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# Investigation and treatment of ovine psoroptic otoacariasis

### Citation for published version:

Crilly, JP, Nunn, F, Busin, V, Marr, EJ, Jennings, AE, Burgess, STG & Sargison, ND 2016, 'Investigation and treatment of ovine psoroptic otoacariasis' Veterinary Dermatology, vol. 27, no. 3, pp. 206-e52. DOI: 10.1111/vde.12314

### **Digital Object Identifier (DOI):**

10.1111/vde.12314

### Link: Link to publication record in Edinburgh Research Explorer

**Document Version:** Early version, also known as pre-print

**Published In:** Veterinary Dermatology

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## 1 Abstract

2 **Background**- Psoroptic otoacariasis has been described world-wide and is caused

3 by a mite morphologically indistinguishable from the sheep scab mite *Psoroptes ovis*.

4 A single treatment of affected sheep with 200 μg/kg of injectable ivermectin is

- 5 reported to be curative.
- 6 **Case report** *Psoroptes* mites were isolated following treatment with ivermectin but

7 treatment with moxidectin at 1mg/kg caused complete cessation of clinical signs.

8 Affected animals were seropositive to Pso o2 antigen ELISA, and had serum

9 haptoglobin concentrations that overlapped with those described for field infections

10 of classical sheep scab.

11 **Conclusions and clinical importance**- Psoroptic otoacariasis is not controlled by

12 single treatments of injectable ivermectin but resolves after a single treatment with

13 injectable moxidectin. Pso o 2 ELISA can detect infection with *Psoroptes* spp. mites

14 but cannot distinguish between sheep scab and psoroptic otoacariasis.

### 15 Background

- 16 Psoroptic otoacariasis has been described in domestic sheep (Ovis aries) in the UK,
- 17 France, Germany, Brazil and Israel.<sup>1,2,3</sup> It is also a widespread problem among
- 18 Bighorn sheep (*Ovis canadensis*). <sup>4, 5</sup> Opinions differ whether the causative mite

19 should be classified as a strain of *Psoroptes ovis* (the sheep scab mite) or of

20 *Psoroptes cuniculi* (the rabbit ear mite) or whether these are homospecific .<sup>6</sup>

21 Clinical signs of psoroptic otoacariasis include brown crusts in the external auditory

meatus (EAM), aural pruritus and subsequent secondary changes. <sup>1, 3</sup> The majority

of the mites are found close to the tympanic membrane.<sup>7</sup> Diagnosis is by isolation of

24 mites. There are no licensed products for the treatment of psoroptic otoacariasis.

Unlike sheep scab, it does not respond to plunge-dipping in organophosphates but is reported to be cured by single injections of ivermectin at 200 µg/kg compared with

reported to be cured by single injections of ivermectin at 200  $\mu$ g/kg compared wi sheep scab for which two injections at a seven-ten day interval are required <sup>1,8,9</sup>.

# 28 Case report

The owner of a small pedigree Wensleydale flock requested investigation of aural pruritus and hair loss affecting the majority of the flock in October 2013. Psoroptic

mange (sheep scab) had not been diagnosed previously in the flock or in any

neighbouring holdings; only two animals had been introduced during the previous 5

years. The owner reported two previous outbreaks of similar disease in 1992 and

34 2002. On both occasions injectable moxidectin treatment was reportedly successful.

The flock was first visited on 14th October 2013 and all of the sheep were examined.

36 This confirmed the presence of lesions of the pinnae and EAM consistent with

37 psoroptic otoacariasis. The yellow scabs typically associated with sheep scab were

not seen. Affected EAM were swabbed as previously described; a sterile, cotton-

tipped, bacteriology swab was gently inserted into the EAM, rotated and then

40 withdrawn, with any loose debris in the EAM also collected.<sup>1</sup> All sheep were blood

sampled using plain vacutainers. The swabs and any debris recovered were

42 examined microscopically. The serum samples were analysed for anti-Pso o 2

antibody by ELISA as previously described and for haptoglobin using a commercial 43 colorimetric assay (Tridelta PHASE Haptoglobin Assay; Tridelta Development Ltd, 44 Maynooth, Co.Kildare, Republic of Ireland).<sup>10</sup> Haptoglobin is an acute phase protein 45 and as such is a non-specific marker of inflammation. Levels are known to be 46 elevated in sheep with active sheep scab infection and drop rapidly after successful 47 treatment.<sup>11</sup> All sheep were treated with subcutaneous injection of ivermectin at 200 48 49 µg/kg (Panomec Injection for Cattle, Sheep & Pigs, Merial Animal Health Ltd, Harlow, Essex, UK). 50

