Efficacy of Macrocyclic Lactone Endectocides Against *Boophilus microplus* (Acari: Ixodidae) Infested Cattle Using Different Pour-On Application Treatment Regimes

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ABSTRACT The efficacy of pour-on formulations of three macrocyclic lactone endectocides (moxidectin, ivermectin, and eprinomectin) was evaluated on cattle against *Boophilus microplus* (Canestrini) using two different treatment regimes. A single application treatment regime with each endectocide showed that fewer ticks per calf were recovered from all treated calves than from untreated cattle, but the level of control among the three treatments was similar (range; 78.7–87.7%) against all stages of ticks on the calves at the time of treatment. The engorged female and egg mass weights of all treated ticks were less than that of untreated ticks. Among the treated groups, the ivermectin and eprinomectin-treated females weighed less and produced lower weight egg masses than those from moxidectin-treated cattle. In a double application treatment regime with a 4-d interval between treatments, there were fewer ticks per calf recovered from the treated cattle than from untreated cattle. In addition, all treated females weighed less and produced lower weight egg masses than those from untreated cattle. Control with moxidectin (90.3%) was lower than with either ivermectin (98.9%) or eprinomectin (99.7%). The mean female and egg mass weight of the ivermectin and eprinomectin-treated groups was also less than that of the moxidectin treatment. A single application treatment against either 18- or 20-d-old adult ticks indicated that both moxidectin and ivermectin were less effective against 20-d-old ticks that were nearer to completing their parasitic development on the animal. In contrast, eprinomectin was the only endectocide tested that was equally effective against both 18- and 20-d-old ticks.

KEY WORDS *Boophilus microplus*, control, macrocyclic lactone, cattle tick, moxidectin, ivermectin

THE U.S. CATTLE FEVER TICK ERADICATION PROGRAM (CFTEP) has been in continuous operation since 1906, and because of this program *Boophilus* spp. ticks have been eradicated throughout the country, except for eight counties that lie adjacent to the Texas–Mexico border in South Texas (Graham and Hourrigan 1977). A permanent quarantine zone is maintained within these eight counties to prevent the reintroduction of *Boophilus* spp. ticks into the United States. Currently, the CFTEP relies exclusively on the systematic dipping of all livestock entering the United States with the organophosphorus (OP) acaricide, coumaphos, to prevent the reestablishment of these vectors of organisms that cause disease. Because of the reliance on a single acaricide and treatment method, there is a critical need for the development and evaluation of additional acaricides and treatment methods that could provide alternatives to the program in case severe OP-resistance in tick populations occurs or in the event that coumaphos and dipping treatments are not available, as has occurred with other OP pesticides in recent years.

Among the relatively few pesticides that have been developed for use against veterinary livestock pests in the last few years, the macrocyclic lactone (ML) endectocides appear to have definite potential. The unique chemistry and mode of action of this class of compounds coupled with their broad-spectrum activity at low concentrations makes them especially interesting as candidates for use in the CFTEP. Ivermectin, which is the oldest and most familar of the ML endectocides, has been shown to be effective against a number of tick species, including *Boophilus* spp. (Nolan et al. 1981; Lancaster et al. 1982; Cramer et al. 1988a, 1988b; Soll et al. 1989, 1990; Davey et al. 2001b). Although few studies have been conducted with some of the newer ML materials, moxidectin has also been

This article reports the results of research only. Mention of a proprietary product does not constitute an endorsement or a recommendation by the USDA for its use.

In conducting the research described in this report, the investigators adhered to protocol approved by the USDA-ARS Animal Welfare Committee. The protocol is on file at the Knipling-Bushland U.S. Livestock Insects Laboratory, Tick Research Unit, USDA-ARS, Kerrville, TX.

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shown to be highly effective against *Boophilus* spp. ticks (Remington et al. 1997). Among the most recently developed ML compounds, eprinomectin (Shoop et al. 1996), while not specifically evaluated against ticks, has not only been reported to be more effective than ivermectin (Williams et al. 1999), but is also more effective than any other registered ML endectocide against internal parasites (Shoop et al. 2001), which makes it of interest as a possible candidate for use in the eradication program.

