

# Parasite resistance trends and practical implementation of fecal egg counts

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## Abstract

Anthelmintic resistance in the United States has recently gained the attention of the veterinary and livestock communities. The rapid rise in resistance to 2 new drug classes shortly after approval outside the United States calls for new strategies and recommendations for maintaining an effective anthelmintic program even as new drugs become available. The development of resistance is product-dependent, with observable differences between classes. Methods used to test anthelmintic efficacy allow the veterinary practitioner to make informed treatment decisions; however, the results should be interpreted carefully due to the limitations for the fecal egg count procedure.

**Key words:** anthelmintic resistance, bovine, fecal egg count reduction test, FECRT

## Résumé

La résistance anthelminthique aux États-Unis a récemment retenu l'attention de la communauté vétérinaire et d'élevage du bétail. La croissance rapide de la résistance à deux nouvelles classes de drogues peu après leur homologation à l'extérieur des États-Unis exige de nouvelles stratégies et recommandations pour maintenir l'efficacité du programme anthelminthique avec l'arrivée de nouvelles drogues. Le développement de la résistance varie d'un produit à l'autre avec des différences notables selon la classe. Les méthodes utilisées pour tester l'efficacité anthelminthique permettent au praticien vétérinaire de faire des choix éclairés. Toutefois, les résultats devraient être interprétés avec prudence en raison des limites de la procédure de comptage des œufs dans les fèces.

## Introduction

The ability to achieve an effective anthelmintic control program is both an economic and health concern for United States cattle operators and veterinarians. Since 1964 when the first case of resistance to benzimidazoles was reported,<sup>4</sup> scientists have been working to discern the causes of resistance and develop methods to diagnose and prevent new occurrences. Fifty years of research has led to many discoveries, but resistance remains the greatest challenge to parasite control.<sup>19</sup>

Resistance typically begins at the farm level and develops more rapidly when animals are regularly dewormed. Management practices such as underdosing and improper dosing are important drivers of resistance due to the inherent selection of resistant and/or tolerant worms. Similarly, the use of generic products, which in some cases do not perform as well as the pioneer products, are also implicated. Regardless of the process, anthelmintic resistance is now common. Along with ongoing research to understand the mechanisms of resistance, there is a need for improved cost-effective diagnostic tools so that intervention strategies can be implemented to maintain the economic and health benefits gained from adequate parasite control.

## Anthelmintic Resistance Trends

Negative effects of parasitism are often expressed subclinically and typically only appreciated in the cattle industry through decreased animal performance. Reduced animal performance is often multifactorial, and any reduction in anthelmintic efficacy can go unrecognized. Anthelmintic resistance occurs when a previously susceptible worm population survives treatment and passes its resistant genes to the next generation. A diminished fecal egg count reduction tests is the primary method for detecting reduced drug efficacy. Research into benzimidazole resistance has shown reduced egg count reductions are only evident once 25% of the gastrointestinal nematodes are resistant.<sup>1</sup>

Gastrointestinal nematode (GIN) resistance to the avermectin/milbemycin (AM) drugs class is usually of most concern, since these drugs have been the cornerstone of parasite control since first gaining approval for use in cattle in 1984. Although avermectin resistance was first reported in small ruminants in South Africa,<sup>22</sup> the first report in United States cattle was in 2004 when resistant *Cooperia punctata* and *Haemonchus* spp worms were confirmed at necropsy in a grazed stocker operation.<sup>7</sup> Further evidence was confirmed in a controlled efficacy study where necropsy of treated animals demonstrated that avermectin was ineffective in reducing developing or arrested *Ostertagia ostertagi* and adult *Cooperia onchophora*.<sup>5</sup>

These findings along with the rapid rise in domestic and global publications on anthelmintic resistance in livestock<sup>20</sup> raised the importance of this issue for producers, the veterinary community, and regulatory agencies. As a result, the FDA Center for Veterinary Medicine (CVM) hosted a

public meeting in 2012 with recognized experts in veterinary parasitology. This meeting led to a CVM-directed education campaign titled Antiparasitic Resistant Management Strategy (ARMS) that called for selective use of antiparasitic drugs along with the promotion of management practices to help maintain effectiveness. These strategies were published,<sup>10</sup> presented at educational meetings, and provided on the CVM website.<sup>21</sup> In 2018, the CVM requested that animal drug companies voluntarily revise drug labels of livestock and equine antiparasitic drugs within 1 year to include information on antiparasitic resistance.

