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Short Communication

Fifteen years later, anthelmintic resistances have dramatically spread over goat farms in Guadeloupe

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ABSTRACT

Faecal egg count reduction tests (FECRTs) were performed on 21 goat farms in Guadeloupe (FWI). Anthelmintic resistance (AR) to netobimin (benzimidazole) was found in all 15 herds in which it was tested. AR to ivermectin (avermectin) and levamisole (imidazothiazole) were also very largely spread (14 out of 17 farms and 7 out of 9 farms, respectively). AR to the final moxidectin (milbemycin) released was already present in 2 out of 9 farms in which it was tested. *Haemonchus* was the dominant genus of gastrointestinal nematodes and was more frequently found to be resistant to netobimin, ivermectin and moxidectin than *Trichostrongylus*, the latter appeared to be more often resistant to levamisole. A first survey 15 years ago revealed only AR to benzimidazoles and one suspected case of AR to ivermectin.

1. Introduction

Gastrointestinal nematode (GIN) parasitism is probably the most common disease in the small ruminant industry in the entire humid tropics. Since the 1970s in the French West Indies, the implementation of various programmes for the development of sheep or goat production has been reliant on, among others, the systematic use of anthelmintic drugs. In many countries, such drenching policies have resulted in the selection of anthelmintic resistant (AR) strains of parasites, as reviewed by Kaplan (2004) and others (Jabbar et al., 2006; Papadopoulos et al., 2012; Torres-Acosta et al., 2012).

The overall goat population of Guadeloupe (16° N, 61° W) was officially estimated to be about 16,000 heads in 2010, mainly in the driest part of the island (2010 agricultural census, available at https://stats.agriculture.gouv.fr/disar/). As in many other Caribbean islands, most of the estimated 1000 goat farmers own less than 10 heads and almost all the goat keepers have other sources of income. About 90 farmers are members of the cooperative of small ruminant farmers (Cabricoop), with only a few of them owning more than 100 goat heads. The different anthelmintic families were successively introduced in Guadeloupe. The first ones were the benzimidazoles: since the 1970s mainly thiabendazole followed by fenbendazole, mebendazole and albendazole and lastly netobimin, an albendazole precursor which was only used occasionally for cost reasons. Levamisole (imidazothiazole) was cheap and widely used since the 1980s; ivermectin (avermectin) was the lead anthelmintic since the 1990s; and moxidectin (milbemycin) was the final to be released in Guadeloupe, during the last decade. A first survey of AR status in goat farms was carried out in 1994–1996 (Barré et al., 1997) and revealed that resistances to benzimidazoles were already

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wide spread in Guadeloupe and that AR to ivermectin was suspected in at least 1 of 16 farms.

The objectives of the present survey were: (i) to provide farmers, vets and extension officers with information about the efficacy of the drugs currently used in each farm and about the drugs used in the past years; and (ii) to update the AR status in goat farms in Guadeloupe 15 years after the first survey (Barré et al., 1997).

2. Materials and methods

The survey was carried out during late 2011 in farms involved in the programme of development of the Cabricoop. Twenty one farms were chosen in agreement with farmers and according to the availability of animals weighing more than 12 kg (aged over 3 months), in order to test 1–4 different drugs on 10 animals each, with a control group. The farms were spread over the main climatic zones and roughly according to goat density.

The farm visits were planned at least 4 weeks after the last drench to allow a free natural infection of animals before testing the drug efficacy by performing a faecal egg count reduction test (FECRT) following the WAAVP recommendations (Coles et al., 2006).

Four anthelmintics (oral formulation) were chosen in agreement with the farmers and veterinary practitioners, according to their present and past utilisation in farms, namely netobimin (Hapadex® 50 mg/ml, Schering-Plough Santé Animale 49500 Sergé, France), which is considered as representative of the whole benzimidazole class, levamisole (Biaminthic® 5%, Laboratoires Biové 62510 Arques, France), ivermectin (Oramec®, Merial 29 Avenue Tony Garnier 69007 Lyon, France) and moxidectin (Cydectine® 0.1%, Pfizer Olot S.L.U. Ctra. Camprodon s/n "La Riba" 17813 Vall de Bianya, Girona, Spain). At the farm level, priority was given to the current anthelmintic, then to either ivermectin or levamisole, and lastly to netobimin.

