subcutaneous ivermectin has also shown a variable response (15,8,16), although some dog breeds show adverse effects (17). A recent therapeutic option for sarcoptic mange in some areas is oral treatment with isoxazolines (18,5), which have demonstrated potent acaricidal and insecticidal activity through a mechanism of binding to neuronal chloride channels activated by gamma-Aminobutyric acid (GABA) and glutamate (19).

Afoxolaner, a molecule belonging to the isoxazoline class, has proven to be effective against *S. scabiei*, showing complete parasitological cure in dogs with sarcoptic mange after monthly oral treatment at the minimum effective dose of 2.50 mg/kg body weight (BW) (20). A chewable formulation that combines afoxolaner with milbemycin oxime, a macrocyclic lactone, has recently been registered to treat and control infestations of fleas, ticks, and infections with intestinal nematodes (21). Although milbemycin oxime is nematocidal, it has also been used in the treatment of sarcoptic mange and has proven useful in therapeutic management of scabies in dogs (22), without adverse reactions in dogs considered potentially sensitive to ivermectin (23). Therefore, the present study was designed to compare the efficacy of afoxolaner plus milbemycin oxime and afoxolaner alone as a treatment in dogs with natural infestation of *S. scabiei*.

Materials and methods

This study protocol was approved by the Ethics Committee of the Amecameca University Center of the Autonomous University of the State of Mexico (UAEM142), client-owned dogs from the State of Mexico, Mexico City, and Guadalajara, Mexico were included. The inclusion criteria were dogs of any age, breed, and gender that were positive for *S. scabiei* on microscopic study and with the prior approval of the owner by a letter of consent. The dogs were considered positive when the skin microscopy showed at least 1 infective form of *S. scabiei*, which was identified according to the morphology of the American Association of Veterinary Parasitologists (AAVP) guide (24), and with characteristic signs of sarcoptic mange.

During the treatment period, the dogs remained in standard accommodation and were fed a standard diet. All dogs were evalu-

ated on days 1, 7, 14, 21, 28, and 56. The evaluations were made by microscopy, with samples obtained by scraping, and by observing clinical signs of sarcoptic mange, such as erythema, comedones, follicular papules, pustules, scales, scabs, and alopecia. Areas such as the face, head, neck, sternum, chest, groin, abdomen, back, sides, front, rear end, perianal, perigenital, and tail were evaluated and each sign assigned a value of 0 (none), 1 (light), 3 (moderate), and 6 (severe), with a maximum value of 864 points. The level of pruritus was evaluated using a scale of 0 to 10, depending on its intensity. The evaluations of the dermatologic and pruritic lesions were carried out by the same person every day, for each dog.

A total of 142 dogs took part in this study. The dogs were divided into 2 groups. Group 1 consisted of 96 dogs and group 2 consisted of 46 dogs. On day 1 (time of positive skin scrapings), the dogs in group 1 were treated orally with a combination of 2.50 to 5.36 mg/kg BW of afoxolaner and 0.50 to 1.07 mg/kg BW of milbemycin oxime in a chewable tablet (NexGard Spectra; Merial, Lyon, France). Dogs in group 2 were treated with 2.50 mg/kg BW of afoxolaner alone (NexGard; Merial). For both groups, the tablets were administered directly into the mouth 10 min after eating. Skin scrapings were repeated on days 7, 14, 21, 28, and 56 and the presence or absence of live mites was recorded. The qualification of dermatologic lesions and evaluation of pruritus were repeated on these same days.

Data analysis

The data were captured in a database for further analysis. At the first statistical moment, the distribution of the data was determined and did not present a normal distribution. The data were analyzed and the information on the variables of pruritus and lesions during the week were analyzed by Tukey's studentized range test with an alpha of 0.05.

Results

The comparison of means of the pruritus level was done on days 1, 7, 14, 21, 28, and 56, as shown in Table I. While the pruritus scale was statistically equal at the beginning of the study, by day 7 there was a significant difference between the 2 treatments, showing a greater decrease in the pruritus level with the combined afoxolaner and milbemycin oxime treatment (group 1). On day 14 and 21, the results were the same and by day 28 the pruritus level had decreased to zero, which was a significant difference from afoxolaner (group 2). On day 56 post-treatment, however, both groups reached 0 on the pruritus scale.

