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Therapeutic and persistent efficacy of a single injection treatment of ivermectin and moxidectin against *Boophilus microplus* (Acari: Ixodidae) on infested cattle☆

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Abstract. The effectiveness of a single treatment with either ivermectin or moxidectin was determined by administering a single subcutaneous injection of each endectocide at 200 μg per kg body weight to cattle infested with all parasitic developmental stages (adults, nymphs, and larvae) of *Boophilus microplus* (Canestrini). The percentage reduction in the number of females that reached repletion following treatment (outright kill) was 94.8 and 91.1% for ivermectin and moxidectin, respectively. In addition, the reproductive capacity of the females that did survive to repletion was reduced by > 99%, regardless of the endectocide. Based on these two factors, the therapeutic level of control obtained against ticks on the cattle at the time of treatment was 99.0 and 99.1% for ivermectin and moxidectin, respectively. Engorged females recovered from either group of treated cattle weighed ≈3-times less than untreated females, and the egg masses produced by treated females weighed ≈5–8-times less than egg masses produced by untreated females. Partitioning of data into three separate 7-d post-treatment intervals allowed for an estimation of the efficacy of each endectocide against each individual parasitic development stage (adult, nymph, and larva). Results indicated that both endectocides were ≥99.7% effective against ticks that were in either the adult or nymphal stage at the time of treatment. However, the level of control against ticks in the larval stage of development at treatment was significantly lower at 97.9 and 98.4% for ivermectin and moxidectin, respectively. Analysis of the persistent (residual) activity of the two endectocides indicated that neither material provided total protection against larval re-infestation for even 1-wk following treatment. Against larvae infested 1–4 wk following treatment, the level of control with moxidectin ranged from 92.4% (1 wk) to 19.5% (4 wk). These control levels were higher at each weekly interval than for ivermectin, which ranged from 82.4% (1 wk) to 0.0% (4 wk). The potential for the use of these injectable endectocide formulations in the US *Boophilus* Eradication Program is discussed.

☆This paper reports the results of research only. Mention of a commercial or proprietary product in this paper does not constitute an endorsement by the U.S. Department of Agriculture. 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 USDA-ARS, Knipling-Bushland U.S. Livestock Insects Laboratory, Tick Research Unit, Kerrville, TX.

Boophilus spp. ticks, which are the primary vector of bovine babesiosis, have been an enormous impediment to livestock production in many of the tropical and subtropical areas of the world for well over a century. For more than three decades requirements mandated by the Animal and Plant Health Inspection Service (APHIS), Veterinary Services (VS) branch of the United States Department of Agriculture (USDA), Cattle Fever Tick Eradication Program (CFTEP) at import facilities have prevented the re-entrance of *Boophilus* spp. ticks into the U.S. on imported cattle through the exclusive use of the organophosphate (OP) acaricide, coumaphos (Graham and Hourrigan 1977). Program procedures require that every animal presented for import from Mexico be thoroughly hand inspected ('scratch' inspected) to determine whether any *Boophilus* ticks are present on any animal. Should any *Boophilus* ticks be found, even on a single animal, the entire herd is denied entry into the country. Even though the herd is not allowed to pass into the US, all animals are dipped in a total immersion vat charged with coumaphos at a concentration of 0.3% active ingredient (AI). The cattle may be resubmitted for import after a period of not less than 10, nor more than 14 d following treatment. If any live ticks are found on any animal at the second submission the entire herd is rejected, and the animals may not be resubmitted for import again. When no ticks are found on any animal at either the first or second submission, all cattle are dipped at 0.3% AI coumaphos and allowed to enter the US. Strict adherence to these prescribed procedures has never failed to prevent the introduction of *Boophilus* ticks into the US on imported cattle. Coumaphos has been shown to kill even fully replete females within 10 d after treatment, and it is 100% effective against the immature stages (nymphs and larvae) of the tick (Davey and Ahrens 1982; Davey et al. 1997). Total efficacy (100%) against immature ticks is critical to the success of the eradication program because they could easily escape detection during inspection as a result of their small size. Thus, the survival of any immature ticks could be a source that might result in an outbreak in tick-free areas of the US.