The purpose of this study was to evaluate the efficacy of pour-on formulations of three different ML endectocides against *Boophilus microplus* (Canestrini) using different application treatment regimes. If any of these materials or treatment regimes provided a level of control that would be suitable for use in the eradication program, they would provide the program with a much needed alternative to the use of coumaphos and dipping vats that are presently the only means of eliminating ticks on cattle.

Materials and Methods

The study was conducted at the USDA, Agricultural Research Service, Cattle Fever Tick Research Laboratory, Mission, TX. The overall study was composed of three separate trials each designed to evaluate different aspects of the efficacy of three ML endectocides. They were applied under differing treatment regimes to cattle infested with adult, nymph, and larval *B. microplus* at the time of treatment.

The three trials were conducted sequentially, rather than simultaneously, because of logistical and labor resources. Naive cattle were used in each of the trials to prevent or reduce bias caused by cattle previously challenged with ticks. All cattle in each trial were maintained individually in stanchions inside an open-sided barn in 3.3 by 3.3-m stalls separated by 1.7-m high cinder block walls. Each trial was conducted under ambient conditions, except that no direct sunlight or rainfall reached the cattle because of the barn roof.

The three ML endectocides that were evaluated in each trial were moxidectin (Cydectin, Fort Dodge Animal Health, Fort Dodge, IA); ivermectin (Ivomec, Merck, Rahway, NJ); and eprinomectin (Eprinex, Merck). Each endectocide was a pour-on formulation obtained from a commercially available source and was registered for use against a variety of internal and external parasites, excluding ticks. The formulation of each endectocide used in the three trials contained 5 mg/ml (0.5%) of active ingredient (AI), and all treatments were applied to the infested cattle in each trial at the rate of 1 ml/10 kg of animal body weight, which was the labeled registered treatment dose for each chemical. The treatment method in each trial was a pour-on application consisting of measuring the appropriate volume of endectocide into a graduated cylinder, then evenly applying the material along the mid-line of the back of each calf from the withers to the tail head.

Trial I and Trial II. Since essentially all procedures followed in conducting trial I and trial II were the same, they are described together, and where the procedures differed between the two trials, the differences are noted. In trial I the infested cattle received a single pour-on application treatment of each endectocide, whereas in trial II the infested cattle were treated twice with each endectocide with a 4-d interval between the first and second pour-on treatment. A total of 16 Hereford heifer calves weighing ≈200 kg each was used in each trial. The calves in each trial were randomly divided into four equal groups containing four calves per group. In each trial the efficacy of each endectocide was based on three separate larval infestations made on each calf before the initiation of the treatment. All infestations on cattle in both trials were made by applying 17 by 60-mm (2-dram) shell vials containing ≈5,000 larvae that were 2–4 wk old to the front shoulder area of each calf using branding cement. After the vial was secure the cotton plug that held the larvae inside the vial was removed to allow larvae to move freely over the body of the calf. In both trials, each calf was infested 18 d before the initiation of treatment, and two additional infestations of ≈5,000 larvae each were made at 11 and 4 d before the initial treatment. This infestation pattern allowed for the evaluation of the efficacy of the endectocides against adult, nymph, and larval ticks at the time treatment was initiated.

The 16 calves used in each of the trials were weighed individually on the day the initial treatment was applied, so that the appropriate volume of test material for each animal could be calculated, then each calf was treated with the appropriate test material in the manner previously described. In each of the two trials, the first group of calves was treated with moxidectin; a second group of cattle was treated with ivermectin; and a third group of calves was treated with eprinomectin pour-on. The fourth group of cattle in each trial remained as an untreated control group. Following the initial treatment, the cattle used in trial II were retreated a second time 4 d after the initial treatment was applied.

Once the initial treatment was applied to cattle in each of the trials, data were collected on each calf in each treatment group for a period of 23 d (27 d after the last pretreatment larval infestation). This 23-d posttreatment evaluation period was based on the report that >95% of all engorged females infested at a given time would detach from the host within 27 d after infestation (Hitchcock 1955). On each day of the posttreatment evaluation period (both trials) all females that detached from each calf were collected and counted. A random sample of 10 engorged females was saved each day from each calf whenever possible to obtain weights, fecundity and fertility data. Engorged females from each daily collection sample on each calf were weighed collectively, placed in a 9-cm-diameter petri dish, and incubated at 27 ± 2°C, 92% RH, and a photoperiod of 12:12 (L:D) h. Females were allowed to oviposit for 20 d, after which the spent females were discarded and the eggs weighed, placed in a coded 25 by 93-mm (8-dram) shell vial and returned to the incubator. After 4 wk, the percentage egg hatch from

each sample group was visually estimated, using a stereo-microscope with an ocular grid, by comparing the proportion of larvae in relation to the proportion of unhatched eggs.

