

## IN VITRO AND IN VIVO EFFICACY OF ANTIMICROBIAL AGENTS FOR CONTROL OF PORCINE COLONIC SPIROCHAETOSIS



Gerald E. Duhamel, DVM,  
PhD, Diplomate ACVP  
University of Nebraska-  
Lincoln, USA

### INTRODUCTION

In the last decade, major advances in the understanding of spirochaetal colitis have led to recognition of *Brachyspira pilosicoli* as a cause of porcine colonic spirochaetosis (PCS). PCS is an enteric bacterial disease that reduces performance in pigs raised under intensive management practices worldwide. Clinically, PCS is characterized by diarrhoea and decreased feed conversion efficiency performance, which increases the prevalence of pigs with lower than expected market weight.

Since approximately two thirds of total feed costs are associated with the grow-finish phases of production, control of PCS may yield significant savings for producers, particularly in systems that use multiple sites and all-in/all-out management. One way to control PCS is with effective and rational antimicrobial therapy, which can reduce infection by *B. pilosicoli* and maximize the productivity and welfare of pigs raised in intensive management systems.

This article reviews current information on the *in vitro* and *in vivo* efficacy of various antimicrobials against *B. pilosicoli*.

### IN VITRO STUDIES

*In vitro* studies designed to determine the minimal inhibitory concentration (MIC) or sensitivity of *B. pilosicoli* to various antimicrobials were

limited in number, but a flurry of investigations have emerged in recent years. The Table 1 on page 3 shows the results for many of them.

One of the first reports came forth in 1996 after Fellström and associates examined the susceptibility of field strains obtained from a few pigs in Sweden.<sup>1</sup> They found that the MIC<sub>50</sub> of tylosin was 16 compared with 0.025 for carbadox.

In 1998, there were several more reports. When Cizek and associates evaluated isolates from the Czech Republic, the MIC<sub>50</sub> was 32.0 for lincomycin, >128.0 for tylosin and 0.250 for tiamulin, a pleuromutilin.<sup>2</sup> Investigators from Belgium found the MIC<sub>50</sub> was 32.0 for lincomycin, 128.0 for tylosin, 0.015 for carbadox and <0.030 for tiamulin.<sup>3</sup> Duhamel and associates tested North American isolates, and the MIC<sub>50</sub> was 50.0 for lincomycin, 0.015 for carbadox and 0.100 for tiamulin.<sup>4</sup>

A report in 1998 summarizing results from investigations in the UK and Denmark showed "a high level of sensitivity to both tiamulin and valnemulin, but particularly to valnemulin," which is a more recently developed pleuromutilin. There was less sensitivity to tylosin compared to the other antimicrobials studied.<sup>5</sup>

In 1999, a Finnish study indicated that *B. pilosicoli* strains that appeared resistant to tiamulin had been identified among field isolates.<sup>6</sup>



However, in 2002, Kinyon and associates examined *B. pilosicoli* isolates from North American pigs for susceptibility to five antimicrobial agents and found that tiamulin susceptibility was generally only one dilution lower than that of valnemulin, which had low MICs. Nearly all isolates were resistant to tylosin and two of those tested were not susceptible to lincomycin.<sup>7</sup> All the isolates were susceptible to carbadox, as other investigators have found, but carbadox cannot be used in many countries around the world.

Comparative trend data became available in 2004. Cizek and colleagues, again using isolates from the Czech Republic, reported that MICs for tiamulin and valnemulin were generally low and, in addition, had remained “practically unchanged” from 2001 to 2003 when compared with 1996 to 1999. MICs were “generally high” for acetylisovaleryl-tylosin and lincomycin, indicating poor sensitivity.<sup>8</sup>

Trends in susceptibility only to valnemulin were studied by Ripley and associates, who reviewed data from investigations of *B. pilosicoli* isolates from Europe and the United States isolated between 1990 and 1997 in addition to 1997 and 2003. They found low MIC values, indicating good sensitivity, and concluded in their 2004 report that “no obvious trends” were emerging.<sup>9</sup>

On the basis of these investigations, *B. pilosicoli*'s overall pattern of antimicrobial susceptibility appears to be somewhat similar to that of *B. hyodysenteriae*, which causes swine dysentery and has been studied extensively for susceptibility for over three decades.

