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Efficacy against nematode and cestode infections and safety of a novel topical fipronil, (S)-methoprene, eprinomectin and praziquantel combination product in domestic cats under field conditions in Europe

Steffen Rehbein[a,*], Balazs Capári[b], Georg Duscher[c], Dace Keidane[d], Zvezdelina Kirkova[e], Saulius Petkevičius[f], Dhimiter Rapti[g], Annegret Wagner[h], Thomas Wagner[i], S. Theodore Chester[j], Joseph Rosentel[j], Eric Tielemans[k], Martin Visser[a], Renate Winter[a], Katrin Kley[a], Martin Knaus[a]

[a] *Merial GmbH, Kathrinenhof Research Center, 83101 Rohrdorf, Germany*
[b] *8330 Sümeg, Hungary*
[c] *Veterinary University Vienna, Institute of Parasitology, 1210 Vienna, Austria*
[d] *Latvia University of Agriculture, 3001 Jelgava, Latvia*
[e] *Trakia University, Department of Parasitology, 6000 Stara Zagora, Bulgaria*
[f] *Lithuanian University of Health Sciences, Veterinary Academy, 47181 Kaunas, Lithuania*
[g] *Universiteti Bujqësor, Fakulteti i Mjeksise Veterinare, Kodër Kamëz, Tirana, Albania*
[h] *83122 Samerberg, Germany*
[i] *86825 Bad Wörishofen, Germany*
[j] *Merial Limited, Duluth, GA 30096, USA*
[k] *Merial SAS, Centre de Recherche de Saint-Vulbas, 01150 Saint-Vulbas, France*

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ABSTRACT

A novel topical combination product (BROADLINE®, Merial) composed of fipronil, (S)-methoprene, eprinomectin and praziquantel was evaluated for safety and efficacy against nematode and cestode infections in domestic cats. The study comprised a multi-centre, positive control, blinded, field study, using a randomized block design based on order of presentation for allocation. In total 196 client-owned cats, confirmed as positive for naturally acquired infections of nematodes and/or cestodes by pre-treatment faecal examination, were studied in seven countries in Europe. Pre-treatment faecal examination revealed the presence of *Toxocara*, hookworm, *Capillaria* and/or spirurid nematode infections in 129, 73, 33 or 1 cat(s), respectively; infections with taeniid and *Dipylidium* cestodes were demonstrated in 39 and 17 cats, respectively. Cats were allocated randomly to one of two treatments in a ratio of 2, topical fipronil (8.3%, w/v), (S)-methoprene (10%, w/v), eprinomectin (0.4%, w/v) and praziquantel (8.3%, w/v) (BROADLINE®, Merial; 130 cats); and 1, topical PROFENDER® Spot-On (Bayer; 66 cats) and treated once on Day 0. For evaluation of efficacy, two faecal samples were collected, one prior to treatment (Day -4 ± 4 days) and one at the end of the study (Day 14 ± 5 days). These were examined for fecal forms of nematode and cestode parasites. For evaluation of safety, cats were examined by a veterinarian before treatment and at the end of the study, and cat owners recorded the health status of their cats daily until the end of the study.

* Corresponding author. Tel.: +49 8032 70750; fax: +49 8032 707525.
E-mail address: steffen.rehbein@merial.com (S. Rehbein).

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For cats treated with Broadline®, the efficacy was >99.9%, 100%, and 99.6% for *Toxocara*, hookworms, and *Capillaria*, respectively; and the efficacy was >99.9%, >99.9%, and 98.5%, respectively, for the cats treated with Profender® ($p < 0.001$ for all nematodes and both treatments). Efficacy was 100% for both cestodes for both treatments ($p < 0.001$).
No treatment related adverse experiences were observed throughout the study. For both treatments, every cat that completed the study was given a safety score of 'excellent' for both local and systemic evaluations. The topical combination product of fipronil, (S)-methoprene, eprinomectin and praziquantel was shown to have an excellent safety profile and demonstrated high levels of efficacy when administered once as topical solution to cats infected with nematodes and cestodes under field conditions.

