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ICASE STUDENTSHIP ON TRANSMISSION AND CONTROL OF ECHINOCOCCOSIS IN THE FALKLAND ISLANDS – FINAL REPORT AND RECOMMENDATIONS

Report From	<p>Prof Mike Rogan Professor of Zoology, University of Salford, UK</p> <p>Dr Alex Mastin Research Fellow in Ecological/Epidemiological Simulation Modelling, University of Salford, UK.</p> <p>Dr Dominic West, Postgraduate researcher, University of Salford, UK.</p> <p>Dr Steve Pointing, Senior Veterinary Officer, Department of Agriculture, FIG, Falkland Islands</p> <p>Dr Haseeb Randhawa, Fisheries Scientist - Biology FIG, Falkland Islands</p>
Action Required	For information
Previous Consideration of the Report	None
Context/Purpose of the Report	To inform and advise the Falkland Islands Veterinary Service as to future direction of the Echinococcosis control and eradication programme
Executive Summary and Analysis	<p>This study has indicated that <i>Echinococcus granulosus</i> is still present in a small number of sheep and dogs in the Falkland Islands. However, levels of <i>Taenia hydatigena</i> in sheep from some farms have been very high over the past 6 years indicating a) that some dogs are accessing infected material from sheep and b) are not being effectively treated with Drontal. Observation of farm practice has indicated that in some cases inadequate disposal of offal and sheep carcasses was a significant risk for dog infection. Recommendations for the enhancement of the hydatid control programme include enhanced monitoring of farm slaughtered animals and the establishment/maintenance of a database, within the</p>

Department of Agriculture, of the presence of all taeniid infections in sheep slaughtered at the Sand Bay abattoir and at individual farms. Strategies for worming dogs could involve less frequent but supervised administration of Drontal and a more selective monitoring of dog infection levels. With these modifications and the assumption that no further dogs would develop mature worm infections, the hydatid control programme can move into a “consolidation phase” with an objective to bring the prevalence of the parasite in sheep and dogs close to zero within the next five to seven years.

Background

The Falkland Islands Government has been trying to eradicate Echinococcosis (hydatid disease) from the Falkland Islands since the mid 1970’s. Over the past 40 years significant progress has been made in reducing the incidence of hydatid disease in the sheep population in the islands and there have been no reported cases of human infection since the late 1970’s. To that extent the disease can be said to be under very good control. However, it has not been totally eradicated despite 40 years of attempting to do so. In 2017 the Department of Agriculture, Falkland Islands Government agreed to sponsor an iCASE studentship (commencing in January 2018) with the University of Salford, UK. This PhD study was set up in order to try and evaluate what factors may be involved in allowing the parasite to persist at low levels. If specific and meaningful results came out of the PhD study then these results would be used to inform the Falkland Islands Veterinary Service as to where the control and eradication programme was failing so that adjustments could be made with a view to completing the eradication programme within a defined period of time.

Project Summary

The project was carried out by Dominic West under a joint supervision team from both the University of Salford and Department of Agriculture, FIG. It was focused not only on the causative agent of Hydatid Disease, *Echinococcus granulosus*, but also on the presence of two closely related taeniid parasites, *Taenia hydatigena* (bladder worm) and *T. ovis* (sheep measles) which share a similar dog-sheep life cycle to *E. granulosus*. It involved the student undertaking two, three month fieldwork visits to the Falkland Islands in 2018 and 2019 with a third visit in 2020 being cancelled due to the Covid-19 pandemic. These were supported by laboratory work at the University of Salford.

The principal objectives of the study were:

- i. To establish current farming practice which may influence transmission of taeniid cestodes.
- ii. To establish the level of *E. granulosus* in dogs by coprological analyses and to identify other possible definitive hosts.
- iii. To evaluate differences in prevalence of *E. granulosus* and other taeniid cestodes in sheep at slaughter in the abattoir over time.
- iv. To evaluate approaches for detecting parasite eggs and biomolecules in soil samples.

- v. To apply mathematical models to explain the geographical distribution of taeniid infections in the Falklands and to simulate control and transmission of these infections.

Key Findings

Occurrence of *E.granulosus* in sheep

The number of hydatid infected sheep going through the Sand Bay abattoir is extremely low with only 31 infected animals being identified in the period 2006-2020. However, the numbers of infected animals which were slaughtered on farms rather than at the abattoir is not known. In 2020, five infected animals were identified through abattoir surveillance from a total of 40,529 slaughtered (0.01% prevalence). These were from five separate farms, four on East Falkland and associated islands and one on West Falkland. From 2006 to 2020 there was no obvious pattern to the distribution of farms having infected animals. One possible explanation for this is that grazing pasture contaminated with *Echinococcus* eggs at the time of infection was more widespread rather than being localised in hot spots. This supports previous observations that eggs can be transferred over considerable distances. However, one farm which had three coproPCR positive dogs in 2010, had cases of hydatid cysts detected in adult sheep at slaughter in 2014, 2015, 2017 and 2018.

