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Drug resistance in nematodes of veterinary importance: a status report

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Reports of drug resistance have been made in every livestock host and to every anthelmintic class. In some regions of world, the extremely high prevalence of multi-drug resistance (MDR) in nematodes of sheep and goats threatens the viability of small-ruminant industries. Resistance in nematodes of horses and cattle has not yet reached the levels seen in small ruminants, but evidence suggests that the problems of resistance, including MDR worms, are also increasing in these hosts. There is an urgent need to develop both novel non-chemical approaches for parasite control and molecular assays capable of detecting resistant worms.

Many parasitic nematodes of veterinary importance have genetic features that favor the development of anthelmintic resistance. Among the most important of these are rapid rates of nucleotide sequence evolution and extremely large effective population sizes that give these worms an exceptionally high level of genetic diversity [1,2]. In addition, most nematode species that have been studied demonstrate a population structure consistent with high levels of gene flow, suggesting that host movement is an important determinant of nematode population genetic structure [2]. Thus, these worms possess not only the genetic potential to respond successfully to chemical attack, but also the means to assure dissemination of their resistant genes through host movement.

Brief history of anthelmintic resistance

The initial reports of anthelmintic resistance were to the drug phenothiazine in the late 1950s and early 1960s, first in *Haemonchus contortus* (barber pole worm) of sheep [3] and then in cyathostomins (small strongyles) of horses [4–6]. In 1961, thiabendazole was introduced as the first anthelmintic that combined efficacious broad-spectrum nematocide activity with low toxicity. The rapid acceptance and widespread use of thiabendazole and then other benzimidazole anthelmintics marked the beginning of the modern chemical assault on helminth parasites. However, within a few years, resistance to thiabendazole was reported, again first in the sheep nematode *H. contortus* [7,8] and then in the equine cyathostomins*. Reports then appeared of benzimidazole resistance in the other major gastrointestinal trichostrongylid nematodes of sheep – *Teladorsagia* (*Ostertagia*) *circumcincta* (brown stomach worm) and *Trichostrongylus colubriformis* (black scour worm). These reports led to studies investigating the prevalence of resistance, which found that, by the mid-1970s, multiple-species nematode resistance to benzimidazole anthelmintics was common and widespread in both sheep and horses throughout the world. This same pattern repeated itself in the 1970s and 1980s following the introduction of the newer imidazothiazole–tetrahydropyrimidine and avermectin–milbemycin classes of anthelmintics and, by the early 1980s, reports of multiple-drug resistant (MDR) worms appeared for the first time (reviewed by Refs [9–16])

By the 1990s, anthelmintic resistance was no longer a potential problem of the future. Widespread reports of MDR worms, including resistance to avermectin–milbemycin drugs, had elevated the issue of anthelmintic resistance from being one of academic interest to being a major threat to small-ruminant production in many areas of the world [17]. Presently, MDR (to all three major anthelmintic classes) *H. contortus*, *T. circumcincta* and *T. colubriformis* have been documented throughout the world, and MDR *H. contortus* now threaten the viability of small-ruminant industries in much of South America [18–21], South Africa [22], Malaysia [23,24] and southeast USA [25]. Recent reviews of the situation in Australia [26] and New Zealand [27] indicate that the problem of anthelmintic resistance, although severe, has not yet reached the crisis levels seen in some of the more tropical areas of the world. However, recent reports of moxidectin resistance in both Australia [28] and New Zealand [29] suggest that the problem may be more severe than past surveys have indicated. In other areas of the world, such as Europe and Canada, MDR worms have been only infrequently reported, and resistance is less of a concern. Nevertheless, in the UK, where drug resistance in nematodes of sheep is not nearly as severe a problem as it is in many other areas of the world, the problem is important enough that a national workshop was recently convened to develop a set of national strategies and recommendations to slow the development of resistance†; see Tables 1 and 2 for summaries of the resistance situation.

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† L. Stubbings (2003) Internal Parasite Control in Sheep. In *Proceeding of a Workshop to decide: short term strategies to slow the development of anthelmintic resistance in internal parasites of sheep in the United Kingdom*, held 11–12 March, 2003, in London, UK.