After all data (tick counts, egg mass weights, and percentage egg hatch) were collected over the 23-d evaluation period, the index of fecundity (IF) was calculated for each calf for each day using the following formula (Davey et al. 2001a): IF = total no. of ♂♂ collected × weight of eggs (grams)/no. of ♂♂ × egg hatch (%). Thus, the IF value is an estimate of the reproductive potential of a given number of females that lay a given quantity of eggs with a given hatching rate. The biological data (female weight and egg mass weight) were also used to provide an indication of whether the endectocide treatments had a measurable sublethal effect on the size and fecundity of the females that survived to repletion following treatment.

The 23 daily IF values for each calf were summed to obtain a total IF value for each animal. The total IF values for each calf in the control group were averaged to obtain a single mean IF value for the control group. This average IF value for the control group was compared with the total IF value for each of endectocide treated groups to obtain the percentage control of each endectocide using the following formula (Abbott 1925): % control = mean total IF; control group - total IF of each calf; treated group/mean total IF; control group × 100.

Trial III. Since it has been reported that the first detachment of *B. microplus* engorged females generally occurs at ≈20–21 d after larval infestation (Hitchcock 1955), the question arose as to whether there would be a difference in the efficacy of any of the three endectocides against adult ticks that still required additional time (≈2 d) to complete their development (as was the case in the 18-d pretreatment tick infestation used in trials I and II), as compared with adult ticks that were closer to repletion and undergoing rapid engorgement at the time treatment was initiated. Thus, to address this question, trial III was conducted. Although the endectocides, treatment method, treatment rate, and other factors followed in conducting trial III were the same as in the other trials, there were other procedures that were different. A total of 24 Hereford heifer calves weighing ≈200 kg each was used in trial III to evaluate the efficacy of the three endectocides against adult ticks that were infested on the cattle at two different times before treatment. The calves were randomly divided into eight equal groups containing three animals per group. The cattle in all eight groups were infested with a single infestation of ≈5,000 larvae that were 2–4 wk old. Four of the groups of cattle were infested at 20 d before the treatment of cattle and the remaining four groups were infested at 18 d before the treatment. The method of infestation was as previously described.

On the day of treatment one group of cattle infested with 20-d-old ticks and one group of cattle infested with 18-d-old ticks was treated with each of the three endectocides (total of six groups of cattle). The remaining two groups of cattle, one with 20-d-old ticks and one with 18-d-old ticks, remained untreated to serve as controls.

After the appropriate endectocide treatment was applied to each group of cattle, data were collected on each calf in each treatment group for a period of 20 d, at which time there were no engorged female ticks remaining on any of the calves. All females that detached each day from each calf were collected and counted, and daily random samples of ≤10 engorged females were saved to obtain weight, fecundity and fertility data as previously described.

After all the data (tick counts, egg mass weights, and percentage egg hatch) were obtained from the engorged females that detached during the 20-d posttreatment evaluation period for each animal in each treatment or control group, the daily IF values were calculated as previously described. The daily IF values for ticks from each calf were then summed over the 20-d evaluation period and the percentage control was determined for each treated calf as compared with the average value of the control group that had the same age class of adult ticks in the manner described above. The biological data of ticks obtained from each calf within each treatment group were also used to evaluate whether an endectocide treatment had a selective effect on the two groups of different age adult ticks.

Data Analysis. All data obtained in the study were analyzed with SAS (SAS Institute 1999). All percentage control data from the three trials was converted by arcsine transformation before analysis. Data obtained in trials I and II (tick number, percentage control, female weight, and egg mass weight) were analyzed with the General Linear model (GLM) procedure (PROC GLM) of the SAS software program, and differences ($P < 0.05$) among means were determined by Ryan-Einot-Gabriel-Welsch multiple range test. The data obtained in trial III were also analyzed with the SAS software program using a series of *t*-tests (PROC *t*-test) by comparing the data obtained from 18-d-old ticks with that of 20-d-old ticks within each endectocide treatment for each measured parameter (percentage control, female weight, and egg mass weight).