With both *B. hyodysenteriae* and *B. pilosicoli*, there appears to be widespread resistance to lincomycin and tylosin. *B. hyodysenteriae* that are resistant to tiamulin have been isolated from pigs in Australia, the Czech Republic, Hungary, Germany and the United Kingdom.<sup>10-13</sup> Although it appears that there could be some tiamulin resistance among *B. pilosicoli* isolated in certain countries, such as Finland, *in vitro* studies elsewhere and trend studies have shown no or little change in susceptibility to tiamulin.

*B. pilosicoli* strains are highly susceptible to valnemulin.

## IN VIVO STUDIES

Determining MICs with *in vitro* studies provides a basis for implementing therapeutic regimens, but the only way to fully assess the *in vivo* clinical efficacy of a specific compound is with field studies and experimental challenges. Currently, there are just a few such studies for antimicrobial control of PCS.

In one of these, the efficacy of in-feed lincomycin administration was evaluated at 22, 33 and 110 ppm for 21 days in pigs challenged with a pure culture of *B. pilosicoli*.<sup>14</sup> Clinical signs of diarrhoea and duration of faecal spirochaete shedding were similar between pigs in the non-medicated control group and pigs fed the same ration containing 22 ppm of lincomycin, but reduced diarrhoea and elimination of faecal shedding were recorded in groups fed rations containing 33 and 110 ppm of lincomycin.

Subsequent studies examined the efficacy of in-feed tiamulin administration for control of concurrent PCS and proliferative enteropathy

**Table 1.** Minimal inhibitory concentration (MIC) values (mcg/mL) of antimicrobial agents against *B. pilosicoli* isolated from pigs in different countries.

Antimicrobial	No. of Isolates Examined	MIC range (mcg/mL)	MIC <sub>50</sub>	MIC <sub>90</sub>	Reference	Country
Carbadox	6	<0.012-0.10	0.025	0.100	Fellström et al., 1996	Sweden
	19	<0.0005-0.015	0.015	0.015	Duhamel et al., 1998	US
	4	0.007-0.015	0.015	0.015	Hommez et al., 1998	Belgium
	25	0.03-0.125	0.060	0.060	Kinyon et al., 2002	US
Lincomycin	19	12.5->100.0	50.0	75.0	Duhamel et al., 1998	US
	10	1.0-64.0	32.0	64.0	Cizek et al., 1998	Czech
	4	8.0-32.0	32.0	32.0	Hommez et al., 1998	Belgium
	12	0.25-12.5	4.32	–	Ripley, 1998	UK
	5	0.5-128.0	13.8	–	Ripley, 1998	Denmark
	25	4.0->128.0	32.0	64.0	Kinyon et al., 2002	US
	33	0.5-128.0	16.0	64.0	Cizek et al., 2004	Czech
Tiamulin	6	0.06-0.50	0.125	0.50	Fellström et al., 1996	Sweden
	19	0.05-0.50	0.100	0.50	Duhamel et al., 1998	US
	10	0.06-2.00	0.250	2.00	Cizek et al., 1998	Czech
	4	<0.03-0.06	<0.030	0.06	Hommez et al., 1998	Belgium
	12	0.031-0.50	0.067	–	Ripley, 1998	UK
	5	0.0156-0.0625	0.041	–	Ripley, 1998	Denmark
	51	<0.063-32.0	<0.063	0.125	Fossi et al., 1999	Finland
	25	0.06-8.0	0.125	1.0	Kinyon et al., 2002	US
	33	0.03-8.0	0.125	1.0	Cizek et al., 2004	Czech
Tylosin	6	8.0->16.0	16.0	>16.0	Fellström et al., 1996	Sweden
	10	2.0->128.0	>128.0	>128.0	Cizek et al., 1998	Czech
	4	128.0-128.0	128.0	128.0	Hommez et al., 1998	Belgium
	12	10.0->200.0	31.6	–	Ripley, 1998	UK
	5	2.0->128.0	24.0	–	Ripley, 1998	Denmark
	25	<16.0->512.0	>512.0	>512.0	Kinyon et al., 2002	North America
Valnemulin	12	≤0.015-0.125	0.018	–	Ripley, 1998	UK
	5	0.0156	0.0156	–	Ripley, 1998	Denmark
	25	0.03-2.0	0.06	0.50	Kinyon et al., 2002	US
	33	0.03-4.0	0.125	2.0	Cizek et al., 2004	Czech
	7	0.125-0.25	0.125	0.25	Ripley et al., 2004	Germany
	10	<0.067-2.0	0.25	1.0	Ripley et al., 2004	Italy
	10	0.0075-0.25	0.04	0.25	Ripley et al., 2004	UK
	9	0.03-2.0	0.5	1.0	Ripley et al., 2004	US

