Measuring the impact of different treatment regimes on the evolution of anthelmintic resistance

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Introduction

Parasitic infections pose a great challenge to both human and animal health [1,2]. The control of parasitic worm infections in livestock largely relies on the use of anthelmintic drugs [3]. Ivermectin (IVM) is the most widely used macrocyclic lactone (MLs) to treat parasitic worms in livestock [4].

Despite being effective in controlling parasitic worms in livestock, there's growing resistance against IVM due its intensive use and anthelmintic resistance (AR) in general is a major challenge to successful parasite control. The prevalence of ML resistance In Scottish sheep has been reported to be about 35% [5].

Teladorsagia circumcincta is the most prevalent gastrointestinal nematode of sheep [6]. Regular IVM treatment of sheep infected by *T. circumcincta* is becoming unsustainable due to ever-increasing levels of resistance. Unlike in benzimidazoles, the genetic basis underlying MLs resistance is poorly understood. Understanding the evolution of genetic markers involved in AR is key to developing novel control approaches for *T. circumcincta* as well as improving surveillance and treatment of teladorsagiasis in sheep.

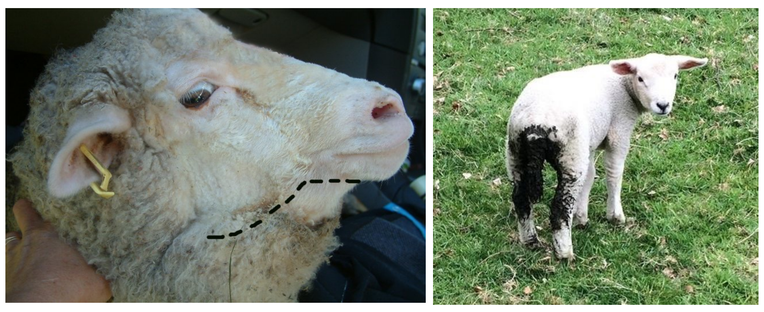


Figure 1. Signs of parasitic worm infections in sheep

Field Trial at Moredun

A 5-year field trial study at Moredun Research Institute [7] compared the impact of four different treatment regimes on the development of AR; It found that targeted treatment led to significantly slower development of resistance compared to the frequent dosing approach. This represents a uniquely well-controlled setting in which to investigate the evolution of AR.

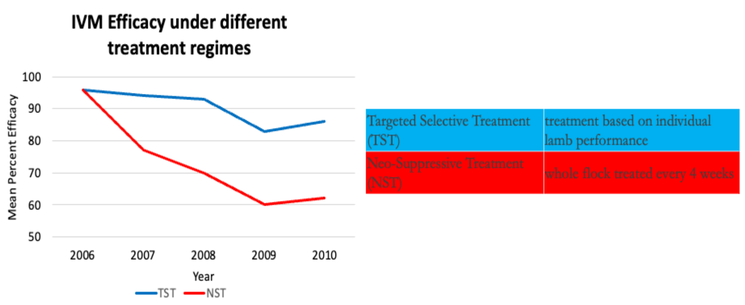


Figure 2. IVM drug efficacy data from the field trial [7].

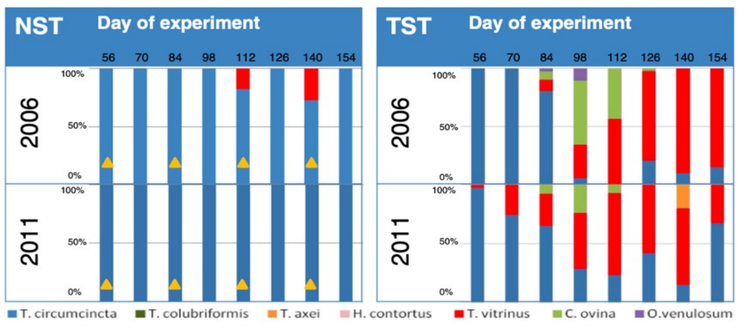


Figure 3. Species composition (%) of L1 larvae between 2006 & 2011 [8].

New Genome assembly

There is a new chromosome-scale genome assembly for the anthelmintic susceptible UK MTci2 strain of *T. circumcincta*.

The single isolate assembly was generated using PacBio long-read sequencing and HiC scaffolding together with extensive manual curation of the genome and annotation.

Genome assembly	Assembly size (Mb)	Scaffolds (n)	N50 (Mb)	N90 (Mb)	Largest (Mb)	Genes (n)	Complete BUSCOs (%) Genome	Complete BUSCOs (%) Protein
T. circumcincta Tci2_WSI3.0	573.0	1,286	84.0	2.4	94.8	22,948	85.2	96.3
T. circumcincta PRJNA72569	700.6	81,734	0.047	0.002	1.5	25,572	67.4	40.2
H. contortus PRJEB506	283.4	7	47.4	43.6	51.8	19,778	83.5	96.2
C. elegans PRJNA13758	100.3	7	17.5	13.8	20.9	18,178	98.8	100.0

This new assembly allows the use of whole-genome approaches and better-designed targeted genetic approaches to study AR in *T. circumcincta.*

Objectives of the study

Use some of the materials from the field trial to;

a) Optimise amplicon-based genotyping approaches for candidate AR loci in *T. circumcincta*.
b) Generate whole-genome sequence data from the pooled L1 larvae and targeted genotypes at candidate loci from individual worms from the field trial, and fit population genetic models to estimate the relative strength of selection acting on candidate ivermectin resistance loci.
c) Estimate the change in frequency of resistance alleles over a grazing season in Scottish sheep flocks and demonstrate the generality of results from the field trial data.
d) Conduct knowledge exchange to inform key stakeholders such as farmers veterinary practitioners and industries on the outcomes of the project.

