Endotoxic Shock in the Bovine: Recent Preventive Approaches

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Immunity directed against common gram-negative core antigens may provide the opportunity for host species to target the environmental contaminants and commensual bacteria, which under the appropriate conditions cause life-threatening gram-negative sepsis and/or endotoxemia. Many of the common livestock diseases, including neonatal coliform septicemia, coliform mastitis, septic metritis and severe enteritis fit this description. The greatest benefits of such immunity may lie in the reduction of nonspecific illthrift and mortality, rather than the treatment or prevention of any single disease. Active and passive immunization and passive immunotherapy targeting the core antigens common to gram-negative bacteria have been attempted in human beings, laboratory animals, and livestocks. Although some studies have failed to demonstrate cross-protective immunity to gram-negative bacteria, prophylaxis has been observed following challenge either by heterologous endotoxin or unrelated, live, virulent gram-negative bacteria. Logistic and theoretical considerations presently limit the widespread application of heterologous, broad spectrum immunoprophylaxis to gram-negative disease.

In gram-negative sepsis, endotoxin stimulates the release of interleukins and arachidonic acid metabolites. Other inflammatory mediators including clotting, kinin and complement cascades are also set in motion, contributing to the altered hemodynamic and hemostatic state frequently associated with fulminant gram-negative sepsis. The early phases of endotoxic shock are characterized by a fall in vascular resistance, inadequate tissue perfusion, hyperventilation and respiratory alkalosis. If shock proceeds uncorrected, these clinical signs are followed by the development of frank metabolic acidosis, cyanosis, altered vascular resistance and cardiac output, coma and death.

Clinical and experimental observations of human patients with fevers, hypotension, metabolic acidosis, complement activation, disseminated intravascular coagulation, and leukopenia followed by leukocytosis, established the central role of endotoxemia in gram-negative infections. These signs have all been duplicated in experimental subjects using purified lipopolysaccharide.

Antigenic Homology of the Gram-negative Cell Wall

The gram-negative bacterial cell wall consists of an inner cytoplasmic membrane and an outer cell wall consisting of (1) mucopolysaccharide-peptidoglycan (2) phospholipid-protein and (3) an outer lipopolysaccharide (LPS) layer. Lipopolysaccharide (LPS) consists of a variable oligosaccharide region linked to highly conserved core polysaccharide and lipid A regions. Variation in oligosaccharide ("O" or somatic) antigens among gram-negative bacteria is well recognized. Variations in hexose molecule numbers, composition and linkage produce a wide array of O antigens. For example, *Salmonella typhimurium* and *S. newport* have identical hexose content, but order and linkage differences result in distinct serologic groups.

In contrast to the heterogeneity of somatic antigens, underlying gram-negative core antigens possess marked chemical, structural and immunologic homology across species, genera and group, making common core antigens attractive candidates as cross-protective immunogens. Lipid A, N-acetylglucosamine, 2-keto-3-deoxyoctonate (KDO), heptose and glucose residues are highly uniform, even across a broad spectrum of distantly related gram-negative bacteria.

Endotoxic activity is associated with the lipopolysaccharide layer of the gram gram-negative cell wall, specifically its lipid A component. Variation in the biologic activity of endotoxin preparations may relate to the presence of lipid A associated protein, aggregation, polysaccharide composition, culture conditions and source organism. Host mediator systems also have profound influence on clinical signs following endotoxin challenge. One example is genetic control of macrophage receptors to endotoxin. The severity of inflammatory responses following LPS challenge has been demonstrated to vary directly with receptor numbers.

A variety of mutants lacking specific enzymes necessary for complete somatic side chain assembly have been isolated. Their absence causes incomplete side chain synthesis, and consequently, exposed common core determinants. These bacteria are termed rough mutant (R-mutant) bacteria, on the basis of colonial morphology. Core

antigen homology has been definitively demonstrated by studies measuring the reactivity of both polyclonal and monoclonal antibodies against heterologous gram-negative bacteria. Rough mutant bacteria are typically described by the capital letter R, followed by a subscript a, b, c, d, or e. In general, Re mutants lack enzymes required in the early stages of somatic side chain assembly. These enzymes link homologous core oligosaccharides to lipid A components. In contrast, Ra mutants have nearly complete lipopolysaccharide synthesis and lack enzymes necessary to attach somatic side chains to the common core oligosaccharide-lipid A complex. *Escherichia coli* (strain J5) is a genetically stable udg-4-epimerase deficient Rc mutant of *E. coli* O111:B4, incapable of incorporating exogenous galactose in the cell wall. Another bacteria often used in investigations addressing cross-protective immunity to gram-negative disease is the R_e mutant of *Salmonella minnesota.*

