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**DETERMINATION OF MINIMAL INHIBITORY CONCENTRATIONS FOR VETERINARY MACROLIDES AGAINST BACTERIAL ISOLATES FROM PIGS WITH RESPIRATORY DISEASE**

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 (<sup>1</sup>Pfizer Animal Health, Madison NJ, USA and <sup>2</sup>Quotient Bioresearch Limited, UK.)

**Introduction:** The objective of this study was to determine the Minimal Inhibitory Concentrations (MIC) of the veterinary-approved macrolides tulathromycin (Draxxin<sup>®</sup>, Pfizer Animal Health), tildipirosin (Zuprevo<sup>®</sup>, MSD Animal Health), tilmicosin (Micotil<sup>®</sup>, Elanco Animal Health) and gamithromycin (Zactran<sup>®</sup>, Merial) against bacterial isolates from pigs with naturally-occurring respiratory disease.

**Materials and Methods:**

**MIC testing of Non-Mycoplasma Isolates:** Thirty *Actinobacillus pleuropneumoniae* and 30 *Pasteurella multocida* isolates were retrieved from the US Pfizer Animal Health Development and Research Culture Collection (AHDRCC). Sensitivity testing against tulathromycin, tildipirosin, tilmicosin and gamithromycin was performed at Pfizer Animal Health. Additionally, 30 *Haemophilus parasuis* strains were randomly selected from the European CEESA VetPath III collection and sensitivity against macrolides was determined at Quotient Bioresearch Limited, UK. All tests were performed according to the methodologies for broth microdilution recommended by the Clinical Laboratory Standards Institute (CLSI) except for *H. parasuis* in which results were obtained on agar plates (Mueller-Hinton with 5% laked sheep blood). It was ensured that the pH of the culture media remained within the range 7.2-7.4, a key factor to obtain reliable MIC data for macrolides (Godinho, 2008, Vet Microbiol 129, 426-432). Tildipirosin and gamithromycin were extracted in-house from commercial drug product.

**MIC testing of *Mycoplasma hyopneumoniae*:** MICs of tulathromycin and tildipirosin against 20 *M. hyopneumoniae* clinical field isolates was conducted at Microbial Research, Inc (MRI) in Fort Collins, CO. There is no CLSI standardized procedure available for the susceptibility testing of veterinary *Mycoplasma* species. Compounds were provided by Pfizer Animal Health.

**Results:**

Compound	<i>P. multocida</i> (n = 30)			<i>A. pleuropneumoniae</i> (n = 30)		
	MIC <sub>50</sub>	MIC <sub>90</sub>	Range	MIC <sub>50</sub>	MIC <sub>90</sub>	Range
Tulathromycin	1	2	0.5-2	8	16	4-16
Tildipirosin	1	1	0.5-2	4	8	4-8
Tilmicosin	2	4	0.5-16	8	16	4-16
Gamithromycin	1	2	0.5-8	2	4	1-4

CLSI breakpoints: Tulathromycin: S ≤16 µg/mL, I 32 µg/mL, R ≥64 µg/mL  
 Tilmicosin: S ≤8 µg/mL, I 16 µg/mL, R ≥32 µg/mL  
 No CLSI breakpoints available for Gamithromycin or Tildipirosin

Compound	<i>M. hyopneumoniae</i> (n = 25)			<i>H. parasuis</i> (n = 29)		
	MIC <sub>50</sub>	MIC <sub>90</sub>	Range	MIC <sub>50</sub>	MIC <sub>90</sub>	Range
Tulathromycin	≤ 0.015	≤ 0.015	≤ 0.015	1	2	0.5-2
Tildipirosin	4	16	0.5-16	1	2	0.5-2

No CLSI breakpoints available

**Discussion and Conclusions:**

For non-mycoplasma isolates, no relevant differences in MIC activity were found between the different macrolide compounds tested. MIC<sub>90</sub> values were within CLSI approved susceptible breakpoint values for both tulathromycin and tilmicosin against all strains tested.

For *M. hyopneumoniae*, MIC<sub>50</sub> and MIC<sub>90</sub> values of tulathromycin were remarkably lower than the tildipirosin values.

Elevated macrolide MIC values may be due to the presence of *msr*(E)-*mph*(E) and the higher tilmicosin and tildipirosin MICs could possibly be due the presence of *erm*(42). Isolates that carry all three genes can show further elevated MICs to existing and new macrolides.