The increasing level of OP-resistant *B. microplus* (Canestrini) populations throughout Mexico has created a critical need in the CFTEP with respect to the identification, development, and implementation of alternative acaricides that will prevent the ingress of OP-resistant ticks into the US. In addition, documented resistance of *B. microplus* to pyrethroid and formamidine acaricides (Fragoso et al. 1995; Miller et al. 1999; Soberanes et al. 2002; Li et al. 2004) has removed these chemical classes from consideration as alternatives to coumaphos. In order for any potential acaricide to be considered as a replacement for coumaphos, it must possess essentially the same efficacy qualities under conditions that are equally as rigorous as those required for the use of coumaphos.

The development and marketing of macrocyclic lactone products, collectively referred to as endectocides, for use against various internal and external parasites may provide the only alternative to coumaphos, since they are the only major group of chemical agents to which *Boophilus* ticks in Mexico have not yet shown any resistance. Studies with a number of these endectocides, such as ivermectin, eprinomectin, moxidectin, and doramectin have shown that these compounds have excellent activity against *B. microplus* under certain circumstances (Cramer et al. 1988a,b; Gonzalez et al. 1993; Remington et al. 1997; Caproni et al. 1998; Guglielmone et al. 2000; Davey and George 2002; Aguilar-Tipancumu and Rodriguez-Vivas 2003). However, little of the available information applies to the utility of these materials under the rigorous conditions associated with eradication that would occur at import facilities, with the exception of a doramectin injectable formulation that would satisfy the standards set by the CFTEP (George and Davey 2004).

The purpose of this study was to evaluate the therapeutic and persistent efficacy of a single treatment of two different endectocides administered to cattle infested with all stages of *B. microplus*. The design of the study not only permitted overall evaluation of the efficacy of the endectocides, but also allowed for an evaluation against each parasitic life stage of the tick (adult, nymph, and larva) individually. Results of the study established the potential feasibility for using these materials at US import facilities, as well as providing insight into their possible use in systematic treatment regimes used by the program to eradicate *Boophilus* ticks on US premises within the permanent quarantine zone that are found to be infested.

Materials and methods

The study was conducted at the USDA, Agricultural Research Service (ARS), Cattle Fever Tick Research Laboratory, Edinburg, TX. Injectable formulations of two different endectocides, ivermectin and moxidectin were evaluated in the study. These agents were applied as a single subcutaneous injection to cattle infested with *B. microplus* that were in the adult, nymphal, and larval stage of parasitic development at the time of treatment. Additional larval infestations were applied to the cattle at regular intervals following treatment to estimate the persistent (residual) efficacy of each compound.

All cattle used in the study were naïve to *Boophilus* ticks prior to experimentation to reduce or prevent any bias that might occur as a result of previous exposure to tick challenge. Throughout the study all calves were maintained in individual stanchions within an open-sided barn in separate 3.3 × 3.3 m stalls separated from each other by 1.7 m high cinder block walls. The trial was conducted under ambient conditions with no exposure to direct sunlight or rainfall due to the presence of the barn roof.

The ivermectin (Ivomec® Injectable) used in the study is a registered product of Merial Inc. (Rahway, NJ) with claims for use against a variety of internal and external parasites other than ticks, whereas the moxidectin injectable formulation made by Fort Dodge Animal Health Inc. (Fort Dodge, IA) is an as yet unregistered product in the US. Both materials were formulated as 1% injectable formulations, and each compound was administered to treated calves at a dose rate of 200 μg per kg of body weight (1 ml per 50 kg).

