

Field trial of the efficacy of praziquantel for the treatment of moniezirosis in naturally infected sheep

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Summary

The purpose of the study was to evaluate the efficacy of praziquantel at 3.75 mg/kg b.w. and 5 mg/kg b.w. in treating *Moniezia expansa* and to observe the appearance of the parasite in the faeces of sheep following the treatment. Thirty-six sheep (24 male + 12 female), naturally infected with *Moniezia expansa*, were allocated to three groups according to the following dosage regimes: Group 1-3.75 mg/kg b.w., n = 12; Group 2-5 mg/kg b.w., n = 12; Control group, n = 12. The sheep were around 6-7 months old and weighed between 17.7 – 35 kg. Sheep were randomly divided into equal groups based on mean weight and sex. Faeces were collected after 12, 36, 60, 84, 108, 132 and 156 hours and just before slaughter (final 24 hours faeces) into faecal collection bags in the treatment groups. The collected faeces were then examined macroscopically for any parasite segments and microscopically for parasite eggs. After treatment the sheep excreted parasite segments as either normal parasite forms or deformed forms (melted and capsule or rosary forms). Equal numbers of animals (3 from each group) were slaughtered 10, 11, 12 and 13 days following treatment and their intestinal contents examined for the presence of parasites. None of the treated animals either in group 1 or group 2 had strobilae or scolices of *M. expansa* in their intestine contents after the slaughter. In contrast, sheep in the control group had 0.5-61 ml strobilae and 1-8 scolices belonging to *M. expansa*, *Thysaniezia giardi* and *Stilesia globipunctata*. It was concluded that praziquantel at the dose rates used in the study was 100 % effective against *Moniezia expansa*.

Keywords: praziquantel, *Moniezia expansa*, anoplocephalid tapeworms, sheep

Praziquantel is a pyrazinoisoquinoline derivative with well-known activity anticestodal and antitrepatodal in humans and animals. It was discovered by Bayer AG and Merck in 1972 (1). It is effective at different dose levels according to the target parasite species. In veterinary medicine praziquantel is used at 5 mg/kg in dogs and cats for the elimination of tapeworms by the oral route. Tapeworms in sheep can be very harmful and may cause economic losses (5, 7, 15, 16). The dosage 3.75 mg/kg was found effective against *Moniezia spp.* and *Thysaniezia spp.* while much higher dosage (8-15 mg/kg) is necessary against *Avitellina centripunctata* and *Stilesia globipunctata* (1-3, 5, 12-15). Anoplocephalid cestodes known from Turkey are to infect sheep *Moniezia spp.*, *Thysaniezia spp.*, *Stilesia spp.* and *Avitellina spp.* (4, 16, 18).

The mode of action of praziquantel makes the drug different, such as shrinking phenomena were observed in both trematodes and cestodes to praziquantel: Tetanic contraction of the parasite musculature and rapid vacuolisation of the syncytial tegument (1). This mode of action might cause different excretions form of tapeworms. Special information about the excretion form of tapeworms after praziquantel treatment is not found in literature.

Mansonil-B® is a tablet formulation of praziquantel (containing 75 mg active ingredient) and it was used for the treatment of sheep. Animal owners reported that no

parasite expelled after drug application and thus the assumption was made that their animals died after the treatment and they had parasites in their intestines. This study was conducted to evaluate the efficacy of praziquantel using two different dosages on *M. expansa* and to observe the appearance of tapeworms in the faeces after treatment.

Material and method

For this study, the faeces of 150 Kivircik sheep, a very popular sheep breed specific to Turkey in the Bolu province (North-West Anatolia/Turkey) were examined for the presence of *Moniezia expansa* microscopically by a flotation and McMaster egg counting method (10). None of the sheep had been previously treated with anthelmintics. Forty-three sheep were positive for *Moniezia expansa* based on the findings of eggs in the faeces. Thirty-seven animals were selected and housed indoors. One of the 37 infested sheep was slaughtered in order to assure that the animals were infected with tapeworms. This sheep had a single *M. expansa* with an approx. volume 25 ml in the small intestine. The weight of the test animals was between 17.7-35 kg (group 1 mean: 26.34 ± 1.09 kg; group 2 mean: 26.61 ± 0.91 kg and control group mean: 26.29 ± 3.46 kg) and there were 12 female and 24 male sheep. Sheep were allocated into three groups according to the dosage regime as: Group 1 – 3.75 mg praziquantel/kg b.w., n = 12; Group 2 – 5 mg praziquantel/kg b.w., n = 12; Control group, n = 12. All animals were

ear tagged, modified McMaster faecal collection bags were used in the study for collecting of the faeces (fig. 1).

