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Do the changes in the behaviours of cattle during parasitism with *Ostertagia ostertagi* have a potential diagnostic value?

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ABSTRACT

We investigated the magnitude of temporal changes in activity, posture and feeding behaviour of cattle infected with *Ostertagia ostertagi*, and their reversal after treatment with an anthelmintic. Twenty-six, 3-month-old, Holstein-Friesian bulls were allocated to one of three treatment groups. Bulls in two of those (groups P and PA) received 100,000 larvae on three occasions (Days 0, 7 and 14) and the remaining animals served as controls (C). The PA group also received an anthelmintic on Day 31. Parasite eggs appeared in the faeces of P and PA bulls from Day 17; from approximately the same time blood pepsinogen levels increased and body weight (BW) gain decreased ($P < 0.001$). The reduction in BW gain persisted until Day 45 for P animals only. There was a decrease in the number of steps taken for P and PA animals, as well as lying and standing episode frequency, by 41 and 44% respectively ($P < 0.001$) from Day 21 onwards. The average lying and standing episode duration increased by 52 and 55% respectively ($P < 0.001$) from the same time in P and PA compared to C bulls. In addition, meal frequency showed a tendency to decrease for P animals only ($P = 0.039$) from Day 39, and this was the only aspect of feeding behaviour affected by parasitism. All behaviours, returned to control levels within a week of anthelmintic drenching of PA bulls, apart from the number of steps taken. Although BW gain and pepsinogen also started to recover after drenching, these had not returned to control levels by Day 45. The magnitude of the changes in activity, and standing and lying episode frequency and duration suggest that these might have a diagnostic value, especially as all can now be monitored by automated means. However, these behaviours did not show the rapid changes we expected before parasitism manifested clinically and following recovery.

1. Introduction

Animal behaviour may be one of the first things that change when an animal is affected by a health or welfare challenge, and can precede any clinical signs of stress or disease (Kyriazakis and Tolkamp, 2010). A number of recent papers have demonstrated that several aspects of cattle behaviour may be modified by (mainly) bacterial challenges (Quimby et al., 2001; Borderas et al., 2008; González et al., 2008; Fogsgaard et al., 2012) and that these changes may have diagnostic value. González et al. (2008), for example, have shown that measurable changes in the feeding behaviour of dairy cattle occur in cases of lameness and ketosis several days before anything can be detected by farm personnel, which allows for early action to be taken. The rate of change in such behaviours is dependent on the nature of the health challenge (González et al., 2008).

The preliminary and short term study of Szyszka et al. (in press) is the only one that has investigated several behavioural changes that may occur in cattle parasitized by helminths. It was found that several aspects of cattle

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posture and activity, such as lying and standing behaviour, and the number of steps taken, were affected to a variable extent by subclinical parasitism with *Ostertagia ostertagi*. In the present study we were interested in the temporal aspects and the magnitude of similar changes in behaviour that might take place during such infections. In addition to measurements of activity and posture, we also measured feeding behaviour because (i) a reduction in food intake is a consistent feature of such infections (Fox et al., 1989; Kyriazakis et al., 1998) and (ii) changes in the grazing behaviour of dairy cattle as a consequence of parasitism have been reported by Forbes et al. (2007). We were interested in whether such changes in behaviours arise before conventional signs of parasitism or clinical symptoms occur, and also in how quickly these changes are reversed when animals are drenched with an anthelmintic. Some abomasal damage by the parasites can occur before any eggs appear in the faeces, concurring with the development of larvae (Murray et al., 1970); this could manifest as a change in animal behaviours, before conventional detection of parasitism. In addition, Kyriazakis et al. (1996) have shown that the food intake of sheep parasitized with *Trichrostonglylus coubriformis* recovered within 5 days post drenching with an anthelmintic. Thus knowledge of such temporal changes may have diagnostic value for the assessment of the effectiveness of a treatment. However, such behavioural changes must be of sufficient magnitude if they are to have diagnostic validity (González et al., 2008). The hypotheses tested here, therefore, were: (1) changes of significant magnitude in aspects of behaviour due to parasitism will appear before any clinical signs, or the presence of eggs in the faeces and (2) such changes will be reversed very rapidly after dosing with an anthelmintic.