Clinical signs reappeared and worsened within 1 month. The flock was visited again 51 52 and deep skin scrapings were taken from the hyperkeratotic areas of affected animals. One animal had died and three had been sold between the first and second 53 54 visits. Both ears of all sheep with visible lesions were flushed following a previously described method; in brief a length of flexible rubber tubing was gently inserted into 55 the EAM and 60 ml of sterile saline was instilled using a syringe, overflow from the 56 ear was captured and negative pressure applied to withdraw as much of the flushing 57 fluid as possible, which was then pooled with the overflow fluid.<sup>2</sup> Repeat blood 58 samples were collected from all animals with visible lesions. All sheep in the flock 59 were treated with 1 mg/kg moxidectin (Cydectin 20 mg/ml LA Solution for Injection 60 61 for Sheep, Zoetis UK Ltd, London, UK). Ears were scored at both visits. An arbitrary 0-3 score was given to each ear using the following criteria: 0- no visible lesion; 1 -62 alopecia and crusting at the ear base; 2 - more extensive alopecia and crusting and 63 the presence of dark brown scabs; 3 - fibrosis and thickening of the pinna, complete 64 occlusion of the external auditory meatus and the presence of several dark brown 65 scabs. Individuals were assigned a score by summing their ear scores. Sheep were 66 67 classified as lesions present/absent and as lamb (<1 year old) or adult (> 1 year old). Statistical analysis was performed using Minitab 16 (Minitab Inc., Sate College, PA, 68 USA). Proportions of affected sheep were compared by 2-proportions test, 69 70 haptoglobin concentration and ELISA titre variation between animals were analysed 71 by 2-sample t-test, and within animals over time by paired t-test; correlation between these variables was analysed by a Pearson test. Comparison to previously published 72 results was performed using a 1-sample t-test. 73 At the first visit 11/23 animals had visible lesions; at the second visit 14/19. A 74

- At the first visit 11/23 animals had visible lesions, at the second visit 14/19. A
   statistically significantly higher fraction of adult sheep than lambs had score 3 lesions.
- 76 on both dates (p=0.043 and p=0.012 respectively).
- No mites were isolated from the swabs or skin scrapes. 3 mites were isolated from 2
- animals by flushing. These were morphologically consistent with *Psoroptes ovis* tritonymphs.
- 80 Pso o 2 titres which exceeded twice the optical density of the negative control (0.21)
- sample were considered sero-positive. At the first sampling, 6/18 samples were
- sero-positive, whilst 7/12 were positive at the second sampling. Not all Pso o 2 or
- haptoglobin results could be assigned to specific individuals due to mismatches
   between sheep ear tag numbers and label numbers. There was no statistically
- between sheep ear tag numbers and label numbers. There was no statistically
   significant difference between the two sets of Pso o2 ELISA titres (p=0.209), or when
- significant difference between the two sets of Pso o2 ELISA titres (p=0
   the identified samples were compared (p=0.591).
- 87 The mean haptoglobin concentration was 0.47 mg/ml at first sampling and 0.24
- 88 mg/ml at second sampling, the haptoglobin concentrations were statistically

significantly different (p=0.019). There was no difference between the haptoglobin

concentrations at first sampling and the pre-infestation mean (0.30mg/ml) previously

described (p=0.072), though some results were higher than those described for field-

acquired sheep scab.<sup>11</sup> There was no correlation between Pso o2 titre and

haptoglobin concentration at either date (p=0.402, p=0.759 respectively). The

94 presence of ear lesions and the age of the animal did not appear to have a

95 significant effect on the likelihood of a positive Pso o 2 ELISA result or the serum

96 haptoglobin concentration.

97 After ivermectin treatment a temporary reduction of the degree of pruritus was

observed but clinical signs resumed within 1 month. After moxidectin treatment the

owner reported a cessation of signs in all sheep. To date (an 18 month period) none

100 have shown further signs of aural pruritus.

### 101 Discussion

102 The appearance and age distribution of clinical signs were similar to those described 103 previously .<sup>1</sup> The lack of lesions or history of sheep scab suggests the mites isolated 104 are a fully ear-adapted strain.