1. Introduction

Cats are hosts to a variety of internal parasites comprising protozoa, trematodes, cestodes, nematodes, and a few acanthocephalans. Some of these parasites, usually depending on their abundance, may impact the health of cats and are known to cause problems ranging from retarded growth or failure to thrive up to clinical intestinal disorders, such as diarrhoea or respiratory disease. In addition, cats, like other companion animals, represent potential reservoirs, carriers and transmitters of several diseases including zoonotic parasitic infections.

The most common helminth parasites of cats worldwide are parasites of the gastrointestinal tract: ascarids (*Toxocara cati* and, with substantially lower prevalence and abundance, *Toxascaris leonina*), hookworms (mainly *Ancylostoma tubaeforme* and, depending on climate, other *Ancylostoma* species and occasionally *Uncinaria stenocephala*), taeniid cestodes (predominately *Taenia taeniaeformis* but occasionally other *Taenia* species and on the rare occasion *Echinococcus multilocularis* in endemic areas) and tapeworms of the family Dipylidiidae (most commonly *Dipylidium caninum* but also species of *Diplopylidium* and *Joyeuxiella*). In addition, infections of cats by metastrongyloid and capillarid lungworms have been diagnosed with increasing frequency in the recent past.

Parasites of domestic cats in Europe have been studied extensively in previous years and nematode and cestode infections have been shown to be a common occurrence. However, factors like category of cats (e.g., stray, feral, shelter, cattery, well-cared-for pet), habitat (urban or rural), or general access to the outdoors, result in considerable variability in the prevalence of endoparasites (e.g., Borkovcová, 2003; Coati et al., 2003; Omeragić, 2003; Miró et al., 2004; Robben et al., 2004; Romaniuk et al., 2004; Ingstrup, 2008; Gracenea et al., 2009; Keidāns et al., 2009; Overgaauw et al., 2009; Duarte et al., 2010; Ładczuk and Balicka-Ramisz, 2010; Mircean et al., 2010; Traversa et al., 2010; Barutzki and Schaper, 2011; Claerebout et al., 2011; Knaus et al., 2011b; Becker et al., 2012; Mugnaini et al., 2012; Näreaho et al., 2012; Zanzani et al., 2012; Capári et al., 2013; Riggio et al., 2013).

Although intestinal helminth infections of cats may cause limited clinical signs, they may cause significant pathology and the presence of these parasites may be unacceptable to cat owners from aesthetic and hygienic points of view as well as for veterinary and medical reasons. Thus, appropriate management practices for the control of feline helminth infections are required including the use of efficacious and safe antiparasitic drugs.

This paper reports the results of a multi-centre field study which was designed to evaluate the efficacy of a novel topical fipronil, (S)-methoprene, eprinomectin and praziquantel combination against naturally acquired nematode and cestode infections and the safety of the product at the recommended commercial dose ranges in domestic cats.

2. Materials and methods

The study was designed in accordance with the International Cooperation on Harmonisation of Technical Requirements for Registration of Veterinary Medicinal Products – VICH GL7, "Efficacy of Anthelmintics: General Requirements" (Vercruysse et al., 2001) and "Efficacy of Anthelmintics: Specific Recommendations for Felines" VICH GL20 (Vercruysse et al., 2002), the "World Association for the Advancement of Veterinary Parasitology (WAAVP) guidelines for evaluating the efficacy of anthelmintics for dogs and cats" (Jacobs et al., 1994) and VICH GL9, entitled *Good Clinical Practice*. An Informed Consent and Agreement was obtained from the owners of the cats before enrolment.

All personnel involved in collecting efficacy and safety data, and the owners, were blinded to the treatment that had been assigned to the animals.

2.1. Study animals

Client-owned cats of any breed and sex, with a minimum age of two months and harbouring naturally acquired nematode and/or cestode infections, as confirmed by pretreatment fecal examination, were eligible to be included in the study. The cats were in good condition and no suspicion of parasitic infestation was made before examination. Fecal samples of 529 cats, from 284 owners, were collected at eight sites in seven European countries (Albania, 57 cats; Austria, 53 cats; Bulgaria, 47 cats; Germany 1, 26 cats; Germany 2, 37 cats; Hungary, 120 cats; Latvia, 74 cats; Lithuania, 115 cats) and subjected to coproscopical examination.