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Table 2. General worldwide situation in levels of anthelmintic resistance among livestock hosts

Drug class	Hosts with high resistance[a,b]	Hosts with emerging resistance[c]	Major livestock-producing areas where drug is still highly effective in sheep, goats and horses
Benzimidazoles	Sheep, goats, horses	Cattle	None
Imidothiazoles–tetrahydropyrimidines			
Levamisole (ruminants)	Sheep, goats	Cattle	None
Pyrantel (horses)	Horses (USA only)	Horses	Unknown – few recent studies outside USA
Avermectin–milbemycins			
Ivermectin	Sheep, goats, cattle	Cattle, horses[d]	Horses – worldwide Sheep, goats – Europe, Canada
Moxidectin	Goats	Sheep, goats, cattle, horses[d]	Horses – worldwide Sheep – most regions

[a]In all cases, references to resistance relate to cyathostomin nematodes of horses and/or trichostrongylid nematodes of ruminants unless otherwise specified.
[b]High resistance is defined as a level and prevalence of resistance that is sufficient to warrant general concern of using that drug on a particular property without prior testing for efficacy. It should be understood that many species of gastrointestinal nematodes infect ruminants and high resistance in any one species is sufficient for inclusion in this list. If high resistance is known to occur in only a single country and/or region, then it is specifically mentioned. If high resistance is known to occur in more than one region, then no reference is made, but this does not necessarily mean that there is high resistance everywhere.
[c]Emerging resistance is defined as a situation where anthelmintic resistance is reported to occur, but prevalence is not known and the level and distribution of resistance is not recognized as a severe problem.
[d]Only in *Parascaris equorum*; presently, there is no evidence of resistance in cyathostomin or *Strongylus* spp. nematodes.

originating from the UK demonstrates a much higher level of pathogenicity than ivermectin-susceptible isolates [47,48]. Explanations for why resistance develops more slowly in nematodes of cattle has been reviewed previously [49], but the fact that resistance is much slower to develop in nematodes of cattle gives strong evidence that many factors other than the genetics of the worms are involved in the dynamic process of resistance selection. Relevant factors that affect the rate with which resistance develops include: the biology and epidemiology of the parasite, the dynamics of the host–parasite relationship, the treatment frequency and the treatment strategies that result in various levels of refugia. An additional factor that has not been fully investigated is differences in anthelmintic pharmacokinetics between host species. Anthelmintic drugs demonstrate considerably lower bioavailability in goats than in other livestock species, and it is frequently suggested that the extremely high prevalence of anthelmintic resistance in nematodes of goats is associated with this unique pharmacokinetic profile.

What about resistance in parasites of humans? To date, there have been no documented cases of anthelmintic resistance in nematodes of humans, although there have been several reports where treatment with mebendazole or pyrantel demonstrated efficacies at much lower levels than expected against hookworms [50–52]. Differentiating reduced efficacy from true resistance is more complicated in nematodes of humans than it is in nematodes of animals owing to several factors that might confound interpretation of fecal egg count data (reviewed by Ref. [53]). Additionally, in the case of human parasites, it can be quite difficult to prove whether reduced efficacies are due to resistance or to some other factor because the confirmatory controlled efficacy experiments carried out with animals cannot be performed on human subjects. Furthermore, we currently lack the molecular knowledge required to develop diagnostic assays that can reliably identify resistance for all drugs except the benzimidazoles. Even with benzimidazole drugs where specific mutations have been correlated with a resistant phenotype in several nematode species [54], we do not have validated tests for use in human parasites. Though the issue of anthelmintic resistance in parasites of humans has received scant attention, the potential is real and this reality should be taken into consideration when implementing drug-based control strategies [53]. Current mass treatment programs for onchocerciasis and lymphatic filariasis may be placing strong selective pressures for resistance on these filarial worm populations, as well as on the important gastrointestinal nematode species. It is of crucial importance that studies be performed to monitor the development of resistance in these nematode species so that these large-scale programs for control can be adjusted if necessary to prevent program failure on the eve of what appears to be their success.

Implications of anthelmintic resistance

The serious problem of anthelmintic resistance is easily appreciated. But what can be done about it? Beginning with phenothiazine in the 1950s, followed by the benzimidazoles in the 1960s, the imidazothiazole–tetrahydropyrimidines in the 1970s and the avermectin–milbemycins in the 1980s; a new class of anthelmintics was introduced into the marketplace each decade. This arsenal of highly effective and relatively inexpensive drugs led to recommendations for parasite control that were based almost solely on the frequent use of anthelmintics, the goals of which were to maximize livestock health, productivity and profitability. Though this approach was highly successful, history clearly suggests that this approach was short sighted and unsustainable. The prospect of a continuous flow of new classes of anthelmintics has not been realized; there has not been a new class of anthelmintics introduced into the marketplace in almost 25 years. During the post-ivermectin period, the investment in discovery and development of new anthelmintics has been greatly reduced and there are few new candidate drugs on the horizon. Development of the cyclooctadepsipeptides and/or paraherquamide would be a valuable addition to nematode parasite control, but it is unlikely that sufficient numbers of new drugs will be

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