Dairy Split Sessions

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A Rational Approach to Septic Calves

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Introduction

Despite the fact that bacterial infection is an important cause of morbidity and mortality in the large animal neonate, the early diagnosis remains a challenge for both the veterinarian and the calf manager. In some situations, a focal/localized site of infection can be identified. This leads the clinician to suspect a bacterial component in the problem of his/her patient and to recommend the appropriate therapy. However, septicemia/bacteremia can occur without a detectable primary site of infection. It is also possible that the septicemia/bacteremia state actually precedes the localization of infection. In such case, early antimicrobial therapy would be beneficial. Experienced calf feeders develop good clinical judgment and often initiate early treatment in sick bovine neonates. If a record keeping system exists on the premises, it is easier to follow and to continue appropriate therapy. Because of the continuing increase in public concern about antimicrobial use, we believe that the veterinarian should provide guidelines to help his client appropriately target calves for antibiotic treatment.

Several questions remain that have no single answer. (1) What are the most informative clinical signs on which to base an adequate antimicrobial therapy? (2) How long should the therapy be continued? (3) How many animals really benefit from antimicrobial therapy? (4) How many animals suffering from a bacterial infection are not treated adequately? (5) Which is the best antibiotic to use in calves?

The purpose of this presentation is to share our experience studying septicemia and to target the future goals of research in this field.

Prevalence and Importance of Bacteremia

Estimation of the prevalence of bacteremia in critically ill neonatal calves with severe diarrhea or depression has been studied in a veal calf operation. One hundred ninety neonatal calves, 1-day to 19-days-old were used in 2 studies to estimate and characterize the prevalence of bacteremia. Bacteremia was detected by blood culture in 31% (28/90) of calves in study 1, and in 24% (19/79) of ill calves and 0% (0/21) of control calves in study 2. Among clinically ill calves, the average age was significantly lower in the blood culture-negative group (5.5 d) than in the blood culture-positive group (7.5 d)

(P=0.004). Mean serum IgG concentration was significantly (P=0.0001) lower in blood culture-positive calves (1.146 g/L) than in blood culture-negative calves (3.077 g/L). The mortality rate was significantly (P<0.0001) higher in the blood culture-positive group (57.4%) than in the blood culture-negative group (15.1%).

Bacteria Involved and Their Characteristics

Family

Bacteria cultured from blood included *Escherichia coli* (51% of all isolates), other gram-negative enterics (25.5%), gram-negative anaerobes (5.9%), gram-positive cocci (11.8%), and gram-positive rods (5.9%) (Table 1).

Antimicrobial Susceptibility

Susceptibility of 25 bacterial isolates from study 1 to 7 antimicrobial agents is presented in Table 2. Comparison of susceptibility of *E. coli* to 6 different antibiotics indicated that susceptibility in the 1st study was significantly different from that of the 2nd study for 4 of the 6 antibiotics examined (Table 3).

Table 1. List of bacterial isolates recovered from blood cultures from critically ill calves in 2 different studies.

Family Bacteria	Study 1	Study 2
	n=29	n=22
Gram-negative enterics	**23**	**16**
Escherichia coli	15	11
Klebsiella pneumonia	4	0
Klebsiella oxytoca	1	0
Klebsiella spp.	0	2
Salmonella dublin	1	1
Salmonella typhimurium	2	0
Campylobacter fetus ssp. *fetus*	0	1
Enterobacter cloacae	0	1
Gram-negative anaerobes	**N/A**	**3**
Bacteroides eggerthii	N/A	1
Bacteroides thetaiomicron	N/A	1
Prevotella bivia	N/A	1
Gram-positive cocci	**4**	**2**
Aerococcus viridans	1	0
Staphylococcus aureus	1	0
Staphylococcus hyicus	1	0
Staphylococcus simulans	1	0
Staphylococcus spp.	0	1
Streptococcus spp.	0	1
Gram-positive rods	**2**	**1**
Bacillus spp.	2	0
Listeria spp.	0	1

N/A: no attempt to culture

Table 2. Antimicrobial susceptibility of 25 bacterial isolates from the blood of bacteremic calves in study 1.

Antimicrobial	*Staphylococcus* (n= 3)	*Escherichia coli* (n= 14)	*Salmonella* spp (n= 3)	*Klebsiella* spp (n= 5)	Total (n= 25)
Penicillin	0	N/A	N/A	N/A	0/3
Ampicillin	1	9	1	0	11/25
Amoxicillin /clavulinic acid	0	13	2	5	20/25
Ceftiofur	0	13	1	5	19/25
Gentamicin	1	4	2	5	12/25
Tetracycline	1	1	1	0	3/25
Trimethoprim/sulfas	2	4	3	5	14/25

Table 3. Change in antimicrobial susceptibility of *Escherichia coli* isolates recovered from the blood of bacteremic neonatal calves between summer 1991 (study 1) and 1993 (study 2), and the statistical significance of changes in the proportion of susceptible isolates.

