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ANTIBIOTIC SUSCEPTIBILITY OF *FUSOBACTERIUM NECROPHORUM* ISOLATED FROM LIVER ABSCESSSES¹

T. G. Nagaraja, K. F. Lechtenberg², and M. M. Chengappa³

Summary

Antibiotic susceptibility patterns of thirty-seven isolates of *Fusobacterium necrophorum* (21 biotype A and 16 biotype B) from liver abscesses of feedlot cattle were determined. These isolates were generally susceptible to penicillins, tetracyclines (chlortetracycline and oxytetracycline), lincosamides (clindamycin and lincomycin), and macrolides (tylosin and erythromycin) but resistant to aminoglycosides (kanamycin, neomycin, gentamycin and streptomycin), ionophores (except narasin), and peptides (avoparcin, polymixin, and thiopeptin). Differences in antibiotic sensitivity patterns were observed between the two biotypes only for clindamycin and lincomycin. The minimum inhibitory concentrations (MIC) of FDA-approved antibiotics for liver abscess control did not parallel their efficacy in preventing clinical liver abscesses in feedlot cattle. Continuous tylosin feeding did not appear to induce antibiotic resistance in *F. necrophorum*.

(Key Words: *Fusobacterium necrophorum*, Liver Abscesses, Antibiotic Susceptibility.)

Introduction

Fusobacterium necrophorum, a gram-negative, anaerobic, rod-shaped bacterium, is the primary causative agent of liver abscesses in feedlot cattle. Two distinct biotypes or subspecies, biotype A (subsp. *necrophorum*) and biotype B (subsp. *funduliforme*), have been recognized. Biotype A is encountered most frequently in liver abscesses. Because of the importance of *F. necrophorum* as an animal pathogen, its antibiotic susceptibility, particularly to clinically relevant antibiotics, has been reported. However, studies on susceptibility to antibiotics used as feed additives have been limited. Also, the difference in susceptibility patterns between the two biotypes have not been reported. Our objectives were to determine the susceptibility of *F. necrophorum* of liver abscess origin to antibiotics, including FDA-approved and certain experimental feed additives, and to determine whether continuous antibiotic feeding during the finishing period would influence susceptibility of *F. necrophorum* to those antibiotics.

Experimental Procedures

Abscessed livers from feedlot cattle in Kansas and eastern Missouri were collected

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at a slaughter house. Lot numbers and cattle origins were recorded to enable obtaining information on antibiotic feeding. *Fusobacterium necrophorum* was isolated from the abscesses. Thirty-seven isolates (21 biotype A and 16 biotype B) were used in the study. The antibiotics tested included: ampicillin, avoparcin, bacitracin, carbenicillin, cephalothin, chloramphenicol, chlortetracycline, clindamycin, cloxacillin, erythromycin, gentamicin, ipronidazole, kanamycin, lasalocid, lincomycin, methicillin, monensin, nalidixic acid, narasin, neomycin, novobiocin, oxytetracycline, penicillin-G, polymyxin-B, salinomycin, streptomycin, tetracycline, thiopeptin, tylosin, vancomycin, and virginiamycin. Antibiotic type is shown in Table 1.

Susceptibility or resistance of *F. necrophorum* isolates to antibiotics was determined by inoculating overnight cultures (16 to 18 hours) into anaerobic media with 100 µg/ml or units/ml (for bacitracin, penicillin-G, polymyxin-B) of antibiotics or no antibiotic. Culture absorbance was measured three times to determine growth. For antibiotics that were inhibitory at 100 µg/ml or units/ml, the minimum inhibitory concentration (MIC) was determined by broth microdilution. The MIC was the lowest concentration of the antibiotic that inhibited growth. Differences in MIC between the two biotypes, between tylosin- and nontylosin-fed cattle, and between chlortetracycline- and nonchlortetracycline-fed cattle were compared by a statistical t test.

Results and Discussion

Fusobacterium necrophorum isolates from liver abscesses were resistant (100 µg or units/ml) to avoparcin, gentamicin, kanamycin, lasalocid, monensin, nalidixic acid, neomycin, salinomycin, streptomycin, tetracycline, thiopeptin, and vancomycin. The

resistance of *F. necrophorum* to ionophore antibiotics explains the lack of influence of monensin or lasalocid on the incidence of liver abscesses.