Passive Immunotherapy

Passive immunotherapy constitutes an adaptation of passive immunization used in clincial settings. These studies have generally involved the administration of high titer antisera to individuals presenting with clinical signs consistent with fulminant gram-negative disease. The salient differences when comparing with passive immunization are 1) the disease process was initiated by natural circumstances, rather than experimental intervention, and 2) inflammatory cascades have already progressed to the point that clincial signs are readily apparent prior to the administration of antisera.

1. *Human bacteremia and shock.* Several clinical trials have reported protection of J5 antisera for severe gram-negative sepsis. The bactericidal capability of J5 antibodies *in vivo* were questioned, but antisera recognizing common core antigens did diminish the serious consequences of gram-negative sepsis and improve the overall prognosis.

2. *Clinical trials in animals.* Studies have shown efficacy of passively administered antibody to rough mutants in canine and equine patients with clinical evidence of endotoxemia due to gastrointestinal disorders. In a recently completed double blind trial, horses with acute abdominal disease were administered either control or J5 hyperimmune plasma, in addition to standard therapy. Decreased mortality and days of hospitalization were noted. A similar study in neonatal foals failed to demonstrate protection from antiserum directed against gram-negative core antigens.

Several studies have suggested that antibody to gram-negative core antigens is a valuable adjunct to rational medical or surgical therapy, improving the prognosis for severe gram-negative disease. Logistic and economic considerations presently limit the widespread prophylactic use of high anti-LPS titer antisera. Additionally, the infectious and idiosyncratic hazards associated with transfusions preclude the routine use of this technique without some positive assurance of benefit. Development of improved assays detecting preclinical, occult endotoxemia may permit clinicians to target immunotherapy on high risk patients; thus, maximizing therapeutic benefits. Furthermore, inflammatory cascades will proceed even after endotoxin is cleared from circulation. Interpretation of the currently available endotoxin assays is complicated by both false positives and false negative results.

Natural Humoral Recognition of Common Gram-negative Core Antigens

A growing body of evidence is beginning to emerge demonstrating the recognition of common gram-negative core antigens in both human and livestock populations.

Humans. Serologic surveys have recently reported the isotype specific recognition of *Escherichia coli* J5 in randomly screened human populations. Hemagglutination titers of the IgM isotype were dramatically higher than IgG titers in all age groups except neonates (less than 1 month). Adult concentrations of either isotype were present by 2 years of age.

Cattle. Randomly selected adult dairy cattle have normally distributed titers to gram-negative core antigens, with a geometric mean IgG1 ELISA reciprocal titer of 544. These titers increase with age, possibly explaining the lower incidence of life threatening sepsis in the offspring of older cows. Cattle with low titers (<1:240) were at a 5-fold greater risk of clinical coliform mastitis.

Pigs. Class specific serologic recognition of gram-negative core antigens has also been reported in neonatal swine (IgG titer, 1,713; IgM titer, 202). Total serum IgG, a measure of total passive transfer, and dam parity were related to higher serum titers recognizing gram-negative core antigens.

Horses. Randomly selected horses had a geometric mean IgG titer of 764 and an IgM titer of 388. Foals, yearlings and adults from a brood mare farm had geometric mean ELISA titers as follows: a) foals: IgG (1,111), IgM (153), b) yearlings: IgG (2,058), IgM (3,013), c) adults: IgG (4,028), IgM (3,840). On the brood mare farm, the addition of new herd mates and the exposures associated with performance, breeding or transportation create potential for repetitive antigenic challenge, possibly accounting for the higher IgG titers observed and the elevated IgM titers noted in yearlings and adults. The lower titers observed in foals were consistent with the isotype selective colostral transfer of immunoglobulins and the decay of passively acquired immunoglobulin.