In total, 196 cats were confirmed as positive for naturally acquired infections of nematodes and/or cestodes and were enrolled (Albania, 33 cats/21 owners; Austria, 15 cats/7 owners; Bulgaria, 34 cats/18 owners; Germany 1, 16 cats/8 owners; Germany 2, 22 cats/12 owners; Hungary, 37 cats/25 owners; Latvia, 16 cats/16 owners; Lithuania, 23

cats/14 owners). These cats, 51 male, 20 male castrated, 98 female, 27 female spayed, were approximately two months to 13 years old and weighed 0.80–8.88 kg; 184 were common European Shorthair or local mixed breed cats, and 12 were pedigree cats or pedigree cat crosses (British Shorthair, European Longhair, Oriental Shorthair, Persian, Siamese, Turkish Angora, PersianX, Turkish AngoraX). All cats enrolled in the study were subjected to a physical examination before treatment; three and four cats, respectively, exhibited mild respiratory distress or had loose stool at this time.

2.2. Faecal examination

To qualify animals for the study and for the evaluation of efficacy based on reduction of fecal nematode egg counts or reduction of proportion of cats testing positive for cestode infection following treatment, fresh faecal samples were obtained before treatment and 14 ± 5 days post treatment (enrolled animals only) for examination. The faecal samples were examined, first grossly for the presence of cestode proglottids. Then, all samples were processed using modified McMaster techniques to establish faecal nematode egg counts. For floatation, zinc sulphate solution adjusted to a specific gravity of 1.3 was used, but the amount of faeces processed (2 g of faeces, six sites; 3 or 4 g of faeces, one site each) and the sensitivity of counting techniques, varied between sites (1 egg ≡ 20 eggs per g [EPG], one site; 1 egg ≡ 25 EPG, six sites; 1 egg ≡ 50 EPG, one site). Oocysts of *Isospora* species, when present, were handled similarly, and oocysts per g (OPG) counts were recorded. For taeniid and *Dipylidium* cestodes, only the presence of either proglottids or eggs was recorded. Identification of parasites was based on the distinct morphology of their faecal forms.

2.3. Evaluation of safety

The cats were examined physically by a veterinarian at enrolment (before treatment) and at the end of the study. Cat owners were required to report any adverse reactions as soon as possible to the investigator and to record the health status of their cats daily, until end of the study. At the end of the study, the investigator evaluated the safety based on the occurrence of any localized reactions at application site and adverse events (Table 1).

2.4. Allocation of cats to treatment groups and treatment administration

At each study site, replicates of three cats each were formed, based on their order of presentation. Within replicates, cats were allocated to one of two treatments according to the allocation sequence in a ratio of 2, topical fipronil (8.3%, w/v), (S)-methoprene (10%, w/v), eprinomectin (0.4%, w/v) and praziquantel (8.3%, w/v) (Broadline®,[1] Merial; 130 cats): 1 topical emodepside/praziquantel (PROFENDER® Spot-On, Bayer; 66 cats) and treated once on Day 0. For dose determination, cats were weighed on the day of treatment administration (Day 0).

Broadline® was available in the applicators of 0.3 mL for small cats (weighing ≥0.8–2.5 kg) and 0.9 mL for large cats (weighing >2.5–7.5 kg). Doses were administered topically after parting the hair on the midline of the neck, between the base of the skull and the shoulder blades, directly onto the skin in one spot. Profender®, available in three pipette sizes, was administered according to the manufacturer's instructions. Cats remained with their owners, in their usual environments and received their usual food and water.

2.5. Data analysis

Data from all study sites were combined for analysis, including data from cats of complete and incomplete replicates. Two cats that did not complete the study (one cat in each treatment group) were not included in any efficacy evaluation, and because they were not available for the follow-up physical examination, they did not provide safety information. Thus, data from 194 cats were retained for analysis.