Results

Trial I: Single Application Treatment Regime. Significantly fewer ($F = 29.9$; df = 3, 12; $P < 0.0001$) engorged female ticks were recovered from all of the ML-treated cattle than from the untreated group of cattle, but among the treated groups there were no differences in the mean number of ticks per calf (Table 1). The level of control achieved with each of the endectocides tested ranged from 78.7% with moxidectin to 87.7% with eprinomectin, but there was no significant difference ($F = 0.1$; df = 2, 9; $P > 0.05$) in the level of control among the ML treatments.

The mean weight of females recovered from all ML-treated cattle was significantly less ($F = 305.4$; df = 3, 345; $P < 0.0001$) than females recovered from the untreated group (Table 2). Within the endectocide treated groups, the females recovered from the

Table 1. Mean ± SD tick number per calf and percentage control of the index of fecundity (IF) of *B. microplus* on untreated cattle and cattle treated with a single pour-on application of three macrocyclic lactones at 0.5% active ingredient (AI)

Macrocyclic lactone treatment	*n*	No. of ticks/calf	Control of the IF, %
Untreated	4	2,902 ± 65a	—
Moxidectin	4	1,445 ± 540b	78.7 ± 13.3a
Ivermectin	4	1,135 ± 238b	84.7 ± 3.0a
Eprinomectin	4	1,015 ± 232b	87.7 ± 3.1a

Means for each parameter were tested by general linear model (PROC GLM); means within the same column followed by the same letter are not significantly different ($P = 0.05$) tested by Ryan-Einot-Gabriel-Welsch multiple range test Number of ticks per calf, $F = 29.9$; df = 3, 12; $P < 0.0001$. Control of the IF, $F = 0.1$; df = 2, 9; $P > 0.05$.

moxidectin-treated cattle weighed significantly more than females obtained from either the ivermectin or eprinomectin-treated groups, of which the latter two treatment groups were not different from each other. As in the case of female weight, the mean weight of the egg masses derived from females obtained from all of the treated groups of cattle was significantly less ($F = 156.4$; df = 3, 345; $P < 0.0001$) than the untreated group (Table 2). Again, the weight of egg masses derived from moxidectin-treated females was significantly greater than that of ivermectin or eprinomectin-treated females, of which the latter two treatment groups did not differ from each other.

Trial II: Double Application Treatment Regime. One of the calves in the control group died of unknown causes 2 d before the initiation of treatments, thus analysis in this group was based on a sample size of three, rather than four calves. There were significantly fewer ($F = 83.5$; df = 3, 11; $P < 0.0001$) females per calf recovered from all ML-treated cattle than were recovered from the untreated group of cattle (Table 3). Within the treated groups, there were significantly more females obtained from the moxidectin-treated animals than either the ivermectin or eprinomectin treatment groups, of which the latter two treatment groups were not different. The level of control achieved with each of the ML acaricides ranged from 90.3% with moxidectin to 99.7% with eprinomectin, but the moxidectin treatment produced significantly lower control ($F = 10.5$; df = 2, 9; $P < 0.005$) than either of the other two treatments, of which the latter two treatment groups were not different.

Table 2. Mean ± SD female weight and egg mass weight of *B. microplus* females recovered from untreated cattle and cattle treated with a single pour-on application of three different macrocyclic lactones at 0.5% active ingredient (AI)

Macrocyclic lactone treatment	*n*	Female wt, mg	Egg mass wt, mg
Untreated	88	319 ± 35a	144 ± 39a
Moxidectin	88	216 ± 65b	70 ± 39b
Ivermectin	87	147 ± 30c	57 ± 20c
Eprinomectin	86	137 ± 41c	53 ± 25c

Means for each parameter were tested by general linear model (PROC GLM); means within the same column followed by the same letter are not significantly different ($P = 0.05$) tested by Ryan-Einot-Gabriel-Welsch multiple range test. Female weight, $F = 305.4$; df = 3, 345; $P < 0.0001$. Egg mass weight, $F = 156.4$; df = 3, 345; $P < 0.0001$.