Antimicrobial	Study 1 (n= 14)	Study 2 (n= 11)	Significance
Ampicillin	9	1	P= 0.02
Amoxicillin /clavulinic acid	13	1	P< 0.0002
Ceftiofur	13	6	P= 0.08
Gentamicin	4	8	P= 0.07
Tetracycline	1	0	P= 0.89
Trimethoprim/sulfas	4	7	P= 0.18

Virulence Factors

Twenty-five bovine isolates of *E. coli* were tested for the presence of several virulence factors. Isolates were obtained from 25 different bacteremic calves. Nineteen isolates were typeable and belonged to 9 different O serogroups. The two main O serogroups were 78 (6 strains) and 119 (3 strains). Virulence factors studied included the following: (1) synthesis and excretion of heat-labile enterotoxin (LT), heat-stable enterotoxin a and b (STa, STb), verotoxin 1 and 2 (VT1, VT2), cytonecrotising factor or aerobactin, (2) presence of the protein EaeA, the P fimbriae or CS31a. Resistance to serum was also studied. The most common virulence factor was aerobactin (17/25). Also 15 isolates were resistant to serum. PAPc was present in 5 isolates. None of the isolates were positive for enterotoxins. Only 2 were Eae positive. Although no virulence factor common to all isolates was identified, the capacity to use iron from the host appeared to be an important mechanism in *E. coli* causing bacteremia in neonatal calves.

Clinical Recognition of the Disease

Our first goal was to develop a tool that the prac-

titioner will be able to use and/or offer to his client dealing with large groups of bovine neonates. For this reason, the following clinical sepsis score was developed based exclusively on clinical observations.

Based on the results of a first study, we performed a subsequent study to evaluate prediction of bacteremia in calves through the use of clinical criteria alone. Clinical evaluation was also kept as basic as possible in order for it to be useful for calf managers.

Logistic regression was used to evaluate the relationship between each variable (clinical scores, temperature, pulse, respiratory rate, infection site, age) and the risk of bacteremia. Scores, presence of localized infection and age were included in the model. The scores for each calf were averaged over two evaluations (A.M., P.M.). A total score was computed by adding the 5 averaged individual scores (fecal, hydration, attitude, umbilicus and scleral vessels). The presence of a localized infection was recorded if any of the following clinical entities existed: hypopyon, septic arthritis, subcutaneous abscess and/or purulent nasal discharge. Temperature, pulse, and respiratory rate were not found to be predictive of bacteremia.

Using the optimum cutoff point, sensitivity and specificity estimates of the clinical sepsis model were 76% and 75% respectively. Negative and positive predictive values were 94% and 38% respectively. The low prevalence of bacteremia in this population, possibly due to the sampling of control calves, decreased the positive predictive value. Overall, the clinical sepsis score appeared to predict bacteremia with reasonable accuracy.

Laboratory Support to Diagnosis

Average GGT, glucose and CO_2 levels were significantly greater in the blood culture negative group than in the blood culture positive group ($p<0.03$). Average immunoglobulin concentration, serum protein concentration, and zinc sulfate optical density were also greater in the blood culture negative group ($p<0.08$). Fibrinogen and ketone bodies were significantly greater in blood culture positive calves than in the blood culture negative calves ($p<0.02$). White blood cell count and band neutrophil count were also greater in blood culture positive calves ($p<0.08$).

The average time required for evidence of growth in blood culture bottles was two days. Sixty four percent of the positive samples became cloudy after 24 hours of incubation. Evidence of growth (cloudiness of the bottle) was noticed in 81% of the positive cultures. Positive isolates that showed no cloudiness in the blood culture bottle were *Staphylococcus* sp. and anaerobes.

Take Home Lesson

(1) Bacteremia is a frequent clinical syndrome in critically ill neonatal calves
(2) Gram negative bacteria, especially *E. coli*, are the most commonly isolated bacteria
(3) Antimicrobial susceptibility is difficult to predict and can vary in time
(4) Virulence factors are numerous and variable among *E. coli* isolates
(5) Clinical evaluation can be usefull to predict bacteremia.

Figure

CRITERIA EVALUATED	RESULT OF OBSERVATION	POINTS
Focal site of **infection**	NO	0
	YES	1.5
Age in days	< 7 days	0
	≥ 7 days	1.2
Clinical Score (C.S)		
Hydration: 0 1 2 3		
Sclera: 0 1 2 3		
Attitude: 0 1 2 3		
Umbilicus: 0 1 2 3		
Fecal: 0 1 2 3		
Total C.S.=_____	Total C.S.	
	≤ 5	0
	> 5 and ≤ 8	2.1
	> 8	2.5
	Sepsis score (cumulative points)