The primary efficacy endpoint was the fecal egg counts for *Capillaria*, hookworm and *Toxocara* nematodes from the individual cat. Percent efficacy was computed comparing the Day 14 (±5) sample with the Day −4 (±4) sample for each treated group separately. Only cats that had the parasite being evaluated in the Day −4 (±4) sample were included in the efficacy evaluation and analysis. For nematodes, the fecal egg counts for each animal were transformed to the natural logarithm of (fecal egg count + 1) for the calculation of geometric means. The percent efficacy of each treated group was calculated using the formula $100 \times [(B - E)/B]$, where B is the geometric mean of the treated group on Day −4 (±4) and E is the geometric mean of the treated group on Day 14 (±5). For cestodes, only presence/absence information was recorded. The percent efficacy of each treated group was calculated as the percent of animals with cestodes present on Day −4 (±4) for which no cestodes were present on Day 14 (±5).

Statistical analysis was conducted using SAS® version 9.1.3. To compare the faecal egg counts of the two measurement days for the nematodes for each treatment group, the Van Elteren rank test was used with sites treated as blocks. The Freq Procedure was used employing the Cochran-Mantel-Haenszel-2 option with score = modridit. For cestodes, because animals were included only if they were positive on Day −4 (±4), comparing the two measurement days equated to testing whether the population proportion of animals testing positive on Day 14 (±5) was less than 1.0. The data were pooled over the sites and it was assumed that the proportion positive for each cestode on Day 14 (±5) could be approximated by a normal probability distribution. The Freq Procedure was used to compare baseline with study end, employing the Binomial option with tables setting the hypothesized value of the proportion to $P = 0.99$.

Non-inferiority analysis was used to compare the novel combination product with the reference product: the null hypothesis that Broadline was worse than Profender versus

[1] Broadline® is a trademark of Merial; all other marks are the property of their respective owners.

Table 1
Categories for assessment of local and systemic safety.

	Score	Safety	Guidance criteria
Local safety	0	'Excellent'	Nothing observed/physiologic
	1	'Acceptable'	Mild localized reaction, e.g., short lasting mild reaction of skin at application site
	2	'Poor'	Serious or unacceptable localized reaction observed, e.g., longer lasting reaction of skin, intense scratching at application site
Systemic safety	0	'Excellent'	No adverse event noted
	1	'Acceptable'	Mild adverse events observed, judged acceptable
	2	'Poor'	Serious or unacceptable adverse event(s) related to treatment observed

the alternative hypothesis that Broadline was equal to or better than Profender was tested. For nematodes, the expected fecal egg count per gram for each treatment group at Day 14 (±5) for each classification of nematode was compared using the mixed procedure in SAS version 9.1.3. The non-inferiority threshold was 2 eggs per gram. The log-fecal egg counts were the dependent variable, Treatment was the fixed effect, and Site was the random effect. A two-sided 95% confidence interval was computed on the Broadline™–Profender® difference. If the upper limit (the one-sided 97.5% confidence limit) was at or below 0.69 (log(2)), then the null hypothesis was rejected and the data supported that Broadline™ was equal to or better than Profender®. The proportion of cats that had cestodes present on Day 14 (±5) of the two groups was compared for each type of cestode represented. The data were pooled over the sites, and the two-sided 95% confidence interval of the difference between the two Treatments was computed, assuming that each proportion could be approximated by a normal probability distribution. The non-inferiority threshold was 0.15 (15%). If the upper limit of the confidence interval on the difference in the proportions was at or below 0.15, then the null hypothesis was rejected and the data supported that the two products were equivalent. To compute the upper confidence limit, the average of the proportions from the two treatment groups was used to estimate the variance for the difference between the treatment groups. When the observed proportions for both group were 0.0, the averaged proportion was estimated by $P_{ave} = 1/(2(n_1 + n_2))$, where n_1 and n_2 are the number of cats treated with Broadline™ and Profender®, respectively, that had that particular parasite on Day −4 (±4).

