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The effect of anthelmintic resistance on the productivity in feedlot cattle

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ABSTRACT

The aim of this study was to evaluate, in a commercial feedlot, the effect of different anthelmintic drugs on the productivity of naturally infected calves from a cow–calf operation, where resistance to ivermectin (IVM) has been previously detected. The study began with the random selection of 80 calves whose weight was 132 ± 12 kg. Four groups were made: IVM, ricobendazole (RBZ), ricobendazole plus levamisol (RBZ + LEV) and a control group (CG) without treatment. On days 0, 21, 42, 70, 98 and 126, manual collection of fecal matter and individual weight were registered. Mixed SAS procedure was used for statistical analysis. The percentages of fecal egg count reduction test (FECRT) calculated 21days post treatment (PT) were 18%, 96% and 100% for the IVM, RBZ and RBZ + LEV groups, respectively. Body weight (± SEM) at the end of the trial was 266 kg (±0.9), 269 kg (±1.1), 276 kg (±1.3), 280 kg (±1.9) for CG, IVM, RBZ and RBZ + LEV groups, respectively. The effect on live weight was highly significant ($p < 0001$).

After 126 days of fattening, the deleterious effect of the combination of *Cooperia* and *Haemonchus* in the IVM group on body weight was evident. Undetected animals carrying anthelmintic resistant (AR) worms entering the feedlot, could cause major productivity losses.

Introduction

Gastrointestinal nematodes (GIN) are an important health problem and cause a significant productivity loss in growing calves (Waller, 2003; Entrocasso et al., 2008). The consequences vary from lower daily weight gain, lower milk production up to animal death (Ames et al., 1969; Entrocasso, 1994; Guzmán et al., 2010). GIN control has been based mainly on the use of anthelmintic drugs, regardless of other epidemiological factors such as parasitic population in shelters (non-drug-exposed), pasture rotation, rainfall and susceptibility according to animal category (Cristel and Suárez, 2006; Sievers and Alocilla, 2007). Prevention and control strategies using chemicals continuously and indiscriminately has favored the selection of resistance in cattle nematodes (Cristel and Suárez, 2006; Kaplan and Vidyashankar, 2012). Anthelmintic resistance (AR) has been reported in North and South America, Africa, Oceania and Europe (van Wyk et al., 1999; Anziani et al., 2001, 2004; Fiel et al., 2001; Coles, 2002; Gasbarre et al., 2009a; Mejia et al., 2003; Demeler et al., 2009; Jackson et al.,

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2006; Waghorn et al., 2006; Soutello et al., 2007; Suarez and Cristel, 2007). In the Northeast of Argentina IVM is used to control the common tick *Rhipicephalus* (*Boophilus*) *microplus*, which contributes to AR due to regular and long exposure of GIN to these compounds (Coles, 2002; Caracostantogolo et al., 2005). Combination of anthelmintic drugs from different chemical groups has been suggested as an alternative parasite control strategy where failure of individual drugs is documented (Entrocasso et al., 2008; Gasbarre et al., 2009b; Bartram et al., 2012).

In Argentina, more than 1.100.000 animals a year are fattened in feedlots (Rossanigo et al., 2010). Weight loss due to lack of drug efficacy as a result of AR could be substantial in these production systems (Fazzio et al., 2012).

The aim of this trial was to test, in a commercial feedlot, the effect of different anthelmintic drugs on the productivity of naturally infected calves from a cow–calf operation, where resistance to IVM has been previously detected.

Materials and methods

This study was carried out in 2013, from April to August, in a commercial feedlot in the city of Marcos Paz (−34°08′S, 58°08′W), Province of Buenos Aires. Calves came from a sub-tropical area near the city of Esquina (27°27′S, 58°49′W), Province of Corrientes, where cattle and sheep coexist in the same fields. *Bos indicus* × *Bos taurus* crossbred animals had a history of frequent tick treatments with IVM, which led to AR in GIN. After being transported 900 km to the south, animals arrived at the feedlot and remained in pens where they had access to rolls of hay and fresh water for 72 h. The sanitary routine was one dose of polyvalent clostridial vaccine (Policlostrigen®-Biogénesis-Bagó) and an injectable broad-spectrum antibiotic (Tilmicosina, Maxityl®-Biogénesis-Bagó). The diet was based on corn grain, sunflower meal, wheat bran and vitamin-mineral supplement. Protein and fiber content in the initial diet was 15% and 25%, respectively. Then, it decreased progressively to 12% and 8% in the finishing diet.