Twelve Hereford heifer calves weighing ≈200 kg each were randomly divided into three equal groups of four calves per group. Evaluation of the therapeutic efficacy of the two compounds was based on three separate infestations on each animal prior to treatment using larvae that were 2–4 wk old. All infestations to investigate the therapeutic efficacy of the agents were conducted by applying 17 × 60 mm (2-dram) shell vials containing ≈5000 larvae to the midline of the back between the shoulders using branding cement. Once the vial was secure the cotton stopper restraining the larvae was removed to allow the larvae to move freely over the entire body of the calf. The first infestation was applied to each calf at 18 d prior to treatment, whereas the next two infestations were made at 11 and 4 d prior to treatment. This pattern of infestation allowed for the evaluation of the effect of the endectocides against adult, nymphal, and larval ticks on the host at the time treatment was applied.

The calves were weighed individually within 3 d of the treatment so the appropriate volume of test material could be calculated. On the day of treatment one group of cattle was treated with ivermectin and a second group of calves was treated with moxidectin. The third group of calves remained untreated to serve as a control.

After treatment was applied to each group of calves, data were collected on each calf within each treatment or control group for 26 consecutive d (30 d after the last pretreatment larval infestation). The post-treatment evaluation period was based on the reported female detachment profile of *B. microplus* stating that ≥95% of all engorged females will detach between 20 and 26 d following their infestation, ≥98% will detach between 20 and 30 d following infestation, and virtually 100% will detach between 20 and 32 d after infestation as larvae (Hitchcock 1955). All females that detached from each calf within each treatment or control group were collected and counted daily. A random sample of ≤ 10 engorged females (whenever possible) was saved each day from each calf to obtain weight, fecundity, and fertility data. The sampled females from each calf on each day were weighed collectively, placed in a 9-cm diameter petri-dish, placed in an incubation chamber at 27 ± 2 °C, 92% RH, under a 12:12 h photoperiod (L:D) and allowed to oviposit undisturbed for 20 d after which the spent females were discarded and eggs were weighed, placed in a coded 25 × 93 mm (8-dram) shell vial, and returned to the incubator. At 4 wk after eggs were weighed, the percentage egg hatch of each sample group was visually estimated using a stereo-microscope with an ocular grid, such that the proportion of larvae was compared to the proportion of unhatched eggs within the sample vial.

Once all data (tick counts, egg mass weight, and percentage hatch) were collected over the evaluation period (26 d post-treatment), the index of fecundity (IF) was calculated for each calf on each day using the formula reported by Davey et al. (2001). This formula takes into account the number of ticks collected, the mean egg mass weight of the sampled females, and the mean percentage hatch of the sampled ticks. Thus, the calculated IF is an estimate of the reproductive capacity of a given number of females that lay a given quantity of eggs (based on weight) with a given hatch level. In addition to the calculated IF values, the biological data (female weight and egg mass weight) of sampled ticks from each calf were used to determine whether the endectocide treatments produced any measurable effect on weight and fecundity of females that survived the treatment.

The daily IF values for each calf on each day were summed over the entire 26 d evaluation period to obtain a total IF value. The total IF values for each calf in the control group were then averaged to obtain a mean IF for the group. This mean IF value for the control group was then compared to the total IF value for each calf within each of the two treated groups to provide the percentage control achieved with each endectocide using a modified Abbott's formula (Abbott 1925), as reported by Davey et al. (2001).

The 7-d interval between each pretreatment larval infestation coupled with Hitchcock's (1955) report that ≥95% of all ticks infested at a given time will detach within 20–26 d after infestation provided a means of classifying and analyzing the effect of each endectocide treatment on each stage of the tick (adult, nymph, and larva) individually. Engorged females that detached on days 2–8 post-treatment were considered to have been in the adult stage of development on the day of treatment, since they were detaching at 20–26 d after infestation (18-d pretreatment infestation), while engorged females collected on days 9–15 post-treatment were considered to have been nymphs on the day of treatment and were detaching 20–26 d after they were infested (11-d pretreatment infestation). Lastly, engorged females collected on days 16–26 post-treatment were considered to have been in the larval stage at the time of treatment, since they were detaching 20–30 d after they were infested (4-d pretreatment infestation), at which time ≥98% would have detached from the animals (Hitchcock 1955). Following calculation of the daily IF values for both untreated and treated calves, the sums were partitioned to provide total IF per calf for each of the three intervals that corresponded to the three life stages of the tick at the time of treatment. A mean total IF value was calculated for the control group for each parasitic life stage, which was then compared to the total IF value of each calf within each treatment group having the same parasitic stage designation.