3. Results

3.1. Prevalence of endoparasite infections in the cats examined for enrolment

Of the 529 cats that were examined for potential enrolment in the study, 207 (39.1%) had evidence of infection with endoparasites (nematodes, cestodes and/or protozoans), as determined by standard coproscopic examination (gross examination plus examination by modified McMaster techniques). Total prevalence and count of faecal forms of endoparasites detected, if applicable, are given in Table 2. Identification of the faecal forms revealed eggs of nematodes (*Toxocara*, hookworm, *Capillaria* and spirurid), *Dipylidium* and taeniid cestode proglottids and/or eggs, and oocysts of protozoans of the genus *Isospora*. Overall, *Toxocara* was the most prevalent parasite followed by hookworm. Of the cats, 21.9% showed infection with a single parasite and 17.2% with multiple parasites (up to four parasites concurrently were observed) (Table 3). Table 4 summarizes the percent prevalence of endoparasite infections in the cats examined in the individual countries.

Table 2
Overall prevalence and number of parasite stages in the faeces of 529 cats from Albania, Austria, Bulgaria, Germany, Hungary, Latvia, and Lithuania examined for enrolment in the study, as determined by gross faecal examination for expelled parasite stages and modified McMaster techniques.

Parasite stage	Prevalence		Range of counts
	Number of cats positive at Day −4 (±4)	%	
Toxocara eggs	129	24.4	25–13,700 EPG[a]
Hookworm eggs	73	13.8	25–4900 EPG
Capillaria eggs	35	6.6	20–1375 EPG[b]
Spirurid eggs	1	0.2	(25 EPG)
Taeniid eggs/proglottids	39	7.4	NR[c]
Dipylidium eggs/proglottids	17	3.2	NR
Isospora oocysts	27	5.1	20–26,400 OPG[d]

[a] EPG = eggs per gram of feces.
[b] For *Capillaria* in two cats in Bulgaria only 'presence' was recorded instead of fecal egg count.
[c] NR = not recorded.
[d] OPG = oocysts per gram of feces.

Table 3
Occurrence of single- and mixed endoparasite infections of 529 cats from Albania, Austria, Bulgaria, Germany, Hungary, Latvia, and Lithuania examined for enrolment in the study, as determined by gross faecal examination for expelled parasite stages and modified McMaster techniques.

	Prevalence: total (%)
Single endoparasite infections	116 (21.9)
Toxocara	59 (11.1)
Hookworm	20 (3.8)
Capillaria	4 (0.8)
Taeniid	18 (3.4)
Dipylidium	4 (0.8)
Isospora	11 (2.1)
Mixed endoparasite infections	91 (17.2)
Capillaria + hookworm	14 (2.6)
Capillaria + *Toxocara*	3 (0.6)
Hookworm + *Toxocara*	17 (3.2)
Hookworm + *Dipylidium*	1 (0.2)
Hookworm + taeniid	1 (0.2)
Hookworm + *Isospora*	1 (0.2)
Hookworm + spirurid	1 (0.2)
Toxocara + *Dipylidium*	9 (1.7)
Toxocara + taeniid	13 (2.5)
Toxocara + *Isospora*	11 (2.1)
Taeniid + *Isospora*	1 (0.2)
Capillaria + hookworm + *Toxocara*	10 (1.9)
Capillaria + hookworm + *Dipylidium*	1 (0.2)
Hookworm + *Toxocara* + *Dipylidium*	1 (0.2)
Hookworm + *Toxocara* + taeniid	1 (0.2)
Hookworm + taeniid + *Isospora*	1 (0.2)
Toxocara + taeniid + *Isospora*	1 (0.2)
Capillaria + hookworm + *Toxocara* + taeniid	2 (0.4)
Capillaria + hookworm + *Toxocara* + *Dipylidium*	1 (0.2)
Hookworm + *Toxocara* + taeniid + *Isospora*	1 (0.2)

3.2. Evaluation of efficacy

Of the 196 (37.1%) cats which had evidence of infection with nematodes and/or cestodes and were enrolled in the study, allocated and treated once on Day 0, two cats did not complete the study (one cat harbouring *Capillaria* and *Toxocara* infections was reported as lost by the owner and one other cat harbouring *Toxocara* was struck by a car and died). Thus, analysis was based on data from 194 cats of which 127, 73 and 32 cats had evidence of *Toxocara*, hookworm, and *Capillaria* nematodes, respectively. Only 'presence' of *Capillaria* was recorded in two cats from Bulgaria instead of faecal egg count, resulting in 30 cats included in the efficacy evaluation for *Capillaria*). Thirty-nine and 17 cats had evidence of taeniid and *Dipylidium* cestodes, respectively, and were included in the efficacy evaluation.