The study began 72 h after arrival, when 80 calves in good health, aged between 4 and 5 months and whose weight was 132 ± 12 kg were randomly selected. All calves were given an individual ear-tag number. On trial day 0, four groups were made: one group was injected 0.2 mg/kg b.w. of ivermectin1% (Ivomec® Merial-IVM); another received 7.5 mg/kg b.w. of ricobendazole 10% (Axilur PI® Intervet-RBZ); a third group was treated with 7.5 mg/kg b.w. of RBZ and 8 mg/kg b.w. of levamisole (Axilur PI® Intervet + Fosfamisol MV® Biogénesis-Bagó-RBZ + LEV). The control group (CG) remained without treatment. On days 0, 21, 42, 70, 98 and 126, manual collection of fecal matter from the rectum and individual weight were registered. Individual egg counts were performed using the modified McMaster technique (Roberts and O'sullivan, 1950) where each egg counted represented 10 eggs/gram of faeces. For third-stage larvae differentiation in every group, pooled fecal culture was carried out twice (Niec, 1968; van Wyk et al., 2004).

The statistical analyses were run with the mixed procedure of SAS (9.1) including the random effect for each animal (experimental unit), and the fixed effects of time (0 vs. 21 vs. 42 vs. 70 vs. 98 vs. 126), treatment (CTL vs. IVM vs. RBZ vs. RBL + LEV) and their interaction (time × treatment). The Slice option was used to detect statistical differences at every time point.

From a total of 20 animals per group, 14 animals with EPG > 120 and <1840 at day 0 were selected to evaluate the dynamics of EPG and FECRT. Finally, the percentages of FECRT were calculated by means of the formula provided by Coles et al. (1992), where reduction percentage is $100 \times [1-(T/C)]$, T is the arithmetic mean of the pos-treatment (PT) group on day 21 and C is the EPG arithmetic mean of the CG on day 21 PT. For statistical analysis, the program FECRT v4.0 available on http://www.vetsci.usyd.edu.au/sheepwormcontrol was used.

Results

Time had a highly significant effect on EPG ($p < 0.001$, Fig. 1). According to contrast this effect was highly significant ($p < 0.001$). Treatment also had effect on EPG ($p = 0.045$, Fig. 1). Finally, time × treatment had an effect on EPG ($p < 0.001$, Fig. 1). On PT day 21, FECRT showed significant differences among groups. The IVM showed an 18% decrease, whereas in calves which were administered RBZ + LEV, the decrease reached 100% (Table 1). This demonstrates presence of AR in the IVM group.

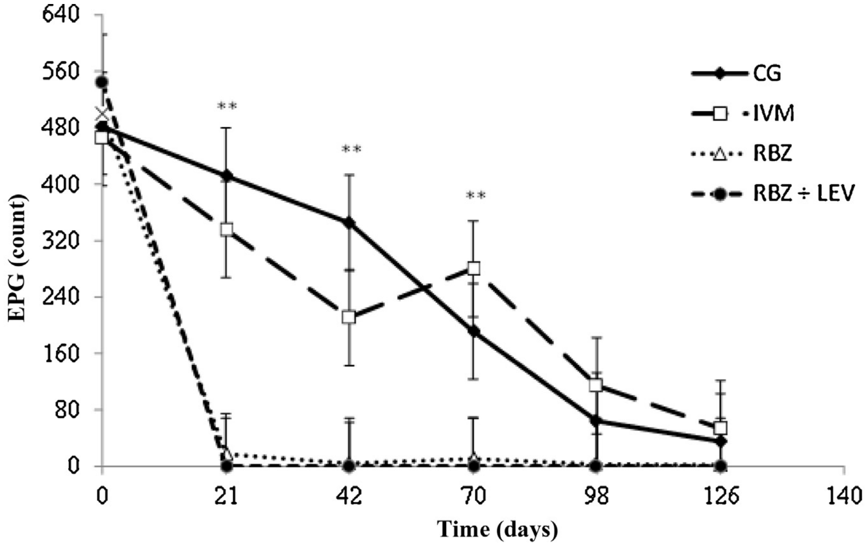
Parasitic genera found in stool cultures of the IVM group were *Cooperia* and – in lesser amount – *Haemonchus* (Table 2). *H. contortus* and *H. placei* L_3 were identified and prevailed according to treatment received (Fig. 2). *H. contortus* L_3 was the only species of this genus recovered after treatment with RBZ, while *H. contortus* and *H. placei* L_3 were recovered in the IVM group coprocultures.