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The environment of the typical farm provides ample opportunity for exposures to gram-negative bacteria. Consequently, one can readily envision ongoing gram-negative challenge, both by pathogenic and opportunist bacteria. Cross-reacting immunoglobulins would then recognize microorganisms and the resulting immune complexes would be cleared by phagocytic cells, removing both antigen and immunoglobulin from circulation. Half-lives of passively acquired core antigen specific IgG are dramatically shorter than those of total serum IgG in both neonatal cattle and swine. Significant decreases in serum ELISA titers recognizing *E. coli* J5 have been noted following a variety of experimental challenges, including intravenous infusion of *S. typhimurium* LPS, oral administration of virulent *S. typhimurium,* and intranasal administration of *H. pleuropneumonia.* Rapid decay of passively acquired immunoglobulins recognizing gram-negative core antigens has also been noted in human clinical trials. The accelerated decay of core antigen specific IgG suggests heterologous humoral recognition of gram-negative bacteria *in vivo* initiating a preferential consumption, rather than passive degradation and decay of biologically inert compounds. These observations support the hypothesis that immumoglobulin recognizing shared gram-negative epitopes exists in nature and is cross-reactive *in vivo*, providing nonspecific immunity to unrelated gram-negative bacterial species. More recently, naturally occurring passive humoral recognition of common core antigens has been related to the decreased severity of clinical signs following endotoxin challenge. Specifically, titers recognizing lipid A have been correlated with decreased fevers following intravenous endotoxin challenge.

Active Immunity Against Common Core Antigens In Livestock

Active immunization with common core antigens has not been investigated in human beings because of the low incidence of life threatening gram-negative disease. Generally, clinical trials have been restricted to 1) passive immunization only and 2) populations either of presumed high risk or already exhibiting signs of gram-negative disease. To a greater extent than observed in human populations, livestock routinely experience well described gram-negative infections, pneumonia, mastitis, septicemia. This permits the prospective analysis of the effect of naturally occurring or vaccinally increased immunity on the incidence or severity of naturally occurring gram-negative disease. These studies provide the best available models for investigating the effects of vaccination with common gram-negative core antigens.

Cullor et. al. found that clinical signs of endotoxic shock in calves vaccinated with *E. coli* J5 were less severe than in controls. Serum IgG_1 ELISA titers against *E. coli* J5 fell dramatically during the course of the endotoxin infusion, suggesting the clearance of cross-reactive immunoglobulin—LPS immune complexes by the reticulo-endothelial system. Similar protection was noted in J5 vaccinated calves in *S. typhimurium* live organism challenge studies.

Following initial observations that cattle with low naturally occurring titers recognizing *E. coli* J5 experienced a 5-fold increase in the risk of clinical coliform mastitis, Gonzales et. al. conducted a series of experiments investigating the efficacy of R-mutant bacterins in reducing the incidence of coliform mastitis. These studies confirmed that immunization with *E. coli* J5 reduced the incidence of coliform mastitis by 80%.

Natural challenge on farms experiencing endemic pleuropneumonia provided Fenwick et. al. a unique opportunity to study the efficacy of a *E. coli* J5 bacterin in the reducing losses due to porcine *H. pleuropneumonia.* Under field conditions experimental bacterins provided protection, measured by mortality, sick days and days to slaughter, that was indistinguishable from commercial bacterins containing *H. pleuropneumonia* and significantly better than non-vaccinates. Similar results were obtained in experimental challenge models.

Limitations of R-mutant Immunology

For an immune mechanism targeting gram-negative core antigens to successfully prevent or decrease the severity of a disease process, the etiologic agents must possess common core antigens. Cross-protection against gram-positive, viral or protozoal disease is not expected. To achieve any degree of cross-protection, the disease in question either must have a gram-negative component or cause sufficient mucosal damage to permit endotoxin absorption from the gut lumen.

Failures of passive immunization efforts come as no surprise when one considers the limited role of immunoglobulin in the immune response. Antibody recognition and binding has minimal inherent micorbicidal activity. Antibodies serve principally to focus innate and cellular effector mechanisms against foreign antigens. Direct effector mechanisms targeted by antibody include complement mediated cytolysis, opsonization and phagocytosis, and antibody dependent cell mediated cytotoxicity. The administration of antisera to immunocompromised hosts lacking a functional complement cascade or reticulo-endothelial/phagocytic system cannot be expected to enhance survival in the face of a virulent challenge. Unfortunately, this may be the premise on which many passive immunotherapy trials have been based. Of particular interest are trials in neonatal calves and foals that failed to demonstrate therapeutic

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benefits in the treatment of experimentally induced or naturally occurring gram-negative sepsis and shock. Both phagocytic and cytolytic (complement) mechanisms are quantitatively and qualitatively decreased in neonatal livestock.