Table 3. Mean ± SD tick number per calf and percentage control of the index of fecundity (IF) of *B. microplus* on untreated cattle and cattle treated twice at a 4-d treatment interval with pour-on formulations of three different macrocyclic lactones at 0.5% active ingredient (AI)

Macrocyclic lactone treatment	*n*	No. of ticks/calf	Control of the IF, %
Untreated	3	3,283 ± 191a	—
Moxidectin	4	1,068 ± 524b	90.3 ± 8.3a
Ivermectin	4	241 ± 75c	98.9 ± 0.8b
Eprinomectin	4	105 ± 68c	99.7 ± 0.3b

Means for each parameter were tested by general linear model (PROC GLM); means within the same column followed by the same letter are not significantly different ($P = 0.05$) tested by Ryan-Einot-Gabriel-Welsch multiple range test. Number of ticks per calf, $F = 83.5$; df = 3, 11; $P < 0.0001$. Control of the IF, $F = 10.5$; df = 2, 9; $P < 0.005$.

Females treated with ivermectin and eprinomectin had similar mean weights, but both groups of females weighed significantly less ($F = 432.1$; df = 3, 279; $P < 0.0001$) than moxidectin-treated females, which in turn, weighed significantly less than females obtained from untreated cattle (Table 4). The trend for the egg mass weights was the same as that of the female weights, where the mean egg mass weight of ivermectin and eprinomectin-treated females was significantly lower ($F = 473.8$; df = 3, 279; $P < 0.0001$) than the egg mass weight of females treated with moxidectin, which was lower than the egg mass weights of untreated females (Table 4).

Trial III: Single Application Treatment Against Adult Ticks of Different Ages. Analysis of the moxidectin treatment against ticks of two different ages showed no statistical difference ($t = 0.4$, df = 4, $P > 0.05$) in the level of control obtained against 18-d-old ticks (56.5%) as compared with 20-d-old females (39.5%), even though the level of control was considerably lower against the 20-d-old ticks that were closer to engorgement at the time of treatment (Table 5). Conversely, analysis showed that 18-d-old females that survived the moxidectin treatment weighed significantly less ($t = 3.5$, df = 96, $P < 0.0009$) and produced

Table 4. Mean ± SD female weight and egg mass weight of *B. microplus* females recovered from untreated cattle and cattle treated twice at a 4-d treatment interval with pour-on formulations of three different macrocyclic lactones at 0.5% active ingredient (AI)

Macrocyclic lactone treatment	*n*	Female wt, mg	Egg mass wt, mg
Untreated	63	315 ± 46a	137 ± 29a
Moxidectin	83	167 ± 56b	37 ± 22b
Ivermectin	81	88 ± 28c	21 ± 13c
Eprinomectin	56	73 ± 32c	18 ± 18c

Means for each parameter were tested by general linear model (PROC GLM); means within the same column followed by the same letter are not significantly different ($P = 0.05$) tested by Ryan-Einot-Gabriel-Welsch multiple range test. Female weight, $F = 432.1$; df = 3, 279; $P < 0.0001$. Egg mass weight, $P = 473.8$; df = 3, 279; $P < 0.0001$.

Table 5. Mean ± SD percentage control of the index of fecundity (IF), female weight, and egg mass weight of *B. microplus* adult females of two different ages recovered from infested cattle treated with a single application of pour-on formulations of three different macrocyclic lactones applied at 0.5% active ingredient (AI)

Macrocyclic lactone treatment	Age of adult ticks at treatment (days)	Control of the IF, %	Female wt, mg	Egg mass wt, mg
Moxidectin	18	56.5 ± 49.9NS	149 ± 75*	36 ± 36*
	20	39.5 ± 35.0NS	210 ± 97*	55 ± 44*
		$t = 0.4$, df = 4, $P > 0.05$	$t = 3.5$, df = 96, $P < 0.0009$	$t = 2.3$, df = 96, $P < 0.03$
Ivermectin	18	97.9 ± 2.5*	111 ± 43NS	29 ± 19NS
	20	87.0 ± 6.8*	119 ± 49NS	34 ± 22NS
		$t = 2.8$, df = 4, $P < 0.05$	$t = 0.9$, df = 92, $P > 0.05$	$t = 1.1$, df = 92, $P > 0.05$
Eprinomectin	18	95.9 ± 3.2NS	99 ± 35NS	31 ± 17NS
	20	91.4 ± 4.5NS	91 ± 48NS	26 ± 22NS
		$t = 1.4$, df = 4, $P > 0.05$	$t = 0.8$, df = 79, $P > 0.05$	$t = 1.2$, df = 79, $P > 0.05$

Means for each parameter within each treatment were tested by *t*-test (PROC *t*-test); means within the same column within the same treatment followed by NS are not significantly different ($P = 0.05$); means followed by "*" are significantly different ($P = 0.05$).

egg masses that weighed significantly less ($t = 2.3$, df = 96, $P < 0.03$) than the 20-d-old females, which were closer to repletion at the time of treatment.