As regards live weight, time had a significant effect ($p < 0.001$, Table 3). Conversely, treatment had no effect whatsoever on live weight ($p = 0.482$, Table 3). Finally, time x treatment had an effect on live weight ($p < 0.001$, Table 3).

Discussion

Resistance to macrocyclic lactones in cattle GIN has been referred to in studies carried out in Argentina (Anziani et al., 2001, 2004; Mejia et al., 2003; Fiel et al., 2005). However, it should be highlighted that there are data of AR to other compounds such as benzimidazoles (Anziani et al., 2001; Mejia et al., 2003; Caracostantogolo et al., 2005). Thus, it is necessary to evaluate the efficacy of the anthelmintics used in animals entering a feedlot.

Great amount of calves are kept in feedlots without parasitic control and without clinical signs of parasitism. The results of this trial show that there exists productivity loss and significantly less weight gain in feedlot calves caused by presence of AR to GIN. However, reports of parasitic gastroenteritis outbreaks are unusual (Coles, 2002). A likely explanation is that, at least in Argentine feedlots, re-infestation due to gastrointestinal nematodes hardly occurs and the prevailing genus in feedlot cattle is *Cooperia* due to the extensive use of IVM in cow–calf operations. Besides, *Cooperia* sp, does not show significant clinical features, as it is the case with parasite genera of higher



CG : control group (without treatment);
IVM: Ivermectin group (0.2 mg/kg b.w. of ivermectin 1%);
RBZ: Ricobendazole group (7,5 mg/kg b.w. of ricobendazole 10%)
RBZ + LEV: Ricobendazole + Levamisol (7,5 mg/kg b.w of ricobendazole and 8 mg/kg b.w. of levamisole)
** P value <0.01, of slice option.

Fig. 1. Effect of anthelmintic treatment on fecal eggs per gram in feedlot calves. CTL: Control group (without treatment); IVM: ivermectin group (0.2 mg/kg b.w. of ivermectin 1%); RBZ: ricobendazole group (7.5 mg/kg b.w. of ricobendazole 10%); RBZ + LEV: ricobendazole + levamisol (7.5 mg/kg b.w. of ricobendazole and 8 mg/kg b.w. of levamisole) ***p*-value < 0.01, of slice option.

pathogenicity such as *Ostertagia ostertagi* (Fiel et al., 2005; Gasbarre et al., 2009b). Nevertheless, production decrease caused by *Cooperia* sp. has already been reported (Herlich, 1965; Keith, 1967; Louvandini et al., 2009; Stromberg et al., 2011) as well as the impact of this genus combined with *Haemonchus* sp. in feedlot calves (Fazzio et al., 2011, 2012). In this trial, the prevailing parasite genus in PT fecal cultures was *Cooperia*, while *Haemonchus* was present in smaller amounts.

FECRT showed the low efficacy of IVM against *Cooperia* and *Haemonchus* genera, since the decrease was only 23% and 0%, respectively (Table 1). AR to IVM is frequent in animals from the Northeast of Argentina, where exposure of GIN to macrocyclic lactones is frequent, since they are used to control *Rhipicepalus microplus* (Coles, 2002; Fiel et al., 2005).

The low efficacy of RBZ and IVM against *Haemonchus* sp. can be explained due to the coexistence of cattle with sheep on the farm of origin and the lack of efficacy of this drug in sheep parasites because of their resistance to it. Different percentages of L_3 of *H. contortus* were found in calves' coproculture according to the anthelmintic drug used (Fig. 2). This finding agrees with Gasbarre et al. (2009a), who recovered *H. contortus* from calves treated with benzimidazoles as well as *H. placei* and *H. contortus* in animals receiving different avermectines. In this trial, L_3 of *H. contortus* in the RBZ group were recovered from the coprocultures of calves until day 70. These data are consistent with those found by Guzmán et al. (2011), who infected calves experimentally with *H. contortus* L_3 and were able to recover adults 45 days post infection, but not in 90 days. This confirms that there is cross-transmission between sheep and cattle, although these parasites have a short half-life in bovines. However, *H. contortus* has been recovered from calves grazing pastures without sheep for more than 40 years and was explained by an adaptation

Table 1
Percentage of egg count reduction in feedlot calves 21 days post-treatment receiving ivermectin, ricobendazole and ricobendazole + levamisole.