Occurrence of other taeniid parasites in sheep.

The prevalence of *T. ovis* (maximum annual prevalence of 0.4% in 2007) and *T. hydatigena* in sheep was considerably higher than that of *Echinococcus*. The maximum prevalence of *T. hydatigena* was 6.0% in 2019 (7.29% in adult sheep). Both parasites are transmitted from dogs and have a much larger egg output (biotic potential) than *E. granulosus*. The number of farms with *T. hydatigena* infections was generally large, again indicating widespread contamination of grazing pasture rather than isolated hotspots.

Annual prevalences of the taeniid parasites varied considerably with peaks and troughs occurring frequently. This suggests that livestock grazing patterns result in high levels of infection some years and lower in others. Although there is no direct evidence, one possible explanation for this is the presence of a small number of infected dogs at different time points. With *T. hydatigena*, significant peaks in prevalence were seen in 2007, 2013, 2016 and 2019. With *T. ovis* similar peaks were seen in 2007, 2013 and 2016. The prevalences of all taeniid cestodes was considerably lower between 2008 and 2012. Analysis of slaughter data comparing new season lambs, yearling lambs and adult sheep, on a farm-by-farm basis frequently showed discrete peaks of infection which were asynchronous i.e. new season lambs first, then yearlings, then adult sheep. This supports the suggestion that infected dogs are appearing sporadically resulting in the infection of lambs which go through the abattoir several years later with detectable cysts. The occurrence of infected dogs over time must indicate that in some cases the dosing with praziquantel (Drontal) is not effective and adult, egg producing, worms are developing in the intestine.

Presence of *E. granulosus* in dogs.

In total, 589 dogs were screened in 2018, using a Copro-ELISA to detect parasite antigen and a Copro-PCR to detect parasite DNA. Both of these tests are specific for *Echinococcus* spp. Four dogs tested positive by Copro-ELISA but none of these could be confirmed by PCR. A dog may test positive by Copro- antigen and not by PCR if the worm/egg count is low or if the infection is immature. Since they were not positive on both tests the dogs were defined as "suspect" rather than confirmed infections. In relation to the history of these suspect dogs, two came from farms which had recorded

cases of hydatid cysts in sheep in 2020 and three had prevalences of *T. hydatigena* in abattoir-slaughtered sheep greater than 3% in 2020 with two farms having more than 6% infection. One of the farms also had detectable levels of copro-antigen present in the soil in the immediate vicinity of the dog kennels. These results suggest the high likelihood of dogs infected with taeniid cestodes being present in a small number of farms.

Possible alternative definitive hosts for *E. granulosus*.

In addition to dogs, a small number of faecal samples (22) from Patagonian Foxes from Weddell Island were analysed by Copro-antigen and Copro-PCR techniques. None of the samples tested positive. In addition, the intestines from 12 feral cats from East Falkland were examined, *post mortem*, for the presence of parasites. No *E. granulosus* or *T. hydatigena* was found. Although the sample size was small there was no indication that these species were acting as alternative definitive hosts for *E. granulosus*. In addition, the pattern of taeniid infection in sheep suggests more sporadic contamination of the environment from a small number of hosts rather than continuous spread of eggs by many hosts. A significant number of bird species were observed feeding on sheep carcasses at cull sites but birds are physiologically not suited for development of taeniid worms.

Evaluation of farm practice in relation to hydatid control measures.

In total, 50 out of 81 farms were visited during the study. In addition, questionnaires on farming practice were issued to 51 farms which were registered as keeping dogs. A 45% response rate was obtained. In general, there was good adherence to measures laid out by the Falkland Islands Government for the control of hydatid disease in relation to dog housing and disposal of offal. However, there were some areas of significant concern.