For cats treated with topical fipronil, (S)-methoprene, eprinomectin and praziquantel, the efficacy was >99.9%, 100%, and 99.6% for *Toxocara*, hookworms, and *Capillaria*, respectively; and the efficacy was >99.9%, >99.9%, and 98.5%, respectively, for the cats treated with topical emodepside/praziquantel ($P < 0.001$ for all nematodes and both treatments) (Table 5). Efficacy was 100% for both cestodes in both treatment groups ($P < 0.001$) (Table 6).

For all three nematodes and taeniid cestodes, the null hypothesis was rejected, and the data supported that Broadline was equal to or better than Profender at the (one-sided) 2.5% significance level; however, because insufficient numbers of animals were infected with *Dipylidium*, the upper confidence limit was not less than the non-inferiority threshold in spite of the two treatments having identical 100% efficacies (Tables 5 and 6).

3.3. Evaluation of safety

No adverse experiences related to the treatments were observed throughout the study. Every cat in each treatment group that completed the study was given a safety score of 0 ('Excellent Safety') for both local and systemic evaluations.

4. Discussion

By examination of faecal samples of domestic cats from seven countries in Europe using conventional coproscopic techniques, it was determined that almost 40% of the cats had evidence of endoparasite infections. The spectrum of parasites identified in terms of their faecal forms comprised mainly parasites of the gastrointestinal tract: coccidians of the genus *Isospora*, ascarids of the genus *Toxocara*, hookworms (Ancylostomatidae), tapeworms of the genus *Dipylidium* and taeniid cestodes. All these parasites are common parasites of cats worldwide, and their occurrence in cats has been documented in the respective countries previously (e.g., Mustejkajte et al., 1961; Genov, 1971; Stoichev et al., 1982; Supperer and Hinaidy, 1986; Fok et al., 1988; Raschka et al., 1994; Schuster et al., 1997; Takács and Takács, 2002; Keidāns et al., 2008, 2009; Barutzki and Schaper, 2011; Knaus et al., 2011a,b; Becker et al., 2012; Capári et al., 2013).

It is noteworthy that *Capillaria* eggs were detected in the faeces of cats from all seven countries with an overall prevalence of 6.6%. *Capillaria* eggs (Fig. 1) found in the faeces of cats are unlikely to be derived from

Table 4
Percent prevalence of endoparasite infections in the cats examined in Albania, Austria, Bulgaria, Germany, Hungary, Latvia, and Lithuania, as determined by by gross faecal examination for expelled parasite stages and modified McMaster techniques.

Parasite	Albania ($n = 57$)	Austria ($n = 53$)	Bulgaria ($n = 47$)	Germany ($n = 63$)	Hungary ($n = 120$)	Latvia ($n = 74$)	Lithuania ($n = 115$)
Capillaria	31.6	3.8	4.3	1.6	5.0	4.1	2.6
Hookworm	54.4	7.5	21.3	0	16.7	8.1	1.7
Spirurid	1.8	0	0	0	0	0	0
Toxocara	19.3	24.5	53.2	38.1	22.5	18.9	13.0
Dipylidium	5.3	0	27.7	1.6	0	0	0
Taeniid	1.8	7.5	0	39.7	6.0	0	3.6
Isospora	10.5	1.9	0	0	3.3	0	7.8
Overall nematode and/or cestode	57.9	28.3	72.3	60.3	30.8	21.6	20.0

Table 5
Faecal nematode egg counts before (Day −4 ± 4) and after (Day 14 ± 5) treatment, percentage efficacy and results of data analysis.

