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Bacterial flora of liver abscesses from feedlot cattle fed with or without tylosin

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BACTERIAL FLORA OF LIVER ABSCESES FROM FEEDLOT CATTLE FED WITH OR WITHOUT TYLOSIN

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Summary

Fusobacterium necrophorum was the predominant bacterial isolate from liver abscesses of feedlot cattle fed with or without tylosin. The major difference in the bacterial flora of liver abscesses between cattle groups was the higher incidence of *Actinomyces pyogenes* in the tylosin-fed cattle. Because the minimum inhibitory concentration of tylosin was not different between bacterial isolates from cattle in the two treatments, we concluded that continuous feeding of tylosin does not induce resistance. The source of *A. pyogenes* infection and significance of *A. pyogenes* interaction with *F. necrophorum* in tylosin-fed cattle are not known.

(Key Words: Liver Abscesses, *Fusobacterium necrophorum*, *Actinomyces pyogenes*, Tylosin.)

Introduction

Liver abscesses in feedlot cattle constitute a serious economic problem; their incidence averages 25 to 30% in cattle at slaughter. Liver abscesses results from a disease complex known as the 'acidosis-rumenitis-liver abscess complex.' It is theorized that ruminal acidosis impairs the integrity of the rumen wall, permitting opportunistic bacteria to colonize, cause infection (rumenitis), and then gain entry into the liver via portal blood.

Fusobacterium necrophorum, a normal inhabitant of the rumen, is the primary causative agent of liver abscesses in cattle. Two

subspecies (subsp) of *F. necrophorum*, subsp. *necrophorum* and subsp. *funduliforme* (previously called biotype A and biotype B, respectively) are recognized.

The most common method of liver abscess prevention is to feed a low level of an antibiotic, tylosin³. Cattle fed tylosin (10 g/ton of feed) have significantly lower incidences of abscesses (about 40 to 50% reduction) compared to nontylosin-fed controls. However, tylosin feeding, in most situations, does not prevent liver abscesses completely. It is reasoned that either (1) liver abscesses in cattle fed tylosin may be caused by bacteria other than *F. necrophorum* or (2) continued tylosin feeding may have induced antibiotic resistance in *F. necrophorum*. In order to test this, a study was conducted to compare bacterial flora of liver abscesses from cattle fed with or without tylosin.

Experimental Procedure

Liver abscesses were collected at the time of slaughter from cattle from feedlots that have fed tylosin continuously for at least 2 years and from cattle from feedlots not having fed tylosin for at least 2 years. Each group included five different feedlots. The number of liver abscesses cultured from each feedlot ranged from 1 to 12; 36 abscesses came from the tylosin group, and 41 from the no-tylosin group (Table 1). Abscesses were collected at the slaughter plant, using the following criteria: a) 1 to 5 cm in diameter and b) relatively soft, with active inflammation at the outer edge.

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Slices of liver with intact abscesses (usually one) were placed in plastic bags, sealed, packed in ice, and shipped overnight to the laboratory. The capsule of the abscess was seared with a hot spatula and then opened with a sterile scalpel. The purulent material was collected aseptically under anaerobic conditions (anaerobic glove box) and cultured for anaerobic and facultative bacteria. Isolation and characterization of facultative bacteria were done at the Veterinary Diagnostic Laboratory in the College of Veterinary Medicine.

Susceptibility and resistance of the predominant isolates to tylosin were determined by a micro broth-dilution method, using micro titer plates. Minimum inhibitory concentration (MIC) was the lowest concentration of tylosin that resulted in the absence of growth in the micro titer plate. MIC determinations were replicated three times.

Results and Discussion

The mean incidences of liver abscesses ranged from 6 to 16.7% in feedlots using tylosin and from 7.3 to 29.2% in feedlots without tylosin (Table 1). Anaerobic bacteria were isolated from all abscesses, and facultative bacteria were isolated from 22/36 (61%) and 23/41 (56%) abscesses from tylosin and non-tylosin-fed cattle, respectively (Table 2).