	All genera	*Cooperia* Mean (95% CI)	*Haemonchus*
IVM[a]	18% (0–56)	23% (0–59)	0% (0–29)
RBZ[b]	96% (86–99)	98% (93–99)	72% (17–92)
RBZ + LEV[c]	100% (100–100)	100% (100–100)	100% (100–100)

Mean (95%CI): mean egg count reduction expressed in percentage (95% confidence intervals).
[a] IVM: Ivermectin group (0.2 mg/kg b.w. of ivermectin 1%).
[b] RBZ: Ricobendazole group (7.5 mg/kg b.w. of ricobendazole 10%).
[c] RBZ + LEV: Ricobendazole + Levamisole (7.5 mg/kg b.w. of ricobendazole and 8 mg/kg b.w. of levamisole).

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Table 2
Effect of anthelmintic treatment on the percentage of recovered larvae in feedlot calves.*

		Day					
		0	21	42	70	98	126
CTL	Coop[1]	91	92	90	95	97	97
	Haem[2]	9	8	10	5	3	3
	n° L_3 identified	211	200	190	200	200	200
IVM	Coop[1]	91	87	95	93	100	100
	Haem[2]	9	13	5	7	0	0
	n° L_3 identified	211	200	180	186	190	200
RBZ	Coop[1]	91	46	55	83	90	100
	Haem[2]	9	54	45	17	0	0
	Trich[3]	0	0	0	0	10	0
	n° L_3 identified	211	52	33	29	10	6

CTL: Control group (without treatment); IVM: ivermectin group (0.2 mg/kg b.w. of ivermectin 1%); RBZ: ricobendazole group (7.5 mg/kg b.w. of ricobendazole 10%).
* Data of ricobendazole + levamisole group (RBZ + LEV) not shown because were not recovered L_3.
[1] Coop: *Cooperia* sp.
[2] Haem: *Haemonchus* sp.
[3] Tricho: *Trichostrongylus* sp.

Table 3
Effect of antiparasite treatment on live weight in feedlot calves.

	Day (LSM kg ± SE)					
	0	21	42	70	98	126
CTL	134 ± 2.1[a]	149 ± 1.8[a]	181 ± 1.6[a]	219 ± 2.0[a]	242 ± 1.1[a]	266 ± 0.9[A]
IVM	132 ± 2.0[a]	149 ± 1.5[a]	184 ± 1.5[a]	222 ± 1.8)[a]	243 ± 0.8[a]	269 ± 1.1[A]
RBZ	132 ± 1.8[a]	153 ± 2.1[ab]	185 ± 1.2[ab]	225 ± 1.5[ab]	249 ± 1.5[b]	276 ± 1.3[B]
RBZ + LEV	131 ± 1.9[a]	155 ± 1.9[b]	189 ± 0.7[b]	227 ± 1.1[b]	252 ± 1.6[b]	280 ± 1.9[B]

CTL: control group (without treatment); IVM: ivermectin group (0.2 mg/kg b.w. of ivermectin1%); RBZ: ricobendazole group (7.5 mg/kg b.w. of ricobendazole 10%); RBZ + LEV: ricobendazole + levamisol (7.5 mg/kg b.w. of ricobendazole and 8 mg/kg b.w. of levamisole). Different letters within column means $p < 0.05$. Different capital letters within column means $p < 0.01$.

of the cycle of the sheep parasite in cattle (Gasbarre et al., 2009a).

The decrease in FECRT in the RBZ and RBZ + LEV groups was higher than 95%. Significant productivity loss due to GIN is unlikely to occur with such percentage of efficacy (Coles et al., 2006). In this trial, groups showing a higher percentage of FECRT also showed a better yield in live weight (Tables 1 and 3). The combined pathogenic effect of *Cooperia* and *Haemonchus* was observed even after 126 days of fattening. There is a difference of 11.4% and 8.3% in b.w. kg when the IVM group is compared with the RBZ + LEV and with the RBZ group, respectively. Reinhardt

