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Short Communication

Efficacy of halofuginone lactate against experimental cryptosporidiosis in goat neonates

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ABSTRACT

Preliminary results obtained in calves, lambs and goat kids infected by *Cryptosporidium* sp. have indicated a partial prophylactic efficacy of halofuginone lactate when administered at 100 μg/kg body weight (BW). In this study, the efficacy of halofuginone lactate was evaluated in goat neonates experimentally inoculated with *Cryptosporidium parvum* oocysts per oral route. The trial consisted in 2 replicated experiments carried out successively at 2 months of interval. Twenty-two 2- to 4-day-old kids were experimentally inoculated once, 2–3 days after the arrival in premises, with 10^6 *C. parvum* oocysts per oral route and were allocated into 2 groups. Animals of group 1 acted as untreated control whereas animals of group 2 received halofuginone lactate for 10 days from the infection day to day 9 post-infection (DPI) at a daily oral dose rate of 100 μg/kg BW. Individual oocyst shedding was monitored by daily examination of faecal smears stained by carbol fuchsin and scored semi-quantitatively (0–5) until 19 DPI. Daily diarrhoea scores, weight gain and mortality were recorded. In the first experiment, oocyst excretion started 1 DPI in the control group, was highest on 4 DPI (mean score 3.6) and became undetectable from 16–19 DPI. In the treated group, oocyst shedding started 1 day later, showed lower scores compared to control on 4, 5, 6, 7 and 10 DPI and vanished from 16 to 19 DPI. No significant difference was seen for weight gains between groups. Five kids died in the control group compared to 1 kid in the treated group. In the second (replicated) experiment, oocyst excretion started 2 DPI in the control group, was highest on 4 DPI (mean score 4.5) and became undetectable 18 and 19 DPI. In the treated group, oocyst shedding started 2 days later, peaked on 13 DPI (mean score 2.3) and persisted until the end of the experiment. No significant difference was seen for weight gains between groups. Ten kids died in the control group compared to 3 kids in the treated group. The results demonstrated the efficacy of halofuginone lactate when given as a prophylactic treatment at 100 μg/kg BW during 10 days in reducing oocyst shedding, diarrhoea and mortality in goat kid cryptosporidiosis.

1. Introduction

Cryptosporidium infection is responsible for diarrhoea, dehydration, weight loss and death in neonatal ruminants (de Graaf et al., 1999). The frequency and the severity of cryptosporidiosis is of a particular concern in goat neonates (Paraud and Chartier, 2012). As a result, the weight loss

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and the mortality (up to 40%) associated with the disease may induce important economic losses for farmers (Naciri et al., 1984; Yvoré et al., 1984; Noordeen et al., 2012). Few herd-level risk factors for *Cryptosporidium* infection in kids have been identified (Delafosse et al., 2006) and implementation of general hygienic measures to control infection remains difficult for farmers. In severe outbreaks, the use of anticryptosporidial drugs is absolutely necessary to control the disease. Many drugs or compounds have been tested against animal cryptosporidiosis and only very few of them have shown some partial effectiveness when administered prophylactically to kids: halofuginone lactate, paromomycin, α- and β-cyclodextrin, activated charcoal and wood vinegar liquid (see Paraud and Chartier, 2012). In France, only halofuginone lactate (Halocur®, MSD Santé Animale) has a licence for the prevention and the treatment of cryptosporidiosis in calves, and the use in other ruminant species as goat kids is feasible "off licence" in the "cascade procedure". Despite a large use by the veterinarians, published data about halofuginone efficacy in kids are scarce and only deal with field studies (Chartier et al., 1999; Giadinis et al., 2008). The objective of the present study was to evaluate the efficacy of halofuginone lactate in preventing cryptosporidiosis in kids in controlled experimental conditions.

2. Material and methods

2.1. Experimental design

For facilities reasons, the study was performed in two identical experiments conducted 2 months apart. The two experiments shared the same experimental design.