- i) Some home slaughter occurs on virtually all farms to provide meat for animal and human consumption and on some farms this is the only location where animals are killed (*i.e.* none are sent to the abattoir). Detailed information on the number of animals killed and the infection status of these animals is very limited. It was estimated that over 20,000 sheep per year are killed on farms but there is no centralised reporting of the presence of hydatid cysts or other taeniid infections in these animals. Several farmers reported that they had never seen *T. hydatigena* (Bladder cysts) in their home slaughtered sheep yet those going through the abattoir had significant levels of infection.
- ii) On many farms old sheep which have no value in terms of wool or meat production are culled at local sites rather than being sent to the abattoir. Carcasses are left in open remote sites without inspection and establishment of disease status. These are open to scavenging by a range of carnivores and the current study provided direct photographic evidence that dogs were also feeding on these carcasses. Old sheep are more likely to have higher levels of taeniid infection and the practice of dumping animals at cull sites, therefore carries a significant risk of dogs accessing this material.
- iii) Practices for the disposal of offal from farm slaughtered sheep is variable. In most cases, plucks (liver, lungs and heart) are “treated” and 19 farms (82.6%) reported carrying out a procedure that would kill hydatid cysts and cysticerci, *i.e.*, long term storage, incineration or freezing before material was dumped on the shore or buried. However, four farms reported either dumping untreated offal directly on the shore or feeding it to pigs. The practice of dumping untreated offal in the immediate vicinity of farms, even if

it is in the sea, poses a significant risk. Even though many dogs are kept in kennels for long periods, some were observed wandering around farms freely during the day. Offal deposited at sea could be washed back on shore and be accessible to dogs. The survival time of taeniid larvae in sea water is unknown. In relation to *T. hydatigena*, cysticerci are frequently associated with the mesenteries around the intestines and these are routinely deposited untreated on shores close to farm buildings. This again is a risk to dogs accessing infected material.

- iv) In most cases kennels used for housing dogs were in well maintained condition. However, in some cases, some were in need of repair. This study showed direct photographic evidence that some dogs were free to roam at some distance from farm buildings at night and could access sheep carcasses at cull sites. Free roaming dogs is again a significant risk.

The overall findings of this study have shown that the prevalence of all three species of taeniid cestode in sheep was very low between 2008 and 2012, but that subsequently a number of peaks of higher prevalences have been observed with levels of *T. hydatigena* in 2019 reaching 7.29% in adult sheep (2659 animals). The likely explanation for this is the sporadic presence of a small number of infected dogs where the adult worms have been able to survive long enough to produce eggs and contaminate the environment. From the distribution of farms where high levels of infection are occurring, it is not possible to say whether infected dogs are present locally or whether eggs are transferred over large distances and remain viable for a long time. Both are likely. The most likely route of dog infection is by scavenging at cull sites or on offal deposited on shores. In addition, for these infected dogs to produce mature, egg laying worms, they must have not had effective dosing with praziquantel (Drontal) for at least 6-8 weeks.

Recommendations for the future of the hydatid control programme on the Falkland Islands

The current study has calculated an initial baseline R_0 value of 0.344 indicating that the parasite is heading towards extinction. However, hydatid cysts are still present in a small number of sheep and a small number of dogs are also suspected to have *E. granulosus* worm infections. Calculations of R_0 in different scenarios where control measures are inadequate show that R_0 could rise to 1.0 and above only when there is a high probability of both dogs accessing infected offal and ingestion of eggs by lambs, coupled with a complete failure in dosing dogs with praziquantel. In order to enhance the progress towards extinction the following recommendations have been made.

1. Surveillance and monitoring infection levels in sheep at Sand Bay abattoir.

Accurate and detailed record keeping of the infection status of animals passing through the Sand Bay abattoir is crucial to monitoring. In addition to hydatid cysts, it is important to record the numbers of animals infected with *T. hydatigena* bladder cysts as although this parasite has a similar lifecycle to *E. granulosus*, its higher biotic potential makes it a useful "sentinel" for potential *E. granulosus* infection. The practice of recording the age of infected animals (new season lambs, yearling lambs and adult sheep) should be maintained as infection levels in lambs relate to the recent presence of viable eggs. Following discussions with veterinarians and Meat Hygiene Inspectors, it has been mentioned that inspections are focussed on the offal still attached to the carcass; the stomach and intestines that are removed from the carcasses down a

different chute are inspected only if there is time. As *T. hydatigena* is often found in the omenta, mesenteries, and peritoneum of infected sheep, less focus on inspecting the stomach and intestines, could result in underestimating the prevalence of *T. hydatigena*. The inspection process should, therefore, be consistent. It is essential that all meat inspectors can accurately differentiate taeniid cestode larvae, especially in young animals and this should be emphasised when there is a change in staffing to maintain consistency. The data from monthly abattoir reports should be regularly maintained in a database so that infection levels can be monitored. It is particularly important to ensure that the outcome of suspect samples sent to the Department of Agriculture for identification is entered into the database. Once confirmed, it would be advisable to undertake an inspection of farms involved to establish that correct control practices are in operation.