Preparatory activities

- Checking viability of the samples; since its over 10 years since their collection from the field.

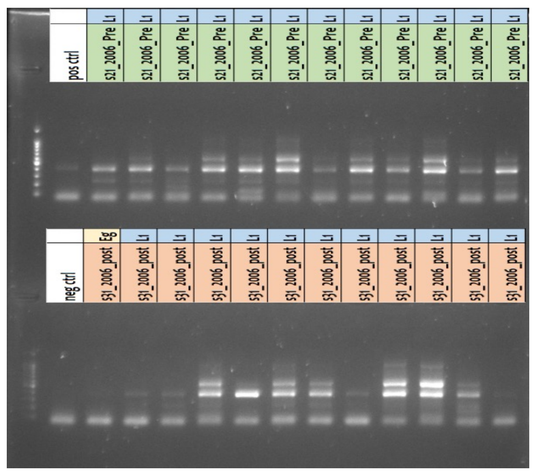
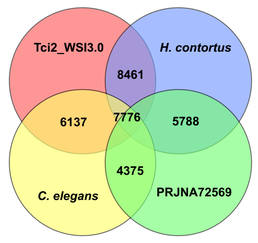


Figure 4. PCR Results of 2006 L1 larvae samples using Generic ITS2 & Tcirc ITS2 primers (double bands indicate *T. circumcincta* positive samples)

- Finding the orthologues of candidate AR genes from *Haemonchus contortus* and *Caenorhabditis elegans* in the new *T. circumcincta* chromosomal genome assembly.



A Venn diagram showing the total number of one-to-one orthologues in the predicted proteomes

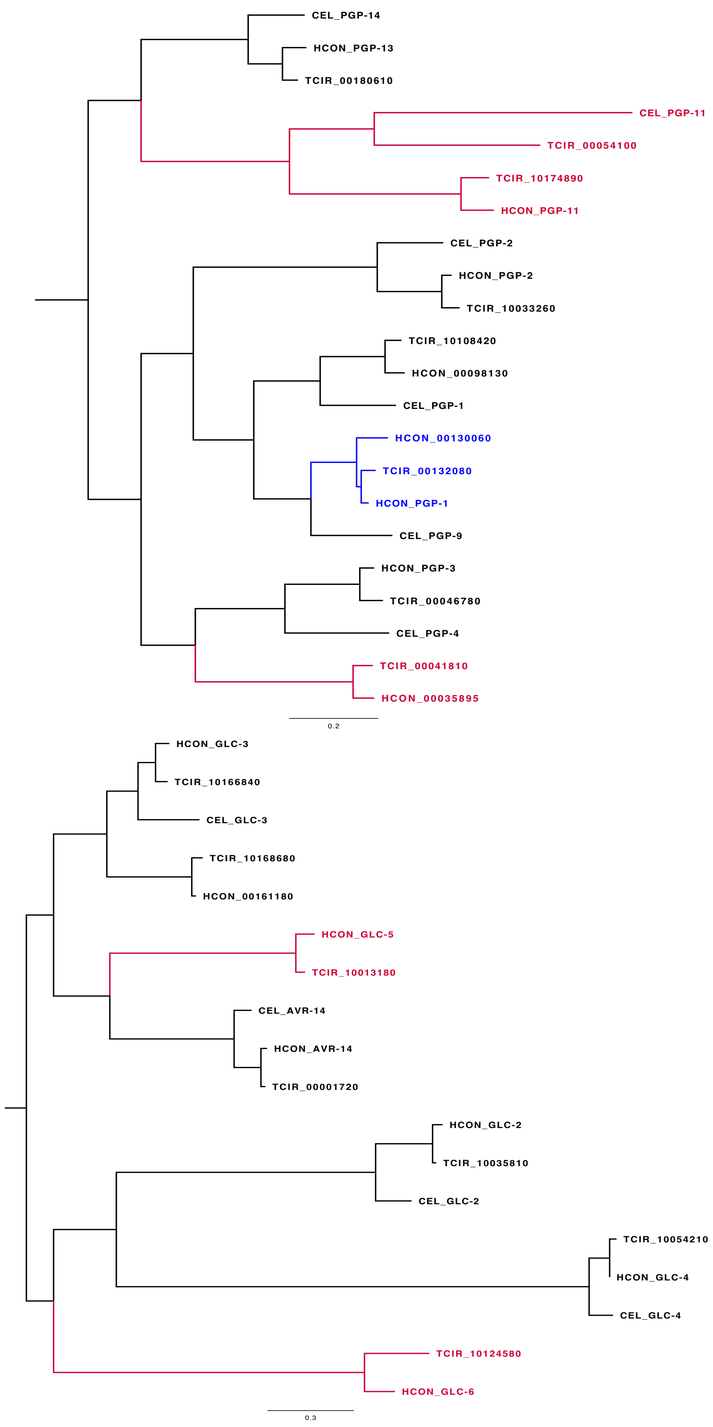


Figure 5. Phylogenetic trees showing orthologs of two families of *H. contortus* and *C. elegans* AR genes in the *T. circumcinta* genome P-glycoproteins and Glutamate-gated chloride channels

Key observations

a) Most of the L1 larvae samples from the field trial are still viable and can reliably be used in this study.
b) Conservation of AR candidate genes across *H. contortus, C. elegans* and *T. circumcincta* is not obvious.

Special Thanks

NORTHWEST BIO

British Society for Parasitology
2024 BSP Spring Meeting
Student Travel Award

Contact

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