The same cattle that were used in the therapeutic portion of the study were used to evaluate the persistent (residual) efficacy of the two endectocides. The persistent efficacy evaluation was based on a series of larval infestations that were applied to the cattle at regular intervals after treatment was applied. Each calf was infested 1 wk following treatment with ≈2500 larvae that were 5-wk-old. Additional post-treatment infestations using the same number of larvae that were 6–8-wk-old were made at 2–4 wk post-treatment. Beginning on Day 27 post-treatment and continuing through Day 60 post-treatment, engorged females that detached from each animal were collected and recorded. Daily random samples of females (≤ 10 females) were obtained from each calf, and egg mass weight and egg hatch was determined as described above. Once all data had been obtained, the IF of ticks from each calf on each day between 27 and 60 d following treatment was calculated as described previously. Again, the 7-d interval between infestations coupled with the *B. microplus* profile of >95% detachment within 20–26 d post-infestation (Hitchcock 1955) was utilized to classify the origin of ticks with respect to one of the four weekly post-treatment infestations. As such, females collected on days 27–33, 34–40, and 41–47 originated from the 1st, 2nd and 3rd weekly post-treatment infestations, respectively, since detachment of these females was occurring 20–26 d after their infestation. Classification of ticks from the 4th weekly post-treatment infestation was slightly different, where females collected on days 48–60 post-treatment were categorized as being from the 4th post-treatment infestation (detachment occurred at 20–32 d after infestation), at which time all ticks had detached from the animals (Hitchcock 1955). To determine the persistent (residual) efficacy of the two endectocides at 1–4 wk following treatment, the mean IF value of the control group in each of the four weekly classification intervals was compared to the total IF value of each calf in each treatment group having the same weekly post-treatment classification interval, as described previously.

The measured variables in the therapeutic portion of the study (number of ticks per calf, percentage control of the IF, female weight, and egg mass weight) were analyzed by General Linear Model (GLM), one-way analysis of variance (ANOVA) (SAS 1999). Data used to evaluate the stagewise and persistent efficacy of the two endectocides were subjected to GLM using either a one-way ANOVA (number of ticks per calf and IF in the stagewise analysis) or two-way ANOVA (control of the IF in the stagewise and persistent efficacy analyses) with main effects of the two-way analyses being parasitic stage by treatment compound (stagewise efficacy) or treatment compound by week of post-treatment infestation (persistent efficacy), then differences among means in all analyses were determined by Tukey's test method (SAS 1999).

Results

In the therapeutic portion of the study significantly fewer ($F = 36.9$; df $= 2,9$; $p < 0.0001$) engorged females were recovered from both treated groups of cattle than were obtained from the untreated group, but neither the ivermectin or moxidectin treated group was different ($p > 0.05$) from the other (Table 1). As compared to the untreated group, the ivermectin treatment produced a 94.7% reduction in the mean number of females per calf, whereas treatment with moxidectin resulted in a 91.1% reduction in female number, meaning that these percentages of ticks were killed outright before they reached repletion. There was no difference ($F = 0.1$; df $= 1,6$; $p > 0.8$) in the overall therapeutic level of control between the ivermectin and moxidectin treatment, where the levels of control were 99.0 and 99.1%, respectively.

The mean weight of engorged females recovered from untreated cattle was significantly greater ($F = 795.6$; df $= 2,203$; $p < 0.0001$) than the engorged

Table 1. Mean ± SD tick number per calf and percentage control of the index of fecundity (IF) of *Boophilus microplus* on untreated cattle and cattle treated with a single subcutaneous injection of ivermectin or moxidectin at 200 μg AI per kg body weight

Treatment	*n*	Number of ticks per calf	Control of the IF (%)
Untreated	4	1844 ± 558 a	–
Ivermectin	4	98 ± 76 b	99.0 ± 1.0 a
Moxidectin	4	165 ± 35 b	99.1 ± 1.0 a

Means for each parameter were tested by GLM one-way ANOVA; means within the same column followed by the same letter are not significantly different ($p = 0.05$) tested by Tukey's method. Number of ticks per calf, $F = 36.9$; df = 2,9; $p < 0.0001$. Control of the IF, $F = 0.1$; df = 1,6; $p > 0.8$.