Passive or active humoral immunity, as confirmed by serology, can only assure the humoral immune status of body fluid compartments that exist in equilibrium with the blood-vascular compartment. Most passive immunization and immunotherapy studies have used hyperimmune serum or plasma as the source of cross-reacting immunoglobulin. Such a preparation will contain immunoglobulins primarily of the IgG isotype. These immunoglobulins are not actively transported across intact mucosal barriers. Consequently, passively administered cross-reacting immunoglobulins will probably not participate in the early phases of enteric or respiratory infections. Furthermore, resident gram-negative flora present at mucosal sufaces may bind and neutralize cross-reactive immunoglobulin, negating the potential for heterologous immunity.

The efficacy of passive immunotherapy can be expected to vary with the length of time between exposure to the pathogen and administration of the antibody. Cellular interactions between the host and endotoxin are set in motion within minutes after exposure, allowing subsequent clinical toxic manifestations to be observed shortly thereafter. The rapid progression of the disease process due to endotoxemia places passive immunotherapy protocols at an obvious disadvantage when viewed in a clinical setting.

Another important limitation of immunoglobulin mediated immunity targeting common gram-negative core antigens is ability of many gram-negative pathogens to survive and persist within phagocytic cells. This property has been best demonstrated in the case of Salmonella species. There is but a poor correlation between serum antibody concentrations in calves and the degree of protection against *Salmonella dublin* challenge. High serum antibody concentrations are often observed in cattle shedding *S. dublin* in their milk or feces, apparently failing to clear the carrier state. Under these circumstances, humoral recognition of homologous core antigens cannot be expected to confer effective immunity. It may, however, decrease the severity of clinical signs by mitigating the effects of endotoxins through one or more of the previously discussed mechanisms.

Summary

Although results to date have been mixed, it appears that at least under some circumstances immunity targeting common core polysaccharide or lipid A epitopes has demonstrated the ability to decrease the severity of diseases caused by heterologous gram-negative bacteria. It probably does this by promoting more rapid reticuloendothelial clearance of gram-negative bacteria and LPS and/or masking pharmacologically active LPS epitopes. This heterologous immunity is probably less efficacious than that provided by homologous immunization. If we accept a concept of disease based on a virulent and unique agent attacking a suceptible host, broad spectrum immunoprophylaxis using R-mutant bacteria will hold little interest. The use of R-mutant bacteria as vaccinal antigens holds the most promise in the case of disease syndromes lacking a single, distinct etiologic agent. Emerging concerns associated with the use of antimicrobial agents as growth promotants may create a niche closely tailored to the use of immunoprophylaxis targeting common core antigens.

Clinical disease is the end-product of pathogen, environmental and host factors. Environmental contamination, meteorologic stressors and impaired host defenses may all tip the delicate balance between health and disease. Additionally, traditional definitions of health and disease may not be adequately descriptive in livestock species, where the unstated goal is optimal productivity, rather than clinical normalcy. Immunity directed against common gram-negative core antigens may provide the opportunity for host species to target the environmental contaminants and commensual bacteria, which under the appropriate conditions cause life-threatening gram-negative sepsis and/or endotoxemia.

Associations between gram-negative disease and a variety of sequelae are a rapidly growing area of research interest. A strong positive correlation between clinical coliform mastitis and altered inter-estrus interval has been noted.[b] Experimental studies have confirmed that intravenous or intramammary LPS infusions are followed by increased serum concentrations of several inflammatory mediators, including PGF_{2a} concentrations in the luteolytic range. R-mutant immunization may provide opportunities to nonspecifically enhance reproductive performance.

In order to gather useful data for evaluation of the efficacy of vaccination, much of the early work in the field of gram-negative core antibodies involved trials using passive protection. Equine antiserum containing antibodies directed against gram-negative core antigens is commercially available.[c] Better methods of delivering passive antibody need to be developed and assessed, including production of monoclonal antibodies. Several such monoclonal reagents have already shown promise in protecting against experimental gram-negative infections. In veterinary medicine, active immunization will likely be economically more feasible. Future vaccinal strategies must focus on ways to stimulate protective immune responses, yet avoid adverse reactions. Colostral transfer of antibodies directed at gram-negative organisms and their interference with active immunization of neonates must be explored.