The shortage of new anthelmintic classes in the marketplace and the need to preserve the effective anthelmintics currently available, has led some researchers to use computer simulation models to provide insight into parasite dynamics and identify potential management interventions.<sup>2,12,23</sup> Results from computer modeling have challenged previously held principles and advanced new recommendations. For example, it is no longer recommended to annually rotate anthelmintic classes and instead many now advocate for a single anthelmintic class be used until it is no longer effective. Although modeling is a useful tool, there is a large complexity of variables that must be considered when making anthelmintic resistance predictions. Variables include, but are not limited to, parasite and host biology, genetics, host immunity, pharmacokinetic dynamics, individual management decisions, and environmental nuances. Computer simulations predicted that the development of anthelmintic resistance to a new drug class would be markedly delayed if treatment intervals were significantly restricted<sup>11</sup> or if 2 drugs from different anthelmintic classes were used simultaneously.<sup>13</sup> *Haemonchus contortus* resistance, however, was reported to a new class of anthelmintic, monepantel, in sheep after only 4 annual treatments. Monepantel, an amino-acetonitrile derivative (AAD), was the first drug from a new anthelmintic class released since ivermectin 30 years prior.<sup>18</sup> Monepantel was released with instructions for dosing regimens to reduce selection pressure for anthelmintic resistance. However, reports of resistance arose after only 2 years of use in New Zealand and then shortly after in the United Kingdom.<sup>9</sup> In 2014 in Australia, a second new class of anthelmintic, derquantel (a spiroindole), was released in combination with abamectin, and within 2 years of the release reduced efficacy against *H. contortus* egg shedding was reported.<sup>18</sup> Results from these studies demonstrate the complexities and pitfalls involved in predicting the development of anthelmintic resistance, and the current models may not be applicable to new classes of anthelmintics.

It was recently suggested that moxidectin may have a role in the control of macrocyclic lactone-resistant nematodes. Although cross resistance between avermectins and moxidectin occurs, a greater resistance to avermectins is often reported. Moxidectin is a milbemycin and compared to avermectins has a different potency, pharmacokinetic profile, a higher safety profile, and perhaps a unique resistance

profile.<sup>16</sup> In vitro studies suggest that the small structural differences between avermectins and milbemycin alter receptor binding and physiological outcomes such as larval pharyngeal pumping, motility, and development.<sup>15</sup> Also, since moxidectin is lipophilic, it has a wide volume of distribution to the adipose tissue, longer half-life (14 days vs 7 days for avermectins) and tissue persistence.<sup>25</sup> However, in the face of anthelmintic resistance, judicious use of moxidectin should always be implemented to mitigate greater selection pressure.

In recent years, feedlots and grazing operators have often given 2 or more anthelmintics concurrently to achieve desired production endpoints. Published US feedlot studies using combination anthelmintic therapies are limited and varied, but some have shown that there is a performance advantage (weight gains and carcass data) for animals treated with a combination macrocyclic lactone/benzimidazole compared to a macrocyclic lactone alone.<sup>17</sup> A 118-day grazing study compared the efficacy of concurrent treatment at pasture turnout with a macrocyclic lactone/benzimidazole or an injectable macrocyclic lactone with extended activity. In the first 32 days, only the concurrent therapy provided nearly 100% efficacy based on fecal egg count reduction test (FECRT) and a weight gain advantage compared to controls. At the conclusion of the 118-day study, both the macrocyclic lactone/benzimidazole therapy and the extended release macrocyclic lactone had statistically similar weight gains, and the gains were statistically greater than controls. In this study, cattle were harboring macrocyclic lactone-resistant nematodes, and a single combination treatment at the beginning of the grazing season provided benefits throughout the entire period and may have helped to preserve refugia.<sup>6</sup>