Table 2. Mean ± SD female weight and egg mass weight of *Boophilus microplus* females recovered from untreated cattle and cattle treated with a single subcutaneous injection of ivermectin or moxidectin at 200 μg AI per kg body weight

Treatment	*n*	Female weight (mg)	Egg mass weight (mg)
Untreated	4	331 ± 47 a	126 ± 42 a
Ivermectin	4	94 ± 42 b	27 ± 19 b
Moxidectin	4	99 ± 33 b	17 ± 14 b

Means for each parameter were tested by GLM one-way ANOVA; means within the same column followed by the same letter are not significantly different ($p = 0.05$) tested by Tukey's method. Female weight, $F = 795.6$; df = 2,203; $p < 0.0001$. Egg mass weight, $F = 292.0$; df = 2,203; $p < 0.0001$.

weight of females recovered from calves treated with either endectocide, which were not different ($p > 0.05$) from each other (Table 2). The females recovered from both groups of endectocide treated calves weighed ≈3-times less than ticks recovered from untreated cattle. Similarly, the egg masses derived from females recovered from treated calves weighed significantly less ($F = 292.0$; df = 2,203; $p < 0.0001$) than egg masses derived from untreated females. The mean weight of egg masses derived from ivermectin treated animals was ≈5-times less than that of untreated females, while moxidectin treated females produced egg masses that weighed ≈8-times less than those derived from untreated females.

Analysis (one-way ANOVA) showed that ticks treated with either endectocide during each of the three developmental stages (adult, nymph, or larva) produced significantly fewer (adult: $F = 10.4$; df = 2,9; $p < 0.005$; nymph: $F = 49.4$; df = 2,9; $p < 0.0001$; larva: $F = 52.0$; df = 2,9; $p < 0.0001$) females than were recovered from untreated cattle, but were not different ($p > 0.05$) from each other (Table 3). Similarly, one-way ANOVA of the IF of the surviving females treated with either endectocide at each developmental stage was also significantly lower (adult: $F = 7.1$; df = 2,9; $p < 0.02$; nymph: $F = 20.4$; df = 2,9; $p < 0.006$; larva: $F = 72.8$; df = 2,9; $p < 0.0001$) than the IF of untreated females at each stage of development, but no difference

Table 3. Mean ± SD of number of ticks per calf, IF and percentage control of the IF of recovered females of *Boophilus microplus* that were in different parasitic stages on untreated cattle and treated cattle when a single subcutaneous injection of ivermectin or moxidectin was administered at 200 μg AI per kg body weight

Parasitic stage at treatment	Treatment	Number of ticks per calf[A]	IF[A]	Control of the IF[B] (%)
Adult	Untreated	447 ± 275 a	42.808 ± 32.230 a	–
	Ivermectin	1 ± 1 b	0.009 ± 0.019 b	> 99.9 ± 0.1 a
	Moxidectin	1 ± 1 b	0.001 ± 0.001 b	> 99.9 ± 0.01 a
Nymph	Untreated	604 ± 166 a	67.386 ± 29.781 a	–
	Ivermectin	4 ± 4 b	0.040 ± 0.052 b	> 99.9 ± 0.1 a
	Moxidectin	30 ± 17 b	0.213 ± 0.167 b	99.7 ± 0.2 a
Larva	Untreated	793 ± 171 a	104.524 ± 23.875 a	–
	Ivermectin	93 ± 72 b	2.189 ± 2.064 b	97.9 ± 2.0 b
	Moxidectin	134 ± 36 b	1.737 ± 1.899 b	98.4 ± 1.8 b