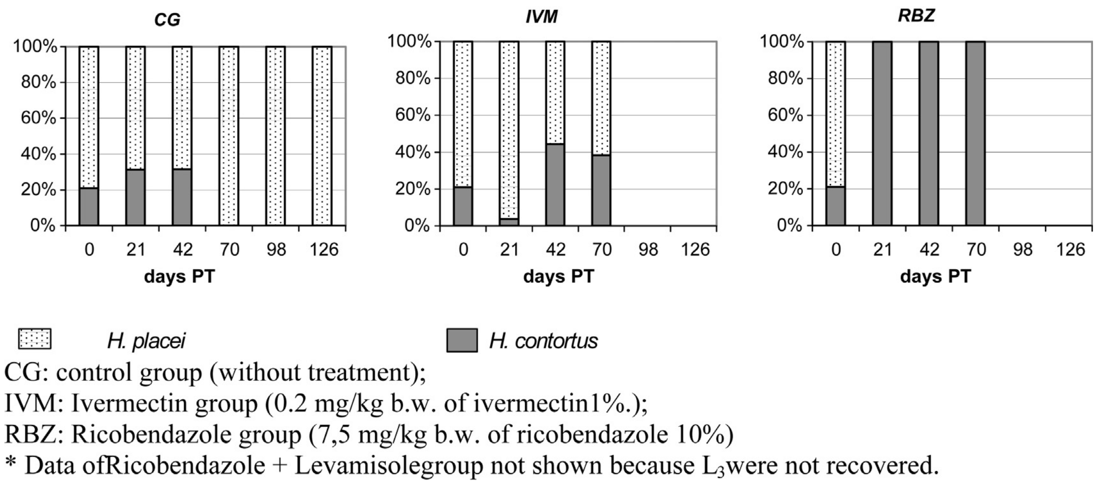

Fig. 2. *Haemonchus* species (L_3) identified in feedlot calves in groups ivermectin, ricobendazole and control*. CG: Control group (without treatment); IVM: ivermectin group (0.2 mg/kg b.w. of ivermectin 1%); RBZ: ricobendazole group (7.5 mg/kg b.w. of ricobendazole 10%). *Data of ricobendazole + levamisole group not shown because L_3 were not recovered.

et al. (2006) noted daily weight loss in feedlot, even with a low worm count and absence of highly pathogenic parasite genera such as *Ostertagia ostertagi*. The study also suggests that drug combination improves antiparasite efficacy and that the rapid effect of bencimidazoles against GIN could benefit weight yield.

EPG counts in feedlot animals decrease over time, even in groups under no anthelmintic treatment (Smith et al., 2000). EPG count in this study also diminished as calves remained in feedlots, regardless of the efficiency of the treatment administered. Nevertheless, EPG count decrease was progressive and hardly significant (Fig. 1), probably because of low immune response to GIN, since animals were young (4–5 months old) and exposed to stressful experiences such as transportation and feedlot adaptation. This phenomenon differs from previous findings by the authors which worked with older animals (Fazzio et al., 2012). The major difference in body weight among groups with low and a high efficacy was observed on PT day 126. On PT day 21, the RBZ group, despite having high antiparasite efficacy, did not show any marked difference with the CG and IVM groups. Presumably, this is because the number of animals used for the test was low. However, in previous trials, significant differences in weight gains were found as from week 3 PT (Fazzio et al., 2011, 2012). Ames et al. (1969) observed in the late months of the 150 days fattening period, compensatory growth in heifers without anthelmintic treatment compared with the treated group. However, the difference was of 29 kg. b.w. that could not be compensated. In Argentina, fattening cycles do not go beyond 100–130 days in the pens and productivity loss due to GIN is unlikely to be compensated in the case of short cycles. Taking into account that weight gain in the different groups, it could be the case that GIN were not eliminated during the trial. Rather, they got older and oviposition diminished. Despite low EPG count in the IVM and the CG groups near the end of the trial (days 98 and 126), differences in weight remained throughout the observation period and even increased over time. Trials involving necropsy and counting parasite burdens would be necessary to know how parasite populations behave while animals remain in the feedlot (mainly when the correlation between parasite burden and EPG is missed) as it occurs when parasites get older due to absence of reinfestation and the greater influence of the immune system on oviposition (Claerebout and Vercruysse, 2000).

Conclusion

Gastrointestinal parasitism in feedlot has a negative impact on productivity. Greater AR makes it essential to carry out PT controls to test the real anthelmintic effect on the parasites in animals finished in feedlots. Failures in anthelmintic treatment may cause losses of more than 10 kg b.w. per animal in each fattening cycle, even when the genus involved is mainly *Cooperia* sp.

Conflict of interest

The authors have no conflicts of interest.

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