Tablets were weighed in accordance to the animals weight with the use of a sensitive electronic balance. In order to ensure that the tablets were swallowed plenty of water was given. Faecal collection bags were put on the animals for the collection of the faeces over the study time. The contents of the bags were collected in the first 12 hours and then in every 24 hours. Faeces of all animals were checked for tapeworm segments during the study time by naked eye.

Microscopic faecal examination by light microscope after faecal flotation (in saturated sodium chloride solution (SG 1.204)) from every faecal sample before and during the study. The count of tapeworm eggs was performed before the start of the study, right after the treatment (for up to 108 hours) and just before the slaughter according to the modified McMaster egg counting method (10).

Three sheep from each group (one female, two male) were slaughtered at days 10, 11, 12 and 13 post treatment. Counts on tapeworms and tapeworm segments were performed according to the recommendations of literature (17). The volume of tapeworms in the intestine was measured by using a cylindrical measure.

In order to observe whether *M. expansa* and *T. giardi* tapeworms recovered from the intestine of the controls, they were treated with a fresh praziquantel suspension, in a dilution rate of 0.009375% (1/2 tablet Mansonil-B to 400 ml water added and mixed). In order to observe the activity of parasites, they were examined in a black water bath with a black bottom. The length of alive parasites were measured before and 2 minutes after exposure.

Tapeworm identification was performed according to literature (7, 8, 11) following staining with Mayer's acid carmin using staining colouring (9).

The statistical analysis of this study was done by using the Mann-Whitney U test of the SPSS 11 package (SPSS, Chicago, IL).

Results and discussion

Macroscopic faecal examination. The type of tapeworms appeared in the dropping after treatment were different in comparison to the untreated controls. After the treatment, sheep excreted parasite segments different to normal forms, where segments of tapeworms are clearly seen. Deformed segments, like melted, capsule, membrane and rosary form (Fig. 2) were present. Most of the sheep in the treatment groups excreted deformed and melted strobilia, which does not clearly seem as a parasite segment. Therefore the volume of the excreted tapeworms could not be measured in the treatment groups. One of the sheep of control group, excreted *Avitellina centripunctata* (about 20 ml) at 8.5 days collection time.

Microscopic faecal examination. All sheep were positive for *M. expansa* eggs before the start of the study. Two sheep in group 1 and group 2 were also positive for *Thysanietzia spp.* eggs. Two sheep in group 2 and two sheep in the control group harboured *T. giardi* eggs after the study start. An



Fig. 1. Modified faecal collection bags



Fig. 2. Group 1 (number 512) 156 hours after the treatment. Deformed form (Capsule type)

increase in *M. expansa* egg released in the 12 hours after the treatment was significant between the 1st group and the control ($p < 0,001$) according to the Mann-Whitney U test, while this result was significant in 36 hours after treatment between the control and groups 1 and 2 ($p < 0,05$). In other faeces collection time, there were no significant difference ($p > 0,05$) among the three groups.

Macroscopic necropsy findings. Only one tapeworm with scolex (*Stilesia globipunctata*, 8 ml) was found in one lamb intestine of group 2. There were different volumes of tapeworms from 0.5 ml to 61 ml in the intestines of the control group. The parasites found in the small intestinal contents of the control group were *M. expansa*, *T. giardi* and *S. globipunctata* (tab. 1).

Tab. 1. Necropsy findings in all groups

Study group	No of scolices excreted	Volume of tapeworms (ml) post mortem	No of scolices at post mortem
Group 1			
11 sheep	Neg.	Neg.	Neg.
1 sheep (no 542)	Neg.	Neg.	Pos. (1 × S)
Group 2			
11 sheep	Neg.	Neg.	Neg.
1 sheep (no 543)	Neg.	Pos. (8)	Pos. (1 × S)
Control			
No 508	Neg.	Pos. (6)	Pos. (1 × M, 1 × T)
No 510	Neg.	Pos. (0.5)	Pos. (1, M)
No 511	Neg.	Pos. (61)	Pos. (4 × M, 3 × S, 1 × T)
No 514	Neg.	Pos. (2.5)	Pos. (1 × M, 1 × T)
No 503	Neg.	Pos. (1)	Pos. (1 × M)
No 519	Neg.	Pos. (1)	Pos. (1 × M)
No 523	Neg.	Pos. (1)	Pos. (1 × M)
No 545	Neg.	Pos. (4)	Pos. (1 × M, 2 × S)
No 525	Neg.	Pos. (0.5)	Pos. (1 × M)
No 552	Neg.	Pos. (25)	Pos. (3 × M, 1 × T)
No 522	Pos. (2,A)	Pos. (5)	Pos. (1 × M, 1 × S)
No 548	Neg.	Pos. (7)	Pos. (3 × M, 2 × S, 1 × T)