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Immunity to common gram-negative core antigens is widely practiced in nature. Although antibodies recognizing common gram-negative core antigens will not eliminate the occurrence of coliform mastitis, etc., this enhanced immunity, either by passive or active immunization, may help to decrease the severity of diseases in which nonspecific gram-negative bacteria or their endotoxins play significant roles.

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[a] Spier SJ, JP Lavoie, JS Cullor, et al: Treatment of gram negative sepsis and endotoxemia in horses using core antibody to a mutant *E. coli* (J5). (abstract) Proceedings: 5th International Conference on Equine Infectious Diseases. October 7-10, 1987, Lexington, KY. (in press).

[b] Moore D: The association of abortion or an altered interestrus interval with mastitis in dairy cows: a retrospective study. Master's Thesis. Department of Epidemiology and Preventive Medicine. School of Veterinary Medicine. University of California at Davis. 1987; 25 pages.

[c] Cross protective *Salmonella typhimurium* antiserum, Immvac Inc., Rt. 1 Box 197A, Bass Lane, Columbia, MO 65201.

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References

1. Braude AI. Endotoxic Immunity. Advances in Internal Medicine 1980;26;427-445. 2. Shenep JL, Barton RP, Morgan KA. Role of antibiotic class in the rate of liberation of endotoxin during therapy for experimental gram-negative bacterial sepsis. J Infect Dis 1985;151:1012-1018. 3. Shenep JL, Morgan KA. Kinetics of endotoxin release during antibiotic therapy for experimental gram-negative bacterial sepsis. J Inf Dis 1984;150:380-388. 4. Bradley SG. Cellular and molecular mechanisms of action of bacterial endotoxins. Annu Rev Microbiol 1979;33:67-94. 5. Morrison DC, Ulevitch RJ. The effects of bacterial endotoxins on host mediation systems: a review. Am J Path. 1978;93:527-617. 6. Moore JN, Hardy MM, Hardy GE. Modulation of Arachadonic acid metabolism in endotoxin horses: A comparison of flunixin megalamine, phenylbutazone and a selective thromboxane synthetase inhibitor (UK-38485) Am J Vet Res. 1986;47:110-113. 7. Giri SN, Chen Z, Carroll EJ, et. al. Role of prostaglandins in pathogenesis of bovine mastitis induced by *Escherichia coli* endotoxin. Am J Vet Res 1984;45:586-591. 8. Smith BP. Understanding the role of endotoxins in gram negative disease. Veterinary Medicine 1986;12:1148-1160. 9. Gay CC. Neonatal enteric infections caused by *Escherichia coli*. Problems of immunization in the control of *Escherichia coli* infections. Ann NY Acad Sci 1971;176:336-349. 10. Koterba AM, Brewer BA, Terplee FA. Clinical and clinicopathological characteristics of the septicaemic foal: review of 38 cases. Eq Vet J 1984;16:347-383. 11. Eberhart, RJ. Coliform mastitis. J Am Vet Med Assoc 1977;170:1160-1163. 12. Smith BP. Bovine salmonellosis. Cal Vet 1980;4:27-30. 13. Kreger BE, Craven DE, Carling C, et al. Gram-negative bacteremia. III. Reassessment of etiology, epidemiology and ecology in 612 patients. Am J Med 1980;68:332-337. 14. Elbien AD, Heath EC. The biosynthesis of cell wall lipopolysaccharide in *Escherichia coli*: Properties of a uridine diphosphate galactose 4-epimeraseless mutant. J Biol Chem 1965;5:1919-1925. 15. Braude AI, Ziegler EJ. Protection against gram negative bacteremia with antiserum to endotoxins. In: Nowotny A: Beneficial effects of endotoxin. Plenum Press. New York. 1983;111-125. 16. Fenwick BW, Osburn BI, Olander HJ. Resistance of C3H/HeJ mice to the effects of *Haemophilus pleuropneumoniae*. Infect Immun 1986;53:474-479. 17. Mutheria LM, Crockford G, Bogard WC, et al. Monoclonal antibodies specific for *Escherichia coli* J5 lipopolysaccharide: Cross-reaction with other gram negative bacterial species. Infect Immun 1984;45:631-636. 