### Practical Implications of Fecal Egg Counts

Anthelmintic treatment selection is generally based on the product label indications, perceived efficacy, and cost. Producers and veterinarians are often unaware of the real-time anthelmintic efficacies of the products they are using. The 2 primary methods for testing product efficacy are the controlled efficacy test and the fecal egg count reduction test (FECRT). The controlled efficacy test determines the actual number of worms present in animals before and after treatment by timed necropsy collection. Given the cost and expertise required for controlled studies, some version of the FECRT is the primary method used in the field to determine product efficacy.<sup>3</sup> Results from the FECRT must be interpreted with the knowledge that reduced post-treatment strongyle egg counts do not always correlate with nematode killing. Also, a FECRT does not report the total number of worms, strongyle nematode genera present, immature stages or surviving female worms that temporarily stop shedding eggs after anthelmintic treatment.<sup>24</sup>

Regardless of the limitations, the FECRT is the best in-field test available for monitoring treatment response. Typi-

cally, the FECRT is conducted by performing fecal egg counts (FEC) on samples from approximately 15 to 20 animals before and after treatment. If the FEC is reduced by 95% or more after treatment, then the treatment is considered effective. Fecal egg counts can be conducted in the veterinary clinic or submitted to a veterinary diagnostic laboratory. Based on the 2007 to 2008 USDA National Animal Health Monitoring Service, only 5.7% of beef cattle operations surveyed conduct fecal egg counts. A recently completed 2017 survey will provide further insight when reported.

A comparison of different fecal egg counting methods revealed that the Mini-FLOTAC technique was the most accurate and precise FEC method in ruminants when compared to the standard Wisconsin or McMaster methods.<sup>13</sup> However, the Mini-FLOTAC device is not sold commercially in the United States and the method is only available through veterinary research laboratories. When requesting or performing a FEC, understanding test sensitivity is critical for result interpretation since procedures differ by location. In most laboratories, the Modified Wisconsin or McMaster methods are available, but the sensitivities can vary widely. For example, the Modified Wisconsin typically ranges from 3 to 5 eggs per gram (epg) while the McMaster procedure can range for 8 to 50 epg.

Often the main obstacle to conducting a FEC is the cost of the procedure. Veterinary diagnostic laboratories charge upwards of \$15 per sample and only some laboratories provide discounts for multiple-sample submissions. To reduce sample costs, individual fecal samples can be composited to reduce the number of fecal egg count procedures needed. Composited fecal samples should be prepared from animals that are physiologically similar (e.g. calf or cow) and include the same amount of feces from each animal so that there is equal animal representation within the composite. A recent article also detailed a method for utilizing composited fecal samples for detection of anthelmintic resistance in cattle.<sup>8</sup> This article provided detailed instructions for preparing composite fecal samples, counting the eggs, and evaluating the results. Based on the results presented, a population of nematodes with greater than 95% FECR or less than 80% FECR could be reasonably classified as susceptible or resistant, respectively. However, results between 80 to 95% should be interpreted with caution.

### Conclusions

Anthelmintic resistance to currently available drugs is a significant and complex issue for the livestock industry. In United States cattle, gastrointestinal nematode resistance to avermectins was first reported in 2004. Increased reporting of reduced drug efficacies has resulted in greater awareness by the cattle industry, veterinary professionals, and regulatory agencies. There is a need for new classes of anthelmintics and new strategies to extend their efficacy in the field. Concurrent use of 2 products from different classes

is often practiced in the face of anthelmintic resistance. The rapid development of resistance outside the US to 2 new drug classes is alarming considering the effectiveness of these products was projected to be much longer. Resistant development is product-dependent, with potential differences between the avermectin and milbemycins. Detection of anthelmintic resistance in the field is dependent on the fecal egg count reduction test. However, the veterinary practitioner must recognize and appreciate test limitations and laboratory variance when applying test results to treatment recommendations.

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