Nematode	Treatment[a]	N[b]	Analysis of fecal egg counts (Van Elteren rank test)				Non-inferiority analysis	
			Geometric mean (EPG)[c]		% Efficacy[d]	*P*-value[e]	Upper 97.5% CI[f]	Threshold[g]
			Day −4 (±4)	Day 14 (±5)				
Capillaria[h]	1	19[i]	110.6	0.5	99.6	<0.001	0.63	0.69
	2	13	65.1	1.0	98.5	<0.001	NA[j]	NA
Hookworm	1	42	258.7	0.0	100	<0.001	0.07	0.69
	2	31	410.0	0.1	>99.9	<0.001	NA	NA
Toxocara	1	86	483.7	0.1	>99.9	<0.001	0.21	0.69
	2	41	439.1	0.3	>99.9	<0.001	NA	NA

[a] Treatment 1 = Broadline, Treatment 2 = Profender.
[b] Number of cats analysed per Treatment.
[c] Geometric mean fecal egg count, EPG = eggs per gram of faeces, pre-treatment (Day −4 ± 4) and post treatment (Day 14 ± 5).
[d] Percent efficacy = $100 \times [(B - E)/B]$, where B and E are the geometric mean fecal egg counts at Day −4 (±4) and at Day 14 (±5), respectively.
[e] (Two-sided) probability comparing Day 14 (±5) with Day −4 (±4) for each Treatment within each nematode.
[f] The upper 97.5% confidence limit on the difference of Treatment 1 −Treatment 2 was computed on the log-counts in the mixed procedure. If the upper limit is less than the threshold, then for that nematode, Treatment 1 was significantly not inferior to Treatment 2 at the 5% significance level, and under the hypothesis that the two Treatments had comparable fecal egg counts if the ratio of the means is less than 2.
[g] Threshold = logarithm of 2.
[h] For *Capillaria* in two cats (Bulgaria) allocated in Treatment 1 only 'presence' was recorded instead of fecal egg count. Thus, they were not included in the efficacy evaluation for *Capillaria* (although they were considered for inclusion in the analyses of the other nematodes). However, because both were assigned '0' (no *Capillaria* present), the '0' was equated to 0 EPG, and both cats were included in the non-inferiority analysis for *Capillaria*.
[i] Nineteen cats were included in percent efficacy evaluation, 21 in the non-inferiority analysis (cf. Footnote h).
[j] NA = not applicable.

bladder worms but may originate from capillarids residing in the gastrointestinal tract or the lungs, or the eggs may have passed through the alimentary tract following ingestion of parasitized prey. Although there was no detailed microscopic examination of the morphology of the eggs to distinguish the eggs of *Capillaria aerophila* from those of other capillarids (cf. Bowman et al., 2002; Traversa et al., 2011), the frequent detection of *Capillaria* eggs in the cats' faeces may support the hypothesis that the lungworm *C. aerophila* is spreading in Europe in both dogs and cats (Traversa et al., 2009, 2010; Di Cesare et al., 2011). Previously, finding capillarids in the stomach or intestine of cats was relatively rare (Thienpont et al., 1981; Raschka et al., 1994). The spirurid eggs found in the faeces of one cat from Albania may have originated from parasites of the gastrointestinal tract (for instance, anecdotal records of *Physaloptera* species in single cats from Hungary (Kávai, 1977) and Greece (Haralampides, 1978) or simply may have passed through the alimentary tract following ingestion of parasitized prey.

The overall rate of nematode and/or cestode infection of the cats varied between sites. However, this variability probably reflects the habitats from where the cats originated. For example, cats examined in Bulgaria, Germany and Hungary originated nearly exclusively from rural areas and those screened in Albania from suburban areas, while cats examined in Austria, Latvia and Lithuania predominately came from urban situations.

The results of this multi-centre study showed the new topical combination of fipronil, (S)-methoprene,

Table 6
Proportions of cestode-positive cats before (Day −4 ± 4) and after (Day 14 ± 5) treatment, percentage efficacy and results of data analysis.