18. Dunn DL, Ewald DC, Chandan N, et al. Immunotherapy of gram-negative bacterial sepsis: a single murine monoclonal antibody provides cross-genera protection. Arch Surg 1986;121:58-62. 19. Ziegler EJ, McCutchan JA, Fierer J, et al. Treatment of gram-negative bacteremia and shock with human antiserum to a mutant *Escherichia coli*. N E J Med. 1982;307:1225-1230. 20. Fenwick BW, Cullor JS, Osburn BI, et al. Mechanism involved in protection provided by immunization against core lipopolysaccharides of *Escherichia coli* J5 from lethal *Haemophilus pleuropneumoniae* infections in swine. Infect Immun 1987;53:298-304. 21. Chedid L, Parant M, Parant P, et al. A proposed mechanism for natural immunity of enterobacterial pathogens. J Immunol 1968;100:292-301. 22. Morris DD, Whitlock RH. Therapy of suspected septicemia in neonatal foals using plasma-containing antibodies to core lipopolysaccharide (LPS). J. Vet Int Med 1987;1:175-182. 23. Wessels BC, Gaffin SL, Wells MT. Circulating plasma endotoxin (LPS) concentrations in healthy and hemorrhagic enteric dogs: Antiendotoxin immunotherapy in hemorrhagic enteric endotoxemia. J Am Animal Hospital Assoc 1987;23:291-295. 24. Morris DD, Whitlock RH, Corbeil LB. Endotoxemia in horses: Protection provided by antiserum to core lipopolysaccharide. Am J Vet Res 1986;47:544-550. 25. Greisman SE, Johnston CA. Failure of antisera to J5 and R595 rough mutants to reduce endotoxemic lethality. J Inf Dis 1988;157:54-64. 26. Garner HE, Sprouse RF, Green EM. Active and passive immunization for blockade of endotoxemia. Proc Am Assoc Eq Pract 1985;31:525-532. 27. Cullor JS, Fenwick BW, Williams MR, et al. Active immunization with *E. coli* J5 and its protective effects from endotoxic shock in calves. In: Immunobiology and immunopharmacology of bacterial endotoxins. Plenum Publishing Co. N.Y., NY 1986;265-268. 28. Elin RJ, Robinson RA, Levine AS, et al. Lack of clinical usefulness of the limulus test in the diagnosis of endotoxemia. N Engl J Med 1975;293:521-524. 29. Law, BJ, Marks MI. Age-related prevalence of human IgG and IgM antibody to core glycolipid of *Escherichia coli* strain J5, as measured by ELISA. J Inf Dis 1985;151:988-994. 30. Tyler JW, Cullor JS, Osburn BI, et al. Relationships between serum IgG[1] titers recognizing gram negative core antigens and the incidence of clinical coliform mastitis. Am J Vet Res (In press). 31. Cullor JS, Spier SJ, Tyler JW, Smith BP. Antibodies that recognize gram negative core antigens: How important are they? Proceedings of the Sixth Annual Veterinary Medical Forum. 1988:503-508. 32. Tyler JW, VL Douglas, JS Cullor, et al: Preferential decay of serum IgG recognizing gram negative core antigens in neonatal livestock. (abstract #287) Proceedings: 68th Annual Conference of Research Workers in Animal Disease. Chicago, IL. November 16-17, 1987. 33. Micheals, FH, KL Banks. Contribution of various host factors to resistance to experimentally induced bacterial endotoxemia in calves. Am J Vet Res 1988;49:557-562. 34. Gonzales RN, JS Cullor, DE Jasper, et al: Efficacy and safety of a heat killed J5 *Escherichia coli* vaccine to prevent post-calving clinical coliform mastitis in dairy cows. (abstract #286). Proceedings: 68th Annual Conference of Research Workers in Animal Disease. Chicago IL. November 16-17, 1987. 35. Fenwick BW, Osburn BI, Cullor JS, et al: Mortaility in swine herds endemically infected with *Haemophilus pleuropneumoniae*: Effect of immunization with cross-reacting lipopolysaccharide core antigens of *Escherichia coli*. Am J Vet Res 1987;47:1888-1891. 36. Banks KL. Host defences of the newborn animal. J Am Vet Med Assoc 1982;180:1053-1055. 37. Smith BP, Habasha FG, Reina-Guerra M, et al. Immunization of calves against salmonellosis. Am J Vet Res 1980;41:1947-1951.

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