Explanations: Neg. – negative, Pos. – positive, T – *Thysanietzia giardi*, A – *Avitellina centripunctata*, M – *Moniezia expansa*, S – *Stilesia globipunctata*

Microscopic necropsy findings. No scolices for *M. expansa* or *T. giardi* were found in the content of intestines of treatment group 1 and 2. One scolex of *S. globipunctata* was recovered from two sheep of group 1 and 2. Different tapeworm scolices (*M. expansa*, *T. giardi* and *S. globipunctata*) were observed in the small intestinal contents of control group (tab. 1).

Tapeworm movement in the praziquantel suspension. Tapeworms recovered from the intestine of one control lamb contracted short after the exposure to the praziquantel suspension and shrunk quickly within 15 sec. Length of *M. expansa* and *T. giardi* were 37 cm; 180 cm and 18 cm; 135 cm after the treatment, respectively.

Statistic analysis of the results showed that both the applied doses of 3.75 mg/kg l.w. and 5 mg/kg l.w. were 100 per cent effective against *M. expansa* (tab. 2). The efficacy tests were applied only for the *M. expansa*, due to low number of infested sheep with *T. giardi* and *Stilesia globipunctata*.

No scolices and strobilae of *M. expansa* and *T. giardi* were recovered from the intestine in groups treated with praziquantel at day 10-13 after the treatment. This result indicates that praziquantel give at 3.75 mg/kg b.w. and 5 mg/kg b.w. as 100% effective against *M. expansa*. The number of sheep infested with *T. giardi* in the treatment groups might be interpreted as not enough for the efficacy trial, however no scolices and strobilae of *T. giardi* were recovered from the sheep belonging to treatment groups at necropsy as well. Efficacy of 3.75 mg praziquantel per kg b.w. against *M. expansa* was also previously reported (2, 3, 5, 6, 12, 15).

Scolex of *S. globipunctata* was recovered from single group 1 and 2 sheep at necropsy. Strobila of *S. globipunctata* was also found in the intestine of one animal. The efficacy of praziquantel against *Stilesia spp.* and *Avitellina spp.* in a dosage of 8-15 mg/kg b.w. has been published previously (1, 13, 14).

Contraction of tapeworm's strobila after exposure to praziquantel was observed during this study, and this may be a reason for destruction of tapeworms. The parasite becomes smaller than the normal size. While tapeworms are being excreted by intestinal peristaltic movement and especially in large intestine, the normal structure of tapeworms may be destroyed easily, which may result in different appearance of tapeworms in the faeces. This process may be explained also by increased number of tapeworm eggs in the faeces after treatment, which was observed in this study. This may explain the different forms of tapeworms observed after treatment. Similar phenomena such as tetanic contraction of parasite musculature and rapid vacuolisation of syncytial tegument of cestodes and trematodes exposed to praziquantel were expressed in a review (1). A secondary effect such as depolarisation of *Schistosoma* tegument and inhibition of glucose uptake, and decrease of glycogen content of *Hymenolepis diminuta*, *Schistosoma mansoni* and *Schistosoma japonicum* was also reported (1).

Tab. 2. Efficacy of praziquantel (3.75 mg/kg b.w.; 5 mg/kg b.w.) against *Moniezia expansa* in natural infected sheep

Group	Dose rate (mg/kg LW)	Number of infected lambs		EE (%)	Sum. of Scolices in the gut	IE (%)	
		Day before the treatment	After slaughter			Arithmetic	Geometric
Control	-	12	12	-	19	-	-
1	3.75	12	0	100	0	100	100
2	5.00	12	0	100	0	100	100

Explanations: EE – Extense efficacy, IE – Intense efficacy

Conclusion

This study has shown that dosage of praziquantel according to the label instructions are 100% effective against *M. expansa*. However, the dosages 3.75 mg/kg b.w. and 5 mg/kg b.w. were not high enough to eliminate infections with *S. globipunctata*.

Tapeworms can not be easily seen in the faeces of sheep after praziquantel treatment, this may be due to the mode of action of praziquantel and peristaltic movements of the intestine and mechanic destruction or absorption of death tapeworms in the intestine.

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