2. Surveillance and monitoring infection levels in sheep slaughtered on farms.

Although many sheep go through the Sand Bay abattoir there are a lot of other sheep that are killed locally on each farm. At present there is no record of the infection status of these animals and this is essential for monitoring infection on a farm-by-farm basis, particularly on farms which send no sheep to the abattoir. In practice this may have some difficulty as it requires farm staff involved in slaughter to be able to identify different taeniid larvae but this could be enhanced by providing on-line/ video training by the meat inspectors based at the abattoir. Monthly slaughter record reports should be produced by each farm, highlighting the number and approximate age of the sheep killed and their intended usage (*e.g.* human consumption or dog food). These reports need to be sent to the Department of Agriculture and entered into the database where they can be viewed in parallel with data from the abattoir.

3. Surveillance of infection levels of *E. granulosus* in dogs.

Analysing the entire dog population by copro-analysis is a time consuming and labour-intensive task, as well as being a costly undertaking. Surveillance could be done in a more focused way by monitoring dogs on farms with a previous history of infected dogs or hydatid infections in sheep or high levels of *T. hydatigena* in sheep. Ideally the tests used should, initially, have a broad specificity which would pick up all taeniid infections such as *T. hydatigena* (*e.g.* taeniid specific copro-antigen test and PCR primers, or microscopic detection of taeniid eggs) followed by more specific identification of *E. granulosus* in positive testing samples via species specific PCR. The further development of molecular biology labs at SAERI in the Falkland Islands could provide an opportunity for local diagnostic testing. Targeted testing on a smaller number of farms could be carried out twice a year, ideally before and shortly after a dosing session. The University of Salford would still be available to advise, initially, on testing dogs, however it should be noted that, at present, a taeniid-specific coproantigen test is not currently available and further investment would be required to produce this. Likewise, the idea of coproantigen testing of soil samples shows some good promise but further work is still required to optimise reagents and sampling strategies.

4. Disposal of offal on farms

The deposition of sheep offal and intestines at sea or on shorelines represents a significant risk in relation to possible dog infection. This practice should be discontinued, and all waste material should be either burned or buried after long term storage. In terms of environmental impact,

waste material from the Sand Bay abattoir should also not be dumped at sea. Although this may have a significant economic impact, alternative approaches should be sought.

5. Culling of old sheep and deposition of carcasses at cull sites.

The practice of culling old sheep and the dumping of carcasses on farms carries a significant risk of infection of dogs which may scavenge on the carcasses. Ideally this practice should be discontinued and these animals should be sent to the abattoir. If it is essential to kill old sheep, then the offal and intestines should be removed and disposed off by burning or burial after the details of the infection status have been established and recorded. Alternatively, whole carcasses could be burned.

6. Dog maintenance.

The condition of dog kennels on each farm should be maintained so that dogs can not escape and are not allowed to roam freely in areas where offal or sheep carcasses could be present. Inspection of kennels could be carried out by Veterinary Service staff during supervised dog dosing visits.

7. Dosing of dogs with praziquantel (Drontal).

Even though dogs are currently dosed every 5 weeks there is still evidence that cestode parasites are being transmitted, as shown by the copro-antigen tests and the levels of *T. hydatigena* in sheep. Failures could arise by accidental or deliberate failure to administer tablets, regurgitation of tablets by dogs or incorrect dosage being given, thereby resulting in an increase in R_0 . Although the current practice maintains the awareness of the hydatid control programme within the farming community it may be more effective to reduce the frequency of compulsory dosing to every two or three months but that this be carried out by veterinary staff who would ensure the correct dose was given and that the tablets had been consumed. This could also be coordinated at times when sheep and dogs are more likely to be in contact (eg at shearing/lambing times). As the number of sheep with hydatid cysts continues to decrease, the probability of a dog accessing infected offal also decreases and, therefore, it is justifiable to increase the period between treatments so long as all dogs can be confirmed as being adequately treated. More frequent dosing could be done on a targeted approach by selecting individual farms which have a history of previously infected dogs, or the presence of hydatid cysts in sheep or evidence of high levels of *T. hydatigena* infection in sheep. The concept of resistance to praziquantel is also possible, but unlikely and this has not been investigated in the current study. Further studies could be carried out with dogs experimentally infected with *T. hydatigena* or *in vitro* studies using *T. hydatigena* cysticerci.

Overall, future surveillance and control of Echinococcosis should be more focused on farms which are more at "risk" of being involved in transmission and at times when dogs and sheep are more likely to be in closer contact (e.g. shearing or lambing). With the current low level of hydatid infection in sheep and the recommended increase in efforts to prevent dogs accessing infected offal and being wormed effectively, the control programme can move to a "consolidation phase" rather than an attack phase". For Echinococcosis to be completely eradicated it is necessary to know where most of the transmission is occurring and there are still some gaps here. The relatively high levels of *T. hydatigena* transmission is also of concern. However, if the recommendations in this report are

carried out effectively it is feasible that the parasite prevalence in sheep and dogs could be close to zero within the next five to seven years.