# Pulmonary Emphysema in Weaned Calves: Laboratory Diagnosis

**Jerre L. Johnson, D.V.M., Ph. D.**
**Jerry L. Pommer, B.S.**
**Donald B. Hudson, D.V.M.**
*Veterinary Science Laboratory, University of Nebraska North Platte Station, North Platte, NE 69101.*
and
**Alan R. Doster, D.V.M., Ph. D.**
*Veterinary Diagnostic Center, University of Nebraska, Lincoln, NE 68583.*

© Copyright American Association of Bovine Practitioners; open access distribution.

Classification of pneumonias takes on many characteristics, each of which seem to add to the confusion in diagnosing respiratory problems. Pierson divides respiratory feedlot yearling deaths into three groups: bronchial, interstitial, and metastatic. He then describes each entity of that group. He classifies pulmonary emphysema of weaned calves as an interstitial pneumonia.

**Other respiratory entities in this group with confusing morphological terms are: bovine pulmonary emphysema; acute bovine pulmonary emphysema, or ABPE; St. George disease; fog fever; atypical interstitial pneumonia, which is currently the most popular; pulmonary adenomatosis; silo filler's disease; cow asthma; hypersensitive pneumonitis; extrinsic allergic alveolitis; "Urner pneumonie"; bovine farmer's lung; acute alveolar emphysema and edema; panters; and lungers.**[8 18 27]

The etiological agents of these conditions may be separate or multiple. The most common etiology of pulmonary emphysema in weaned calves in central Nebraska appears to be bovine respiratory synctial virus (BRSV). This generally occurs as a mixed infection. *Pasteurella multocida* and *Pasteurella hemolytica* are the most common secondary bacteria we isolate. Other bacteria isolated have been: haemophilus, *E.coli,* corynebacterium, streptococcus, and pseudomonas, with viruses of PI, BLVD, and IBR being identified by virus isolation or fluorescent antibody (FA) testing.

Emphysema is overinflation of the alveoli. The alveoli are distended with air which cannot escape via the bronchioles or through the capillary bed. This disease is characterized by extensive alveolar and less extensive interstitial emphysema. The immediate cause of the emphysema is a dilatation, thinning, and eventual rupture of the alveolar walls.[27] Emphysema is common in cattle in lesions of pneumonia and inflammatory diseases of the lung. Forced breathing and gasping exaggerate the emphysema, causing possible rupture of affected alveoli. In severe cases, large, thin-walled, air-filled bullae may develop, leading to fatal pneumothroax. Hypertrophy of the right heart accompanies diffuse emphysema, and if the disease is prolonged and progressive, cardiac failure occurs.[4]

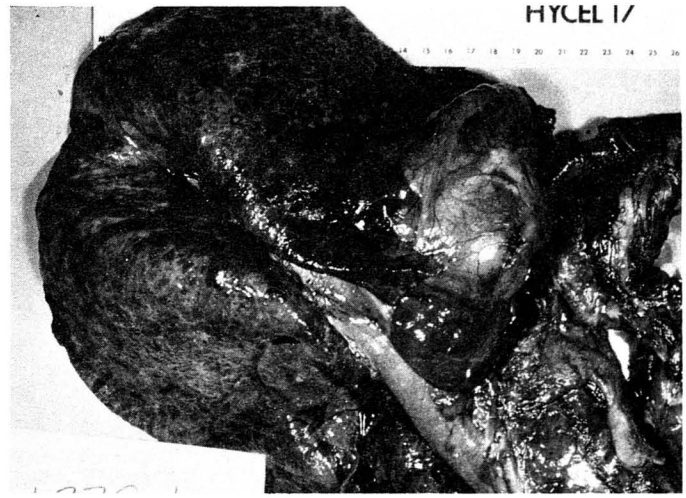

Figure 1. Lung of a 3-day-old calf with interstitial emphysema.

*Pathogenesis*

The respiratory syncytial virus (RS) is classified in the genus *Pneumovirus* of the family paramyxoviridae. The virus is antigenically related to the human RS virus.[5]

The BRSV virus is widespread with reports from Japan, Europe, United Kingdom, Ireland, Canada, and the United States. Serologic studies from these areas indicate the virus is probably widespread in the cattle population.[1 3 9 10 16 17 21]

In humans, the RS virus is the most significant respiratory viral pathogen of infants and children, especially during the first 6 months of life; however, it is uncommon in the first 4 weeks of life.[12] It is probably transmitted by the airborne route.

Hall *et al.* found the virus in 35% of the babies hospitalized out of 82 neonates studies. The mean age was 15 days; the

youngest infant was 6 days of age. The clinical characteristics of the neonates infected were that they had a shorter mean gestation and mean birth weight. Sixty-one percent of the infants showed respiratory illness; the others had nonspecific signs such as lethargy, irritability, or poor feeding.[7]

Stokes *et al.* have indicated 50% of children with documented RSV infection develop future wheezing.[26]

Welliver *et al.* reported 70% of children with RSV infection develop nasopharyngeal epithelial cell-bound IgE. Persistence of the cell-bound IgE is more common in RSV bronchiolitis than upper respiratory infection or pneumonia. This also seems related to family history of wheezing.[31]

In cattle, the natural route of transmission of the virus is obscure. Airborne, direct contact, and inanimate objects such as feedbunks, water troughs, and vaccination with common needles have been incriminated. Fetuses of naturally infected dams have been found to be infected. The presence of the virus in the milk may indicate that newborn calves are infected from colostrum.[29] In calves ingesting colostral antibody, the antibody disappears at about 12 weeks after birth.[32]

In a herd we studied, we found positive FA cells in 2 cows and calves out of 14 we examined.