[A] Means tested by GLM one-way ANOVA; means within the same column and stage followed by the same letter are not significantly different ($p = 0.05$) tested by Tukey's method. Number of ticks per calf, Adult: $F = 10.6$; df = 2,9; $p < 0.005$; Nymph: $F = 49.4$; df = 2,9; $p < 0.0001$; Larva: $F = 52.0$; df = 2,9; $p < 0.0001$. IF, Adult: $F = 7.1$; df = 2,9; $p < 0.02$; Nymph: $F = 20.4$; $p < 0.006$; Larva: $F = 72.8$; df = 2,9; $p < 0.0001$.
[B] Means tested by GLM two-way ANOVA; means followed by the same letter are not significantly different ($p = 0.05$) tested by Tukey's method.

($p > 0.05$) was observed between the treated groups at any life stage. Analysis (two-way ANOVA) of the control of the IF obtained from a single injection of either ivermectin or moxidectin on each of the parasitic life stages of the tick (adult, nymph, and larva) showed that both endectocides provided significantly higher control ($F = 6.9$; df = 2,18; $p < 0.006$) against ticks that were in either the adult or nymphal stage of development (≥99.7%) than was observed against ticks that were larvae (97.9 and 98.4% control for ivermectin and moxidectin, respectively) at the time treatment was applied. However, there was no difference ($F = 0.1$; df = 1,18; $p > 0.8$) in the treatment effect of either endectocide, regardless of life stage, and there was not a significant interaction ($F = 0.3$; df = 2,18; $p > 0.7$) between treatment compound and parasitic stage.

The results obtained from the analysis of persistent (residual) efficacy (two-way ANOVA) of a single injection of ivermectin or moxidectin showed a significant treatment effect on the level of control that was achieved ($F = 12.6$; df = 1,24; $p < 0.002$), where the moxidectin treatment provided a higher level of control at each weekly post-infestation interval than was obtained from the ivermectin treatment (Table 4). The week at which the post-treatment larval infestation was made also produced a significant effect ($F = 16.4$; df = 3,24; $p < 0.0001$) on the level of control, regardless of the endectocide. The control obtained against larvae infested at 1 wk post-treatment was significantly higher than all other post-treatment larval infestations. Similarly, the level of control against ticks infested at 2 wk post-treatment was higher than that of ticks infested at 4 wk post-treatment, whereas against larvae infested

Table 4. Mean ± SD percentage control of the index of fecundity (IF) of *Boophilus microplus* females that survived to repletion from larval infestations applied to untreated and treated cattle at weekly intervals following a single subcutaneous injection of ivermectin or moxidectin at 200 μg AI per kg body weight

Post-treatment larval infestation (week)	Control of the IF (%)		Analysis between treatments at each weekly interval	Analysis within treatments at each weekly interval
	Ivermectin	Moxidectin		
1	82.4 ± 22.1	92.4 ± 8.7	*	a
2	23.3 ± 38.3	79.8 ± 17.2	*	b
3	12.5 ± 15.7	43.8 ± 30.1	*	bc
4	0.0 ± 0.0	19.5 ± 29.9	*	c

Means tested by GLM two-way ANOVA; tested by Tukey's method ($p = 0.05$). There was a significant treatment effect ($F = 12.6$; df = 1,24; $p < 0.002$) at each weekly interval; asterisk indicates significant difference ($p < 0.05$) between treatments at the indicated weekly interval. There was a significant week effect, regardless of endectocide ($F = 16.4$; df = 3,24; $p < 0.0001$); effects shown in last column for both endectocides; the same letters indicate no difference. There was not a significant treatment × week interaction ($F = 1.5$; df = 3,24; $p > 0.2$).

3 wk post-treatment the level of control was not different from larvae infested at either 2 wk or 4 wk post-treatment. There was not a significant interaction between main effects (treatment by week of post-treatment infestation) for the level of control ($F = 1.5$; df = 3,24; $p > 0.2$).