Swabbing failed to detect any mites, flushing isolated mites from two animals. This is 105 similar to a previous report of psoroptic otoacariasis in Brazilian sheep .<sup>2</sup> Only sheep 106 with visible lesions were swabbed or flushed, as this investigation began as an 107 investigation of the cause of the clinical signs. This is likely to have missed infected 108 sheep as ear mites have been isolated from sheep showing no clinical signs of 109 otoacariasis.<sup>1</sup> Animals with lesions may be those which are hypersensitive and thus 110 responding strongly to a relatively small number of mites. Mites were recovered from 111 10.5% (2/19) animals, which is lower than previously reported (46%-83%).<sup>2</sup> This, and 112 the fact that only immature stages were isolated, is consistent with ivermectin 113 treatment eradicating the adult mites which were then replaced by newly hatched 114 115 mites.

Previously, cessation of pruritus and elimination of living mites from the ear canal 116 following a single injection of ivermectin (200 µg/kg bw), has been reported for ovine 117 psoroptic otoacariasis.<sup>1</sup> This is in contrast to the situation with sheep scab. 118 Injectable moxidectin, both in the 1% and 2% formulation, is widely used for the 119 treatment and control of sheep scab.<sup>12, 13, 14</sup> Similarly, a sustained release ivermectin 120 implant has been successfully used to treat psoroptic otoacariasis in Bighorn sheep. 121 <sup>15</sup> The period of persistent activity of 1% moxidectin against infection by *P.ovis* is 35 122 days; this is longer than the incubation period of *P. ovis* eggs and the maximum 123 survival period of the mite away from the host. 12,13,16,17, 18 124

Several animals at each sampling were seropositive for anti-Pso o 2 antibodies in a flock with no clinical history of sheep scab. These results suggest that positive Pso o2 ELISA results indicate either sheep scab or psoroptic otoacariasis, as has previously been described for crude *Psoroptes* antigen ELISAs in both domestic and Bighorn sheep.<sup>19, 20, 21</sup> Anti-*P. ovis* antibody ELISAs have been proposed for the detection of asymptomatic sheep scab infections; these results suggest that such a test cannot distinguish between these two *Psoroptes* spp. infections.<sup>10,22</sup>

Haptoglobin concentrations in sheep artificially infected with sheep scab increased to
 above 3 mg/ml, well above that of any sheep in this investigation.<sup>11</sup> However the

- range of concentrations seen at the first sampling did overlap that described for
- naturally acquired sheep scab infection, as would be expected from an active
- 136 infection.<sup>11</sup>

137 There was no difference in the proportion of animals with positive Pso o 2 ELISA

titres at the first and second sampling. This is unsurprising given the first treatment

- 139 was unsuccessful in eliminating infection and that circulating anti-*Psoroptes* antibody
- levels drop over a period of weeks to months after successful treatment in cases of
   sheep scab and Bighorn otoacariasis. <sup>20, 22, 23</sup> Haptoglobin levels fell between first
- and second sampling; this would be consistent with ivermectin killing the adult mites
- and so temporarily reducing the antigenic stimulation.

### 144 **Conclusions and clinical importance**

145 Psoroptic otoacariasis is an uncommon parasitic infection of sheep causing clinical

signs related to aural pruritus. Mites may be most successfully isolated ante-mortem

by flushing of the external auditory meatus. Treatment with ivermectin was

unsuccessful in this case; moxidectin treatment resulted in the resolution of clinical

signs. Ear mite infection resulted in positive titres using the recombinant Pso o2

150 ELISA test.

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- 209 Figure captions
- Figure 1.

This figure shows the range of lesions associated with psoroptic otoacariasis in this case. Top: A score 1 lesion- alopecia and crusting at the ear base. Middle: A score 2 lesion with more extensive alopecia and crusting and the presence of dark brown scabs. Bottom: A score 3 lesion with fibrosis and thickening of the pinna, complete occlusion of the external auditory meatus and the presence of several dark brown scabs. Ears without any visible lesions received a score of 0.

- 217 Table 1.
- 218 This table shows the total ear lesion score, age of affected sheep and Pso o2 ELISA
- titres and haptoglobin concentrations where these could be ascribed to individual
- animals. The negative control OD was 0.21, the positive control OD was 2.0. Pso o2
- values which were considered to be positive are in **bold**. Sheep which were absent
- from the flock in December (dead or sold) are denoted by N/A in the applicable cells.