The score of the lesions showed a significant difference from day 1, with a higher value in the dogs given afoxolaner and milbemycin oxime (group 1) on day 7 (Table II). The afoxolaner and milbemycin oxime treatment subsequently presented a notable difference and a greater decrease in the lesions despite the fact that the mean was higher at the beginning of the treatment in dogs treated with afoxolaner alone (group 2). Subsequently, on days 14, 21, and 28, there was also a significant difference between treatments, with a lower score in dogs given afoxolaner and milbemycin oxime (group 1). However, on day 56, which was the last day of testing, the lesion score of both groups had decreased to 0.

Table I. Weekly comparison of means of pruritus level in dogs with sarcoptic mange treated with 1 dose of afoxolaner (NexGard) or afoxolaner and milbemycin oxime (NexGard Spectra).

	Day					
	1	7	14	21	28	56
Afoxolaner	7.47 ^a	6.23 ^a	3.43 ^a	1.67 ^a	0.41 ^a	0 ^a
Afoxolaner & milbemycin oxime	6.87 ^a	3.55 ^b	1.01 ^b	0.07 ^b	0 ^b	0 ^a
CV	24.08	29.36	58.83	123.39	290.50	—
MSE	2.89	1.68	1.11	0.53	0.15	—

^{a,b} Means with different letters within a row represent a significant difference ($P < 0.05$).

CV — Coefficient of variation; MSE — Mean standard error.

Table II. Weekly comparison of means of lesion scores in dogs with sarcoptic mange treated with 1 dose of afoxolaner (NexGard) or afoxolaner and milbemycin oxime (NexGard Spectra).

	Day					
	1	7	14	21	28	56
Afoxolaner	195.0 ^a	138.50 ^a	54.18 ^a	19.39 ^a	4.60 ^a	0 ^a
Afoxolaner & milbemycin oxime	204.6 ^b	71.85 ^b	23.53 ^b	9.67 ^b	2.46 ^b	0 ^a
CV	0	56.82	76.52	95.47	125.00	—
MSE	0	2857.332	748.44	152.08	15.80	—

^{a,b} Means with different letters within a row represent a significant difference ($P < 0.05$).

CV — Coefficient of variation; MSE — Mean standard error.

Discussion

According to the results of the present study, oral administration of 2.50 to 5.36 mg/kg BW of afoxolaner combined with 0.50 to 1.07 mg/kg BW of milbemycin oxime (NexGard Spectra) significantly reduces the severity of the lesions and reduces the pruritus level to 0 in dogs naturally infested with *S. scabiei* by 28 d. The variability of results in the 142 dogs evaluated in this study demonstrates that sarcoptic mange has no predilection for breed, sex, or age (18,25).

The clinical resolution of pruritus and skin lesions associated with a decrease in the number of mites has previously been reported for products such as selamectin (26), imidacloprid/moxidectin (27,28), fipronil (25), amitraz/fipronil/S-methoprene (29), and topical fluralaner (5). Ivermectin is the most used treatment due to its convenience, low cost, and relative safety in dogs, except in some breeds (30), although cases have been reported that are refractory to treatment (25).

In the present study, the intensity of pruritus decreased significantly over the course of 2 weeks, indicating that afoxolaner plus milbemycin oxime has rapid acaricidal activity. In a previous study, afoxolaner demonstrated a reduction in pruritus to 0% associated with a total eradication of mites at 56 d (20). In the present study, although the pruritus level decreased, it continued until day 28 in the dogs given afoxolaner alone (group 2). It is therefore possible that the dead mites remaining on the skin continue to cause local irritation for more than 28 d. Fluralaner has been used in similar studies and a decrease of 98% in pruritus was observed on day 28 (3). This was

similar to what was previously observed on day 60, with 2 monthly administrations of sarolaner (18). The results obtained show that pruritus was reduced by 87.4%, lower than previously reported with other isoxazolines. It is therefore likely that some mites may still be alive after a month, requiring a second dose to achieve full antiparasitic efficacy.

Milbemycin oxime as a single treatment for sarcoptic mange has also shown good results in reducing pruritus after the third weekly dose, with no adverse effects observed in dogs considered potentially sensitive to ivermectin (23). No adverse effects were observed in this study after the administration of the treatment.