In experiment 1, twenty-two male or female 1- to 3-day-old kids of Saanen or Alpine breed were selected at random in three farms where no sign of clinical cryptosporidiosis in kids was detected the preceding years. The neonates were given colostrum before being transported to the experimental premises. Then, the animals were allocated into 2 groups of 11 animals each. Each group was housed in a pen (3 m × 3 m) with straw bedding and neonatal kids were fed a milk replacer twice a day. All the kids were orally inoculated using a 1 ml syringe containing 10^6 oocysts of *Cryptosporidium parvum* (Waterborne Inc, New Orleans, USA) in water 3 days after the arrival (D0 = inoculation day).

Animals of group 1 acted as untreated control group, animals of group 2 received halofuginone lactate for 10 days from day 0 to day 9 at a orally daily dose rate of 100 μg/kg BW. The drug was given individually with a syringe. The duration of administration was extended to 10 days according to previous field clinical trials (Chartier et al., 1999). The halofuginone lactate was given after evening feeding. The treatment was thus administered before oocyst output or clinical signs were present in kids.

The criteria used to assess the efficacy of the treatment were daily oocyst shedding and diarrhoea scoring, weekly weighing and mortality.

Faecal samples were individually taken rectally from all neonatal kids every day from D1 to D19. Faecal smears were stained with carbol fuchsin for visualisation of oocysts (Heine, 1982). Oocyst shedding was scored semi-quantitatively according to the average number of oocysts at 200× magnification: 0 (no oocyst), 1 (≤1 oocyst), 2 (1–10 oocysts), 3 (11–20 oocysts), 4 (21–30 oocysts) and 5 (>31 oocysts) on faecal smear at 200× magnification. 11 goat kids per group

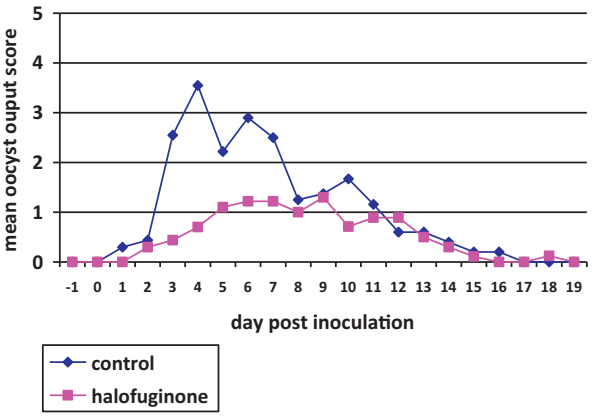

Fig. 1. Mean oocyst output scores in kids experimentally inoculated by *C. parvum* per oral route according to the group (control group, halofuginone treated group): experiment 1. Day 0: day of inoculation (10^6 *C. parvum* oocysts). Day 0 to Day 9: halofuginone lactate (Halocur©) at a daily dose rate of 100 μg/kg BW in the treated group, oral route
Oocyst ouput scores: 0 (no oocyst), 1 (≤1 oocyst), 2 (1–10 oocysts), 3 (11–20 oocysts), 4 (21–30 oocysts) and 5 (>31 oocysts) on faecal smear at 200× magnification. 11 goat kids per group

The experiment was replicated 2 months later with a new mob of kids. All goat kids of Saanen breed came from a unique farm with no recent history of cryptosporidiosis.

2.2. Statistical analysis

Comparisons of mean values in the different groups were made using ANOVA for oocyst shedding and for weight gains, χ^2 analysis for mortality and Kruskal–Wallis for weight gain (Statistica® Ver. 9.01 for Windows®). Statistical significance of the variables was tested at the 0.05 level of confidence.

3. Results

3.1. First experiment

In the control group, oocyst shedding started 1 DPI in 3 animals out of 11, was then maximal 4 DPI (mean score 3.6) with all kids excreting oocysts and decreased thereafter to be undetectable from 16 to 19 DPI (Fig. 1). On 12 and 13 DPI, 50% of kids were still shedding oocyst. Large individual variations in oocyst excretions were seen with more than half of the animals excreting high levels of oocysts (≥3). In the treated group, oocyst shedding started 1 day later, in 2 animals out of 11, peaked 9 DPI (mean score 1.3) with 8 kids out of 10 excreting oocysts and was around zero from 16 to19 DPI. On 12 and 13 DPI, between 20 and 50% of kids were still excreting oocysts. When compared to control, oocyst output was seen later and of lower magnitude.