In the ivermectin treated groups, the level of control was significantly lower ($t = 2.8$, df = 4, $P < 0.05$) against the 20-d-old females than was observed against 18-d-old females (Table 5). However, in contrast to the difference in the level of control, analysis of female biological data showed no difference in either the mean female weight ($t = 0.9$, df = 92, $P > 0.05$) or the mean egg mass weight ($t = 1.1$, df = 92, $P > 0.05$) of the different age females that survived to repletion following treatment.

Results of the measured parameters against ticks treated with eprinomectin were similar in both age classes of females (Table 5). Although the level of control against 18-d-old ticks was slightly higher (95.9%) than for 20-d-old females (91.4%), there was no difference between the two means ($t = 1.4$, df = 4, $P > 0.05$). Likewise, the mean weight of females and the mean weight of the egg mass were slightly greater for 18-d-old females than for 20-d-old females, but neither factor was significant (female weight, $t = 0.8$, df = 79, $P > 0.05$; egg mass weight, $t = 1.2$, df = 79, $P > 0.05$).

Discussion

While the results of this study demonstrated that a single application treatment (trial I) of any of the endectocides tested provided reasonably good control (range, 78.7–87.7%), the application of two treatments spaced at 4 d apart (trial II) provided appreciably higher control (range, 90.3–99.7%), with each ML averaging ≈12% higher control in the double treatment regime than in the single treatment regime. The female and egg mass weight of ticks recovered from treated cattle following initiation of treatments in both trials I and II were substantially lower than that of untreated females, indicating that each of the endectocides tested had a dramatic adverse effect on the fecundity and fertility of the females that survived. However, the weight of females obtained from treated cattle in the single application treatment regime (trial I) was 1.3- to 1.9-fold greater than ticks obtained from treated cattle in the double application treatment regime (trial II), and the egg mass weights of single treatment females was 1.9- to 2.9-fold greater than females that were treated two times. Once again, these data indicated that the double application treatment regime was more effective than a single treatment. Finally, while moxidectin and ivermectin were less effective against ticks that were undergoing rapid engorgement (20-d-old ticks) than ticks that had not reached the rapid engorgement phase of development (18-d-old ticks) at the time of treatment, eprinomectin was equally effective against both age groups. This suggested that eprinomectin was either absorbed into the blood stream of the cattle and reached efficacious levels more rapidly than the other endectocides or that eprinomectin molecules bound better to their target site than the other compounds, thus increasing the level of toxicity.

Although there are a number of studies that have been conducted in which ivermectin was evaluated against ticks, few studies have evaluated the efficacy of moxidectin, and none has examined the effect of eprinomectin against any tick species. The results of this study (trial I) compare favorably with Cramer et al. (1988a, 1988b), who reported that a single topical application of ivermectin against *B. microplus* ticks at concentrations of 50 and 100 μg/kg provided 85 and 91% overall control, respectively, and that the weights of female ticks that survived a single ivermectin treatment were consistently reduced. Similarly, Caproni et al. (1998) reported that a single injection treatment with ivermectin at 200 μg/kg provided 83.2% reduction in *B. microplus* ticks at 12 d posttreatment. In another study that showed some similarities to our results (trial I), it was reported that a single pour-on treatment of moxidectin applied at 0.5 mg/kg markedly reduced egg production in female ticks and produced a mean level of control that was >92%, but the range of control was 71–100% (Remington et al. 1997). Although no studies have been conducted with eprinomectin against any tick species, this ML was reported to be significantly more effective against internal parasites than ivermectin (Williams et al. 1999),

and better than any of the newer ML endectocides (Shoop et al. 2001), which is consistent with our findings against *B. microplus* ticks.