The efficacy of afoxolaner plus milbemycin oxime as an acaricide was also accompanied by a decrease in the rate of lesions characteristic of sarcoptic mange. Fluralaner was used in a previous study and a decrease of 98.92% in the score of the lesions was observed on day 28 (3). This is higher than that obtained in the present study, in which the decrease was 96% on day 28. This is probably related to the higher rate of pruritus until day 28, which could perpetuate the damage. In another study with afoxolaner, a decrease of 60% was observed in the crust index on day 28 (20). This is similar to the results of another study with fluralaner in which a decrease in erythematous papules was observed after 28 d and with sarolaner where a decrease of scabs, erythema, papules, and alopecia was observed on day 60 (18).

The rapid ectoparasitocidal effect from the use of afoxolaner alone and afoxolaner plus milbemycin oxime was previously evaluated in a field study of sarcoptic mange in dogs, in which both treatments resulted in substantial improvement of pruritus, papules, and crusts,

and alopecia and was statistically significant within 1 mo after the initial treatment (31).

In the present study, the treatment with afoxolaner combined with milbemycin oxime was associated with a significant reduction in the scores of the lesions between days 1 and 14. Although there were no significant differences between the following days, the score continued to decrease during the rest of the study, although the 28 d between the treatment and the evaluation of the lesions were probably too short to allow complete clinical resolution. It has also been observed that the resolution of the alopecia increases significantly after day 56 (20). However, the results of the present study are consistent with those obtained by Hampel et al (31), with both treatments gradually improving the clinical signs in dogs, in addition to being a safe treatment against canine sarcoptic mange.

In conclusion, the medication in which nematocide milbemycin oxime is added to afoxolaner in order to extend the ectoparasitocidal spectrum of afoxolaner demonstrates a slightly more rapid clinical resolution of clinical signs in spite of the fact that the milbemycin oxime dose is too low. Both afoxolaner alone and the combination of afoxolaner with milbemycin oxime in an oral chewable form are valuable tools for the treatment of scabies since both show high efficacy against canine sarcoptic mange after a monthly dose.