The isolates were susceptible to ampicillin, bacitracin, carbenicillin, cephalothin, chloramphenicol, chlortetracycline, clindamycin, cloxacillin, erythromycin, ipronidazole, lincomycin, methicillin, narasin, novobiocin, oxytetracycline, penicillin-G, polymyxin-B, tylosin, and virginiamycin. Mean MICs of antibiotics to which *F. necrophorum* was susceptible are shown in Table 1. The MICs of all antibiotic compounds except for clindamycin and lincomycin did not differ between the two biotypes of *F. necrophorum*. For clindamycin and lincomycin, MICs were lower ($P < 0.05$) for biotype A than biotype B (Table 1).

Only 31 of the 37 isolates were from cattle with known antibiotic feeding status during the finishing period. Twenty-three isolates were from tylosin-fed (10 g/ton of feed) cattle and eight were from cattle that did not receive tylosin. Only four isolates were from chlortetracycline-fed (75 mg/head/day) cattle. The mean MICs for tylosin were similar whether or not the cattle had been fed tylosin (Table 2). Similarly, continuous feeding of chlortetracycline had no effect on the MIC of either chlortetracycline or oxytetracycline (Table 2). Apparently, continuous feeding of tylosin or chlortetracycline did not induce antibiotic resistance *F. necrophorum* isolates.

According to the U.S. Feed Additive Compendium, five antibiotics (bacitracin, chlortetracycline, oxytetracycline, tylosin, and virginiamycin) are approved for use in the prevention of liver abscesses in feedlot cattle. Based on MIC, chlortetracycline and oxytetracycline were most effective and bacitracin was the least effective. However tylosin is

most effective in preventing clinical liver abscesses. Except for bacitracin, MIC does not appear related to

clinical efficacy. The mode of action of these antibiotics in preventing liver abscesses is possibly inhibition or reduction of the population of *F. necrophorum* in ruminal contents and (or) in the liver.

Table 1. Mean Minimum Inhibitory Concentrations (MICs) of Antibiotics for *Fusobacterium necrophorum* Isolates from Liver Abscesses^a

Antibiotics ^b	Antibiotic Type	<i>Fusobacterium necrophorum</i> Biotypes		Total (n = 37)	
		Biotype A	Biotype B	Mean	Range
		(n = 21)	(n=16)		
Ampicillin	Lactam	1.6	1.4	1.5	.2-9.4
Bacitracin		50.2	42.7	46.8	9.4-100.0
Carbenicillin	Lactam	1.8	4.4	2.9	.3-18.8
Cephalothin	Lactam	.1	.2	.2	.1-.3
Chloramphenicol		14.1	20.1	16.7	1.6-42.5
Chlortetracycline	Tetracycline	.8	1.5	1.1	.1-6.3
Clindamycin	Lincosamide	.04	.8 ^c	.4	.02-3.1
Cloxacillin	Lactam	.4	1.1	.7	.1-3.1
Erythromycin	Macrolide	3.5	3.0	3.2	.8-6.3
Iprnidazole		.3	2.7	1.9	.2-5.6
Lincomycin	Lincosamide	.04	.7 ^c	.3	.01-3.1
Methicillin	Lactam	.4	.9	.6	.1-6.3
Narasin	Ionophore	2.5	3.3	2.9	.8-4.7
Novobiocin		8.0	4.3	6.4	.2-12.5
Oxytetracycline	Tetracycline	.4	1.5	.9	.03-4.1
Penicillin-G	Lactam	.1	1.3	.7	.1-2.1
Polymyxin-B		37.4	46.0	41.1	8.1-100.0
Tylosin	Macrolide	3.7	7.4	5.3	2.0-12.5
Virginiamycin	Streptogramin	2.8	3.9	3.3	2.3-6.3

^aMean of six replications, each utilizing twofold dilutions from 0.01 to 100.00.

^bConcentrations in µg/ml except for bacitracin, penicillin-G, and polymyxin-B, which are in units/ml.

^cDifferent from biotype A.

Table 2. Effect of Continuous Feeding of Tylosin or Chlortetracycline On Antibiotic Susceptibility of *Fusobacterium necrophorum* Isolates from Liver Abscesses

Antibiotics	Minimum Inhibitory Concentration, µg/ml			
	Tylosin-Fed ^a	No Tylosin	Chlortetracycline-Fed ^b	No Chlortetracycline
	(n = 23)	(n = 8)	(n = 4)	(n = 27)
Tylosin	5.9	5.1		
Chlortetracycline			2.2	1.2
Oxytetracycline			.1	1.1

^aTylosin was fed at 10 g/ton of feed throughout the finishing period.

^bChlortetracycline was fed at 75 mg/head/day throughout the finishing period.