The individual drug doses were calculated according to the goat live weight which was estimated by measuring the heart (or chest) girth (growing goats) or the heart and paunch girth (adult goats) with a tape measure (Mahieu et al., 2011). Goats were given 1.5 times the sheep dose in order to take into account the specificity of goat pharmacokinetics (Hennessy et al., 1993a,b,c; Sangster et al., 1991; Sanyal, 1996). All of the anthelmintic were given orally: 0.23 ml Hapadex® (11.25 mg netobimin) per kg live weight (kg LW); 0.23 ml Biaminthic® (11.25 mg levamisole) per kg LW; 0.38 ml Oramec® (0.3 mg ivermectin) per kg LW; and 0.30 ml Cydectine® (0.3 mg moxidectin) per kg LW.

For practical reasons, the animals were randomly allocated to experimental groups, weighed and dosed and faecal samples were collected during the first visit of the farm (d0). The second faecal samples were collected 14 days later (d14) according to the WAAVP recommendations (Coles et al., 2006).

The faecal samples (ideally about 10–15 g, sometimes less than 5 g in the field conditions) were taken from the rectum (latex gloves) and kept in plastic tubes to avoid contamination and immediately transported to the laboratory for processing. The faecal samples were kept at ambient temperature until processing to avoid species-related bias in egg hatch and larval development (O'Connor et al., 2006). The individual subsamples for FEC (about 4–5 g each) were precisely weighed (±0.01 g), crushed as soon as possible in 20–30 ml of tap water and stored at 4 °C to stop egg development. GIN eggs were counted within two days using a modified McMaster method. After centrifugation (15 min, 2800 rpm), the sediment was thoroughly mixed with 35 ml of saturated NaCl solution ($d = 1.19$) and centrifuged again to eliminate faecal particles. The homogenised supernatant was then sampled to fill the two cells of a McMaster slide, and GIN eggs were counted under a microscope (40×). The actual sample weight was used for the calculations and each egg counted represented about 30 epg. The remaining faeces were pooled according to the FECRT design and cultured at ambient temperature for 7 days. The GIN infective larva were recovered with a Baerman apparatus, identified at the genus level and counted according to Van Wyk et al. (2004). The FEC attributed to each GIN genus was calculated by using the estimate of the contribution of this genus to the overall infective larvae population.

Only animals with pre-treatment FEC over 150 epg were kept for analysis (Coles et al., 2006).

FEC reduction values were estimated from Cabaret and Berrag (2004), by adapting the individual "iFECRT4" calculation method from Dash et al. (1988) to groups with an unequal number of individuals:

$$\mathrm{FECR}j = 100 \times \left(1 - \left\{ \left(\frac{\frac{1}{nj}\sum_{i=1}^{nj} \mathrm{FEC}ijd14/\mathrm{FEC}ijd0}{\frac{1}{nc}\sum_{i=1}^{nc} \mathrm{FEC}icd14/\mathrm{FEC}icd0} \right) \right\} \right)$$

where ij (nj) represents the ith (number of) individual(s) in the jth treatment, ic (nc) the ith (number of) individual(s) in the control group, d0 and d14 the pre- and post-treatment days of sampling. The 95% confidence interval of FECRj was empirically obtained by a 2000 bootstrap re-sampling of the individuals of both drenched and control groups, for each farm. For the three farms without a control group, the corresponding term of the equation was set to 1 and the calculation was the same than the individual "iFECRT3" calculation method in Cabaret and Berrag (2004), derived from Kochapakdee et al. (1995).

AR was declared when the FECR mean was below 95% and the lower bound of the confidence interval was below 90%. AR was suspected when only one of these conditions is true (Coles et al., 2006).

We used the *R* statistical software (R Development Core Team, 2014) for all calculations.

All of the animal handling and sampling operations complied with the European Union rules.

3. Results and discussion

The small size of the goat farms, the organisation of farm visits and animal handling constraints did not always allow 10 individuals per group; therefore the number of individuals was reported for each group. Overall 482 individuals (79%) with a FEC over 150 epg were kept for FECR