References

1. Angelone-Alasaad S, Molinar MA, Pasquetti M, et al. Universal conventional and real-time PCR diagnosis tools for *Sarcoptes scabiei*. *Parasit Vectors* 2015;8:587.
2. Currier RW, Walton SF, Currie BJ. Scabies in animals and humans: History, evolutionary perspectives, and modern clinical management. *Ann NY Acad Sci* 2011;1230:E50–E60.
3. Romero C, Heredia R, Pineda J, et al. Efficacy of fluralaner in 17 dogs with sarcoptic mange. *Vet Dermatol* 2016;27:353–e88.
4. Arlian LG, Morgan MS, Rider SD Jr. *Sarcoptes scabiei*: Genomics to proteomics to biology. *Parasit Vectors* 2016;9:380.
5. Taenzler J, Liebenberg J, Roepke RK, Frénais R, Heckerroth AR. Efficacy of fluralaner administered either orally or topically for the treatment of naturally acquired *Sarcoptes scabiei* var. *canis* infestation in dogs. *Parasit Vectors* 2016;9:392.
6. Patel JS, Patel PR, Panchasara HH, Brahmaxatri KG. Epizootiology of sarcoptic mange in Buffalo calves. *Indian Vet J* 2003;80:972–974.
7. Gill NK, Kaur M. To evaluate the efficacy of diagnostic tests for canine sarcoptic mange in dogs. *Indian J Sci Res* 2014;8:107–111.
8. Pin D, Bensignor E, Carlotti DN, Cadiergues MC. Localised sarcoptic mange in dogs: A retrospective study of 10 cases. *J Small Anim Pract* 2006;47:611–614.
9. Diwakar RP, Diwakar RK. Canine scabies: A zoonotic ectoparasitic skin disease. *Int J Curr Microbiol App Sci* 2017;6:1361–1365.
10. Curtis CF. Current trends in the treatment of *Sarcoptes*, *Cheyletiella* and *Otodectes* mite infestations in dogs and cats. *Vet Dermatol* 2004;15:108–114.
11. Chosidow O. Scabies. *N Engl J Med* 2006;354:1718–1727.
12. Walton SF, Currie BJ. Problems in diagnosing scabies, a global disease in human and animal populations. *Clin Microbiol Rev* 2007;20:268–279.
13. Curtis CF. Ectoparasites. In: Jackson HA, Marsella R, eds. *BSAVA Manual of Canine and Feline Dermatology*. England: Imprint Digital, 2015:153–163.
14. Fourie LJ, Du Rand C, Heine J. Evaluation of the efficacy of an imidacloprid 10%/moxidectin 2.5% spot-on against *Sarcoptes scabiei* var. *canis* on dogs. *Parasitol Res* 2003;90:S135–S136.
15. Behera SK, Dimiri U, Singh SK, Mohanta RK. The curative and antioxidative efficiency of ivermectin and ivermectin + vitamin E-selenium treatment on canine *Sarcoptes scabiei* infestation. *Vet Res Commun* 2011;35:237–244.
16. Currie BJ, Harumal P, McKinnon M, Walton SF. First documentation of in vivo and in vitro ivermectin resistance in *Sarcoptes scabiei*. *Clin Infect Dis* 2004;39:e8–e12.
17. Dowling P. Pharmacogenetics: It's not just about ivermectin in collies. *Can Vet J* 2006;47:1165–1168.
18. Becskei C, De Bock F, Illambas J, et al. Efficacy and safety of a novel oral isoxazoline, sarolaner (Simparica), for the treatment of sarcoptic mange in dogs. *Vet Parasitol* 2016;222:56–61.
19. 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 Mol Biol* 2014;45:111–124.
20. Beugnet F, de Vos C, Liebenberg J, Halos L, Larsen D, Fourie J. Efficacy of afoxolaner in a clinical field study in dogs naturally infested with *Sarcoptes scabiei*. *Parasite* 2016;23:26.
21. Letendre L, Harriman J, Drag M, Mullins A, Malinski T, Rehbein S. The intravenous and oral pharmacokinetics of afoxolaner and milbemycin oxime when used as a combination chewable parasiticide for dogs. *J Vet Pharmacol Therap* 2016;40:35–43.
22. Bergvall K. Clinical efficacy of milbemycin oxime in the treatment of canine scabies: A study of 56 cases. *Vet Dermatol* 1998;9:231–233.
23. Miller WH Jr, de Jaham C, Scott DW, Cayatte SM, Bagladi MS, Buerger RG. Treatment of canine scabies with milbemycin oxime. *Can Vet J* 1996;37:219–221.
24. Chapter 5, Diagnosis of arthropod parasites. In: Zajac AM, Conboy GA, eds. *Veterinary Clinical Parasitology*. 8th ed. Oxford, United Kingdom: Wiley-Blackwell, 2012:217–303.
25. Terada Y, Murayama N, Ikemura H, Morita T, Nagata M. *Sarcoptes scabiei* var. *canis* refractory to ivermectin treatment in two dogs. *Vet Dermatol* 2010;21:608–612.
26. Shanks DJ, McTier TL, Behan S, et al. The efficacy of selamectin in the treatment of naturally acquired infestations of *Sarcoptes scabiei* on dogs. *Vet Parasitol* 2000;91:269–281.
27. Fourie LJ, Heine J, Horak IG. The efficacy of an imidacloprid/moxidectin combination against naturally acquired *Sarcoptes scabiei* infestations on dogs. *Aust Vet J* 2006;84:17–21.
28. Krieger K, Heine J, Dumont P, Hellmann K. Efficacy and safety of imidacloprid 10% plus moxidectin 2.5% spot-on in the treatment of sarcoptic mange and otoacariasis in dogs: Results of a European field study. *Parasitol Res* 2005;97:S81–S88.
29. Gaxiola S, Gaxiola J, Perez A, et al. Effectiveness of two topical treatments with a combination fipronil/amitraz/(S)-methoprene against natural infestations of mites (*Sarcoptes scabiei* var. *canis*) on dogs. *Int J Appl Res Vet Med* 2013;11:10–15.

30. Correa-Salgado RA, Castaño E. Analysis of the canine ABCB1-1Δ mutation and its therapeutic and toxicological implications. (Evaluación de la mutación ABCB1-1Δ en perros y sus implicaciones terapéuticas y toxicológicas). *Rev Bio* 2014;13:65–75.
31. Hampel V, Knaus M, Schäfer J, Beugnet F, Rehbein S. Treatment of canine sarcoptic mange with afoxolaner (NexGard) and afoxolaner plus milbemycin oxime (NexGard Spectra) chewable tablets: Efficacy under field conditions in Portugal and Germany. *Parasite* 2018;25:63.