It has been suggested by other investigators that the use of multiple treatments with ML endectocides, as was the case in trial II of this study, could provide greater efficacy against some tick species, particularly one-host ticks, such as *Boophilus* spp. (Horak et al. 1983). Results of the double treatment regime evaluated in this study compared favorably with that of Nolan et al. (1985), who reported that while two or three ivermectin treatments spaced 3 or 4 d apart failed to cleanse animals of ticks, a two treatment regime applied at 4 d apart, as in trial II, was highly effective against *B. microplus.* Likewise, multiple treatments with moxidectin applied at 4-wk intervals at a concentration of 0.5 mg/kg with a pour-on formulation produced excellent results, with tick counts being either zero or very low over a 21-wk study period (Remington et al. 1997).

Results in this study showing that ivermectin and moxidectin were less effective against ticks that were in the rapid engorgement phase of development (20-d-old adults) when treatment was applied were consistent with other studies. Nolan et al. (1981) reported that a significant number of *B. microplus* females survived during the initial 2 d following treatment with ivermectin. These investigators suggested that adults in the final stages of engorgement were either less susceptible to the chemical or there was a lag phase immediately following treatment during which lethal amounts of the chemical were not ingested by the ticks. Similarly, it was reported that the efficacy of a single topical application of ivermectin was low at 1–3 d following treatment, indicating that the material was not as effective against adult ticks that were near repletion at the time of treatment (Cramer et al. 1988a). Contrast this with our results with eprinomectin, which indicated little difference in efficacy against adult ticks regardless of their developmental stage at the time of treatment. These results suggest that eprinomectin is probably absorbed into the blood stream of the calves more rapidly and at higher levels than ivermectin or moxidectin, thus reaching lethal levels before engorging females detach from the host.

The results obtained in the single application treatment regime (trial I) seem to indicate that any of the ML endectocides tested would be suitable in a program where strategic treatments are applied to infested cattle with the objective of reducing the tick burden to a level that prevents economic injury to the animals. However, even in such a strategic treatment control program, the use of eprinomectin would appear to be the best choice of the three ML compounds tested because, even though the level of control was not statistically higher, it was still somewhat greater than ivermectin or moxidectin, and in addition, eprinomectin would likely be more effective against adults that were near engorgement when the strategic treatments were applied.

In an eradication program where total elimination of the ticks is the objective, the use of a single application treatment regime would be unacceptable, regardless of which ML endectocide was used, because the control falls far short of the 99% level that is traditionally used as the standard by which an acaricide is considered for potential use in the U.S. Fever Tick Eradication Program. Even in the double application treatment regime (trial II), both moxidectin and ivermectin failed to reach a level of control (>99%) that would be suitable for use in the eradication program, although ivermectin came very close. However, the use of eprinomectin in the double application treatment regime (trial II) provided encouraging results regarding its potential for use in the eradication program. In areas within the United State where tick infestations are detected on cattle that are being held on premises, the use of this treatment regime in a systematic, repeated treatment approach where double treatments are applied at 4 d intervals followed by a 21 or 28 d nontreatment interval, followed by a repeated double treatment regime, and so on, it is likely that eradication of a field population of ticks could be achieved in a relatively short time. In such a systematic treatment scenario each double application treatment regime would provide >99% control of the ticks on the host at the time of treatment, thus there would be virtually no viable ticks being added to the tick population in the field between each of the double application treatment regime intervals, leading to a collapse in the field tick population. Obviously, this type of treatment scenario needs to be evaluated under field conditions to determine if successful eradication of a field tick population can be accomplished, but if it is successful it would provide a superior advantage over the present regulations, which require repeated treatments of cattle on infested premises every 14 d for a period of 6–9 mo.

Another area for additional studies with these compounds may lie in evaluating their efficacy at a higher dose than was used in this study. Since the concentration used in this study was specifically recommended for the control of internal parasites, perhaps a higher dose (i.e., 2-three times higher than this study) would provide the >99% control level necessary for use in the fever tick eradication program. If the use of these compounds, even at a higher dosage level, resulted in > 99% control of the ticks, then it would provide an attractive alternative to the use of coumaphos that is presently the only acaricide approved for use in the program.

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References Cited

Abbott, W. S. 1925. A method for computing effectiveness of an insecticide. J. Econ. Entomol. 18: 265–267.

Caproni, L., Jr., O. Umehara, E. Moro, and L.C.B. Goncalves. 1998. Field efficacy of doramectin and ivermectin against natural infestation of the cattle tick *Boophilus microplus.* Rev. Bras. Parasitol. Vet. 7: 151–155.