Fusobacterium necrophorum was isolated from all abscesses in both groups. However, the occurrence of subsp. *necrophorum* alone (82 vs 50%) was greater ($P<.05$), and conversely, the occurrence in association with subsp. *funduliforme* (12 vs 42 %) was lower ($P<.05$) in the no-tylosin than the tylosin group. Subspecies *funduliforme* is

less pathogenic than subsp. *necrophorum* and is generally associated with mixed infections.

The other anaerobes isolated from liver abscesses included species of *Porphyromonas*, *Bacteroides*, *Clostridium*, and *Peptostreptococcus*. Among facultative bacteria, *Actinomyces pyogenes* was the most predominant isolate. The incidence of *A. pyogenes* was greater in tylosin-fed than non-tylosin-fed cattle (53 vs 10%). The other facultative bacteria included species of *Streptococcus*, *Staphylococcus*, *Escherichia*, and *Pasteurella*.

Fusobacterium necrophorum and *A. pyogenes* isolates were susceptible to tylosin. MIC values of tylosin for *F. necrophorum* and *A. pyogenes* isolates were similar between isolates from abscesses of cattle fed with or without tylosin (Table 3).

The major difference in the bacterial flora of liver abscesses between cattle fed with and without tylosin was the higher incidence of *A. pyogenes* in the tylosin-fed cattle. This is surprising, because the organism is quite sensitive to tylosin, and we saw no evidence of resistance being induced. Although the origin of *F. necrophorum* found in liver abscesses is well known, the source of *A. pyogenes* is not known. Because *A. pyogenes* is aerobic, it is not a normal inhabitant of the rumen. However, the bacteria may remain dormant and multiply if conditions (such as entry into the liver) become conducive. Also, some synergistic interaction may exist between *F. necrophorum* and *A. pyogenes* in causing liver abscesses.

We conclude that long-term usage of tylosin in feedlots does not promote development of resistance to tylosin in *F. necrophorum* populations. However, tylosin feeding permits a greater incidence of another bacterium, *A. pyogenes*; the reason is not known.

Table 1. Incidence and Severity of Liver Abscesses in Feedlots Using or Not Using Tylosin

Feedlots	Severity of Abscesses ^a			Total	No. of Abscesses Cultured
	A-	A	A+		
	----- % -----				
<u>Tylosin Feedlots</u>					
A	3.4	.7	1.9	6.0	5
B	6.3	2.4	6.1	14.8	4
C	8.3	3.0	5.4	16.7	12
D	7.2	2.7	3.1	13.0	6
E	3.0	4.2	5.3	12.6	9
<u>Non-tylosin Feedlots</u>					
F	7.0	2.6	5.9	15.5	10
G	9.9	3.3	4.9	18.1	10
H	9.4	5.6	6.7	21.7	11
I	2.9	2.2	2.2	7.3	9
J	8.5	7.1	13.6	29.2	1

^aA- = Livers with one or two very small abscesses or abscess scars ; A = Livers with two to four well-organized abscesses generally < 2.5 cm in diameter; A+ = Livers with one or more large active abscesses or many abscesses.

Table 2. Bacteria from Liver Abscesses of Cattle Fed with or without Tylosin ^a

Bacteria	Tylosin	No Tylosin
No. of abscesses cultured	36	41
Anaerobic bacteria	36/36	41/41
<i>Fusobacterium necrophorum</i>	36/36	41/41
Subspecies <i>necrophorum</i> alone ^b	18/36 (50)	34/41(82)
Subspecies <i>necrophorum</i> mixed with <i>funduliforme</i> ^b	15/36 (42)	5/42 (12)
Other anaerobes	22/36 (61)	23/41 (56)
Facultative bacteria	23/36 (64)	13/41 (32)
<i>Actinomyces pyogenes</i> ^b	19/36 (53)	4/41 (10)
Other facultative bacteria	5/36 (14)	9/41 (22)

^aNumbers in parentheses are percentages.

^bChi-square test P<.01.

Table 3. Minimum Inhibitory Concentrations of Tylosin (Mg/L)

Bacteria	Tylosin	No Tylosin	SEM
<i>Fusobacterium necrophorum</i>	10.5	11.5	1.2
<i>Actinomyces pyogenes</i>	3.4	2.6	.8