Discussion

The high level of therapeutic control achieved with a single subcutaneous injection of ivermectin or moxidectin (99.0 and 99.1%, respectively) was a consequence of two factors that suffered upon comparison of endectocide treated ticks to untreated ticks, namely the reduction in tick numbers (outright kill) on the cattle and the reduction in reproductive capacity (IF) of females that did survive to repletion following treatment. Results indicated that a high percentage of ticks treated with ivermectin or moxidectin (94.7 and 91.1%, respectively) were killed outright before they were able to reach repletion, and the reduction in reproductive capacity of females that survived to repletion following treatment with either endectocide was ≥99%. In addition, data associated with the female and egg mass weight of treated females clearly indicated that treatment with either endectocide had a dramatic adverse effect on the fecundity of treated females, reducing the weight of females by ≈3-fold and the weight of egg masses by ≈5–8-fold. These results compared favorably with the 99.4% reduction in tick numbers reported by Maske et al. (1992) following an injection treatment with ivermectin. A previous study conducted to evaluate a single injection of ivermectin (Cramer et al. 1988a) reported a lower reduction in tick numbers and reproductive capacity of treated females than the data obtained herein, while reported mean female weight and egg mass weight of treated females was comparable to our study. Caproni et al.

(1998) reported considerably lower reduction in tick numbers at 12 and 28 d following an injection treatment with ivermectin (83.2 and 63.8%, respectively). Studies conducted in Australia and Mexico that evaluated the moxidectin injectable also compared favorably with findings in this study, reporting reduction in tick numbers that ranged from 95 to 100% for up to 28 d following treatment (Remington et al. 1997; Aguilar-Tipacumu and Rodriguez-Vivas 2003).

It is important to note that the timing for administering treatments to cattle in this study on the 18th day following the first pretreatment infestation may have resulted in an overestimation of the therapeutic level of control. As reflected by the fact that no engorged females were recovered from treated cattle until 2 d after treatment, it could be assumed that virtually none of the adult females were in the rapid engorgement phase of development at the time the two treatments were applied. Thus, there was a 24–48 h time period following treatment during which absorption of the AI into the blood and lymph of treated calves would have been increasing to efficacious levels prior to female ticks ingesting large quantities of endectocide contaminated blood during the rapid engorgement phase of development. If treatments had been administered on the twentieth day after the first pretreatment infestation, it is possible that some of the engorging females would not have remained on the host for a long enough period to ingest an adequate quantity of endectocide necessary to kill them or have as great an impact of their fecundity or fertility. Consequently, while it was clearly evident that the endectocides were highly efficacious against ticks that were infested at ≤ 18 d before treatment, the efficacy of the materials against ticks infested 19–20 d before treatment might be less certain. Reports of other investigations with these endectocides stating that the level of control was somewhat lower against ticks that were very near to repletion when treatment was applied support this idea (Cramer et al. 1988a; Davey and George 2002).

The persistent (residual) efficacy of the two endectocides produced remarkably different results. The moxidectin treatment produced significantly higher control at each of the four weekly post-treatment larval infestations than was observed in the ivermectin treatment, indicating that moxidectin remained in the blood stream of treated cattle at a high enough level to adversely affect ticks for a substantially longer period of time than ivermectin. However, results indicated that the level of the endectocides in the blood of treated cattle remained above the threshold level needed for eradication-type control (≥99%) for only a few days following treatment (therapeutic efficacy), then dropped below levels that would sustain the 99% control necessary for eradicating ticks on the calves. Hence, neither endectocide provided the necessary protection against re-infestation for even 1 wk following treatment. It is interesting to note that coumaphos, like both endectocides evaluated in this study, also has a residual activity that is < 7 d following treatment, therefore, due to the 21 d development time of ticks on the host, procedures used by the US CFTEP require that livestock be re-treated with coumaphos at 14 d intervals to prevent any viable ticks from reaching repletion between treatments. In a program where control (≥80% efficacy) is the objective, moxidectin would be the clear choice for a period of up to 2 wk following treatment, whereas ivermectin would sustain an adequate level of control (≥80%) for only 1 wk following treatment. This is supported by a study in which it was reported that the use of subcutaneous injections of moxidectin applied at 4-wk intervals provided very effective control of cattle fever ticks for up to 79 d (Sibson 1994). However, in an eradication program where total control (≥99%) is the goal, re-treatment of cattle at frequent intervals ($<$ 21 d intervals) would be necessary to ensure that virtually no viable ticks would be able to survive to repletion. The results on persistent activity of the two endectocides evaluated in the study compared favorably with other studies that reported persistent activity at ≥80% could only be sustained for 1–2 wk following treatment (Cramer et al. 1988b; Caproni et al. 1998; Aguilar-Tipacamu and Rodriguez-Vivas 2003).