Cestode	Treatment[a]	Analysis of proportion of cestode-positive cats				Non-inferiority analysis	
		Number of cats[b]		% Efficacy[c]	*P*-value[d]	Upper 97.5% CI[e]	Threshold[f]
		Day −4 (±4)	Day 14 (±5)				
Dipylidium	1	10	0	100	<0.001	0.16	0.15
	2	7	0	100	<0.001	NA[g]	NA
Taeniid	1	26	0	100	<0.001	0.07	0.15
	2	13	0	100	<0.001	NA	NA

[a] Treatment 1 = Broadline, treatment 2 = Profender.
[b] Number of cats that had evidence of the indicated cestode pre-treatment (Day −4 ± 4) and post treatment (Day 14 ± 5), respectively.
[c] Percent efficacy = $100 \times [(B - E)/B]$, where B and E are the numbers of cats that had evidence of the indicated cestode at Day −4 (±4) and at Day 14 (±5), respectively.
[d] Probability value testing that the probability was 1 of finding evidence of the cestodes at Day 14 (±5) given that it was present at Day −4 (±4) was 1.
[e] Upper confidence interval $= (P_1 - P_2) + 1.960\sqrt{\{1/n_1 + 1/n_2)P_{ave}(1 - P_{ave})\}}$, where P_1 and P_2 are the proportion of evidence of the indicated cestode at Day 14 (±5) for treatments 1 and 2, respectively, 1.960 is the 97.5th ordinate associated with the standard normal distribution, n_1 and n_2 are the number of cats that had the indicated genera at Day −4 (±4), and P_{ave} is the arithmetic average of P_1 and P_2. When $P_1 = P_2 = 0$, then $P_{ave} = 1/(2(n_1 + n_2))$.
[f] Threshold = 0.15.
[g] NA = not applicable.

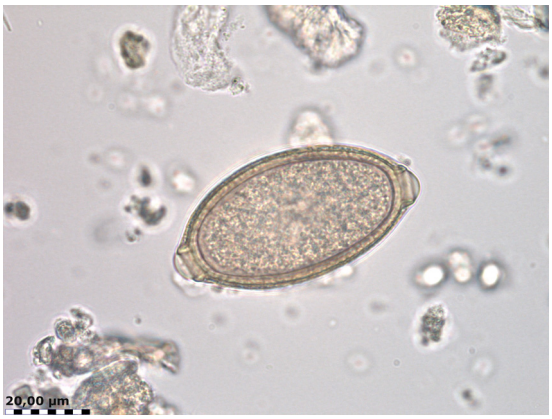

Fig. 1. Egg of *Capillaria aerophila* (© Merial).

eprinomectin and praziquantel is highly efficacious in the treatment of naturally acquired nematode and cestode infections under field conditions and thus confirm the results of the range of dose confirmation studies conducted under laboratory conditions using both induced and naturally infected cats (Knaus et al., 2014a,b,c,d; Prullage et al., 2014; Tielemans et al., 2014a). The cats included in the study represented a wide range of geographic origins, ages, body weights, breeds, and spectrum of parasite infections. The nematode and cestode parasites that were treated successfully with the novel combination product are important causes of health issues in cats and may have zoonotic potential (Štěrba and Baruš, 1976; Ekanayake et al., 1999; Lalošević et al., 2008; Bowman et al., 2010; Lee et al., 2010; Lloyd, 2011). Almost 50% of the parasite-positive cats included in the study harboured infections of at least two helminth parasites, and approximately 17% had evidence of concurrent nematode and cestode parasites. These findings are indicative of the common occurrence of multiparasitism in cats and thus favour the use of products providing both broad-spectrum nematocidal and cestocidal activity.

The novel topical combination would appear to offer a highly effective product for convenient administration in one spot to treat the most common parasitic infections of cats as its spectrum of activity covers not only adult and developing nematodes and cestodes but also ectoparasites, i.e. fleas and ticks (Baker et al., 2014; Tielemans et al., 2014b). The results demonstrate that fipronil, (S)-methoprene, eprinomectin and praziquantel, when administered once as topical solution to cats infected with nematodes and/or cestodes, is highly efficacious against a broad range of important nematode and cestode parasites and demonstrates excellent safety under field conditions.

Conflict of interest

The work reported herein was funded by Merial Limited, GA, USA. All authors are current employees or contractors of Merial.

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