caused by *Lawsonia intracellularis* under field conditions.<sup>15,16</sup> In these studies, administration of 150 ppm of tiamulin for 14 to 21 days significantly reduced clinical signs of diarrhoea, improved weight gain and reduced faecal shedding of spirochaetes. In addition to therapeutic use, in-feed administration of tiamulin at 200 ppm for 18 to 30 days - depending on the age of the pigs - combined with thorough sanitation but not depopulation was effective in eradicating PCS from a 60-sow farrow-to-finish farm in Finland.<sup>17</sup> Whether or not this approach is cost-effective for larger operations remains to be determined.

More recently, the efficacy of in-feed valnemulin administration at a rate of 25 ppm (dose rate of 1.25 mg/kg bodyweight/day) for 28 days was evaluated for control of spontaneous PCS.<sup>18</sup> At this concentration, valnemulin significantly reduced diarrhoea and improved weight gain. Similarly, the therapeutic efficacy of in-feed valnemulin at approximately 25 ppm for 14 to 27 days was independently confirmed in two separate studies using pure-culture challenge models of PCS.<sup>19,20</sup> In these studies, valnemulin given after challenge significantly reduced diarrhoea and colonization by the spirochaete.

#### STRATEGIC MEDICATION

In addition to administration of antimicrobials, improving hygiene and limiting access to a manure-contaminated environment are essential management strategies for controlling PCS.

The goal of strategic medication is to reduce transmission of *B. pilosicoli* among commingled

susceptible pigs. A cost-effective scheme in all-in/all-out pig flow systems involves environmental cleaning and disinfection between groups along with a therapeutic level of an effective antimicrobial compound administered in several alternating batches of feed.

When control of environmental contamination is less than optimal, natural exposure should be allowed for 7 to 10 days before initiation of pulse medication. It is possible that hosts other than pigs can be a source of *B. pilosicoli* infection, and exposure of pigs and feedstuffs to rodents and wildlife, particularly mice and birds, should be restricted.

#### ANTIMICROBIAL RESISTANCE

Resistance may stem from indiscriminate long-term use of a single antimicrobial agent as the only control measure for spirochaetal colitis. Various classes of antimicrobials have different modes of action. Therefore bacteria that are resistant to one class of antimicrobials might still be susceptible to antimicrobials from a different class.

The best safeguard against the development of antimicrobial resistance among pathogens may be rotation of compounds effective against pathogenic *Brachyspira*.

#### CONCLUSIONS AND FUTURE PERSPECTIVES

New information generated over the last decade has improved our understanding of PCS and led to the implementation of cost-effective control strategies to reduce the impact of *B. pilosicoli*

infection on pig production. Before an appropriate control strategy can be devised, however, a thorough laboratory diagnostic investigation is essential to correctly identify PCS and any other possible contributing factor(s).

Currently, valnemulin has low MIC values against *B. pilosicoli* *in vitro* and so does carbadox, followed by tiamulin. These appear to be the most effective compounds for control of PCS under field conditions, although carbadox cannot be used in many countries.

Increased MIC values of tiamulin against *B. pilosicoli* have been documented in Finland. However, trend studies incorporating data from several other countries indicate no change in the sensitivity of *B. pilosicoli* to tiamulin over the last decade. Further monitoring will be required to determine whether or not *B. pilosicoli* resistant to tiamulin are emerging and whether or not the efficacy of tiamulin against field outbreaks of PCS remains unchanged.

When the response to antimicrobial therapy is poor, veterinarians should consider potential contributing factors, such as other infectious or non-infectious causes of diarrhoea, an error in dosage and/or inadequate delivery or uptake of antimicrobial agents, poor control of environmental contamination with re-infection and/or antimicrobial resistance of the spirochaete.

Because the pig industry is global and there are a limited number of available, effective antimicrobial agents against pathogenic *Brachyspira*, the potential for emergence of resistant strains has become an international concern. Continued monitoring of pathogenic *Brachyspira* for development of antimicrobial resistance and understanding the mechanisms of resistance are of paramount importance.

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