Under present procedures, when US premises are determined to be infested with ticks the owner may either vacate the infested area of all livestock for a period of 6–9 mo. (depending on the time of year the infestation is detected) or, if the owner wishes to maintain cattle on the infested premises, all livestock must be treated every 14–17 d in coumaphos at a concentration of 0.165% AI for a period of 6–9 mo., thus requiring 14–22 treatments (depending on the time of year). Repeated dipping treatments at 14 d intervals are conducted since continual exposure of ticks to coumaphos at short intervals is potent enough to prevent any viable ticks from reaching repletion. Based on the results of this study, injectable formulations of either ivermectin or moxidectin are excellent prospects as an alternative replacement for coumaphos for eradication of *Boophilus* spp. from US premises. Since both of the endectocides evaluated in this study provided ≥99.0% therapeutic control, the systematic repeated application of either endectocide would almost certainly achieve the same results (eradication) as those of coumaphos over the 6–9 mo. quarantine period, even though the same 14 d interval that is presently required for using coumaphos would likely be necessary. Therefore, the use of either of these endectocides as replacements for coumaphos in the above described treatment scenario certainly warrants their serious consideration as an alternative to coumaphos. In addition, although they have no direct impact on the control of ticks *per se*, other added benefits of using these endectocides as a replacement for the dipping of livestock in coumaphos are that labor cost, human exposure to toxic materials, and adverse environmental impact could be substantially reduced by applying injections of these endectocides to treated livestock.

The perspective ramifications associated with the potential use of these endectocides at import facilities is considerably less positive. Based on the previously described rigorous procedures followed at import facilities for allowing entry of cattle into the US, there would be great cause for concern by the CFTEP regarding the potential use of ivermectin or moxidectin, where rapid and complete elimination of *Boophilus* ticks is critical in preventing an inadvertent introduction of viable ticks into the US. While the ≥99.7% therapeutic control obtained against adult and nymphal ticks on the calves would meet the efficacy standards required by the CFTEP for use at import facilities, the 97.9 and 98.4% control for ivermectin and moxidectin, respectively against larval ticks fell below the acceptable standard set for use at ports of entry. Although the efficacy of these endectocides would provide nearly the same level of therapeutic control against adults and nymphs as coumaphos, nevertheless, the difference in efficacy against larval ticks, though subtle, revealed that the endectocides were critically less effective than coumaphos against this development stage. Coumaphos has been reported to be 100% effective against nymphs and larvae (Davey et al. 1997), whereas these endectocides, although virtually 100% effective against adults and nymphs, were significantly less effective against larvae. The importance of this fact, particularly at import facilities, is that larvae, because of their small size, are difficult or impossible to detect on animals during inspection, whereas adults and, to a lesser degree, nymphs, because of their relatively larger size, are more easily detected during the inspection process, which would lead to the cattle being rejected and not allowed to move into the US. Consequently, the ability of any larval ticks to survive to repletion and produce even small numbers of offspring would pose an enormous threat to the success of the CFTEP by providing a source for an outbreak in areas far removed from the permanent quarantine zone. Therefore, the use of these endectocides at import facilities should be carefully considered before recommending them as a replacement for coumaphos. Although it might be possible to reach the necessary level of control against larval ticks if a higher concentration (perhaps 400 μg/kg) were used, this assumption would need to be further evaluated and confirmed.

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