Cramer, L. G., L.A.F. Carvalho, A. A. Bridi, N. K. Amaral, and R. A. Barrick. 1988a. Efficacy of topically applied ivermectin against *Boophilus microplus* (Canestrini, 1887) in cattle. Vet. Parasitol. 29: 341–349.

Cramer, L. G., A. A. Bridi, N. K. Amaral, and S. J. Gross. 1988b. Persistent activity of injectable ivermectin in the control of the cattle tick *Boophilus microplus.* Vet. Rec. 122: 611–612.

Davey, R. B., J. E. George, and D. E. Snyder. 2001a. Efficacy of a single whole-body spray treatment of spinosad, against *Boophilus microplus* (Acari: Ixodidae) on cattle. Vet. Parasitol. 99: 41–52.

Davey, R. B., J. A. Miller, and J. E. George. 2001b. Efficacy of daily oral treatments of ivermectin administered to cattle infested with *Boophilus microplus* (Acari: Ixodidae). J. Agric. Urban Entomol. 18: 127–137.

Graham, O. H., and J. L. Hourrigan. 1977. Eradication programs for the arthropod parasites of livestock. J. Med. Entomol. 13: 629–658.

Hitchcock, L. F. 1955. Studies on the parasitic stages of the cattle tick, *Boophilus microplus* (Canestrini) (Acarina: Ixodidae). Aust. J. Zool. 3: 145–155.

Horak, I. G., J. Boomker, S. A. Kingsley, and V. de Vos. 1983. The efficacy of ivermectin against helminth and arthropod parasites of impala. J. So. Afr. Vet. Assoc. 54: 251–253.

Lancaster, J. L., Jr., R. L. Kilgore, and J. S. Simco. 1982. Efficacy of low level daily doses of ivermectin in calves against three species of ticks. Southwest. Entomol. 7: 116–118.

Nolan, J., H. J. Schnitzerling, and P. Bird. 1981. Evaluation of the potential of systemic slow release chemical treatments for control of the cattle tick (*Boophilus microplus*) using ivermectin. Aust. Vet. J. 57: 493–497.

Nolan, J., H. J. Schnitzerling, and P. Bird. 1985. The use of ivermectin to cleanse tick infested cattle. Aust. Vet. J. 62: 386–388.

Remington, B., P. Kieran, R. Cobb, and D. Bodero. 1997. The application of moxidectin formulations for control of the cattle tick (*Boophilus microplus*) under Queensland field conditions. Aust. Vet. J. 75: 588–591.

SAS Institute. 1999. SAS user's, manual, version 8.2 for Windows. SAS Institute, Cary, NC.

Shoop, W. L., J. R. Eggerton, C. H. Eary, H. W. Haines, B. F. Micheal, H. Mrozik, P. Eskola, M. H. Fisher, L. Slayton, D. A. Ostlind, B. J. Skelly, R. K. Fulton, D. Barth, S. Costa, L. M. Gregory, W. C. Campbell, R. L. Seward, and M. J. Turner. 1996. Eprinomectin: A novel avermectin, for use as a topical endectocide for cattle. Int. J. Parasitol. 26: 1237–1242.

Shoop, W., B. Michael, J. Egerton, H. Mrozik, and M. Fisher. 2001. Titration of subcutaneously administered eprinomectin against mature and immature nematodes in cattle. J. Parasitol. 87: 1466–1469.

Soll, M. D., I. H. Carmichael, G. E. Swan, and S. J. Gross. 1989. Control of induced infestations of three African multihost tick species with sustained-release ivermectin. Exp. Appl. Acarol. 7: 121–130.

Soll, M. D., G. W. Benz, I. H. Carmichael, and S. J. Gross. 1990. Efficacy of ivermectin delivered from an intraruminal sustained-release bolus against natural infestations of five African tick species on cattle. Vet. Parasitol. 37: 285–296.

Williams, J. C., A. F. Loyacano, A. DeRosa, J. Gurie, B. C. Clymer, and F. Guerino. 1999. A comparison of persistent anthelmintic efficacy of topical formulations of doramectin, ivermectin, eprinomectin and moxidectin against naturally acquired nematode infections of beef calves. Vet. Parasitol. 85: 277–288.

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