2. Materials and methods

The experiment took place at the facilities of Newcastle University after approval of the experimental protocols by the Animal Experiments Committee and under license according to the UK Animals (Scientific Procedures) Act for experimental challenge and regulated procedures.

2.1. Animals and housing

The animals used were 26 Holstein-Friesian bulls, approximately 3 months of age, with an average weight of 117 ± 25 kg, derived from a commercial farm. All animals were housed together in a single straw-bedded pen measuring 10 m × 4 m. Both food and water were available on an *ad libitum* basis with the feeder providing 23 feeding spaces, in accordance to the space requirements set by the UK Animals (Scientific Procedures) Act (Wolfensohn and Lloyd, 2003). The food offered was a total mixed ration, consistent throughout the experiment, containing 31.25% barley, 18.45% sugar beet pellets, 15.98% soya bean meal, 13.42% barley cereal, 9.76% distillers maize, 7.41% molasses and 3.75% chopped barley straw. The chemical composition of the food was 13.01 MJ Metabolizable Energy and 197 g Crude Protein per kg dry matter as estimated from AFRC (1993) feed tables. The animals had not received any prior challenge with parasites and were treated with 2 ml of the anti-inflammatory dexamethasone (Rapidexon, Eurovet, Cambridge, UK) and 8 ml of the antibiotic florfenicol (Nuflor, Shering-Plough, Milton Keynes, UK) prior to being parasitized, to reduce the risk of potentially confounding bacterial infections; such a treatment was not expected to affect the development of parasitism (Greer et al., 2005).

2.2. Experimental design

The animals were randomly assigned, taking into account their initial body weight, to one of four treatment groups. The experiment was considered to start when the animals received the first challenge (designated as Day 0). Prior to the application of the health challenges (Day −8) animals were fitted with a pedometer (IceRobotics, South Queensferry, UK) secured with Velcro on their front left leg, and video recordings of feeding behaviour started in order to provide background data before the parasite challenge. Also on the same day (Day −8) the animals received 7.5 mg/kg body weight of the anthelmintic albendazole (Albenil, Virbac, Woolpit, UK).

The first treatment group (P) consisted of seven animals, which received a trickle dose of 300,000 L3 *O. ostertagi* larvae in total, administered by gavage in doses of 100,000 L3 on Days 0, 7 and 14 of the experiment. The dose and its expected consequences on feeding behaviour and activity had been established in a previous experiment (Szyszka et al., in press). Trickle dosages of *O. ostertagi* have been previously reported in literature to mimic a repeated challenge (Fox et al., 1989; Forbes et al., 2009), which is more likely to be encountered in the field. The animals in this group remained infected for the duration of the experiment (Day 45). The second treatment group (PA) also consisted of seven animals and received the same trickle infection as group P. However, on Day 31 of the experiment, these animals received 7.5 mg/kg of the broad-spectrum anthelmintic ablendazole (Albenil, Virbac, Woolpit, UK).

The third (C) and fourth (CA) treatment groups, which acted as unchallenged controls, each consisted of six animals. They were given by gavage 20 ml water on Days 0, 7 and 14. Group CA was also treated with the anthelmintic on Day 31 (Albenil, Virbac, Woolpit, UK) to coincide with the drenching of the second treatment group, thereby controlling for any potential behavioural side effects of the anthelmintic dosing on the animals. The L3 larvae used as the challenge were obtained from Ridgeway Research (Gloucestershire, UK) and were of an Ivermectin susceptible strain that was isolated in South Gloucestershire, UK, 3 months before use (reference label OOSG10). Upon arrival the 3.6 M larvae were split in six glass beakers and diluted in 500 ml of water each, which was changed every other day until use. Just prior to dosing, 410 ml of surplus water was removed from the top leaving six doses of 15 ml with 100,000 L3 per dose. Each dose was topped up with 5 ml of water and administered to each animal by gavage.

The experiment lasted for 45 days, throughout which faecal samples and body weight measurements were taken twice a week, and blood samples were taken once a week; the same measurements were taken on the last day of the experiment. All animal handling took place in the morning,