The youngest pulmonary emphysema we have observed was a 3-day-old calf which was positive on FA for BRSV. Leipold *et al.* have described a 24-hour-old calf with a large air cavity on radiograph. This calf was posted at 3 days of age and grossly had large bullous interstitial emphysema.[13]

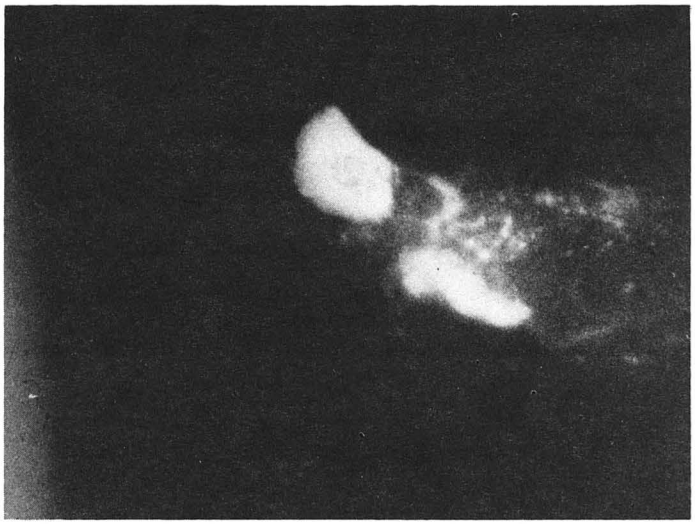

Figure 2. Right lung of a weaned calf. The apical and cardiac lobes are consolidated while the diaphragmatic lobe has large areas of bullous and interstitial emphysema.

*Clinical Signs*

This disease occurs mostly in the fall. It may occur in the spring, while the calf is still on the cow, in the form of a dry cough. The calves stand with heads distended, show severe depression, increased respiration, frequent coughing and abdominal expiration. Nasal discharge is from serous to mucopurulen conjunctivitis, salivation, and in the more advanced cases, subcutaneous emphysema developed all around the body.[1,2,8,20,30,32] This readily appears around the throat and neck areas. Observations in some herds have shown edema of the throat and neck.

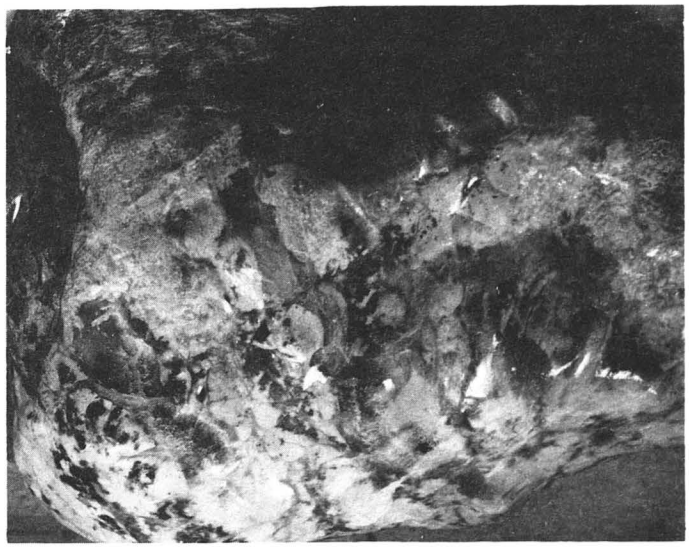

Figure 3. Immunofluorescence of nasal epithelial cells from a positive BRSV herd.

**Table 1. Clinical differential diagnosis.**

| Signs | IBR | BVD | BRS |
|---|---|---|---|
| Upper respiratory dyspnea or mouth breathing | + | | |
| Severe depression | | + | + |
| Decreased appetite | + | + | |
| Increased respiration rate | | Highest in BRS | |
| Frequent coughing | | | + |
| Excessive salivation | | + | |
| Lesion-mucous membrane of the mouth: | | + | |
| of the nose: | + | | |
| Pneumonia | | + | + |

© Copyright American Association of Bovine Practitioners; open access distribution.

*Postmortem Findings*

On opening the thoracic cavity, the lungs do not collapse. Subpleural and interstitial emphysema may be present in all lobes, with emphysema most commonly present in the diaphragmatic lobes. The diaphragmatic lobes may contain large emphysematous boluses. Lobular demarcation is accentuated and 75% of the parenchyma is affected. The affected lung is firmer than normal (hepatized). Its color is reddish-pink and it is slightly cyanotic appearing and meaty-like (adenomatosis). On cutting, the lung is edematous and foamy fluid flows from the cut surface. This is not limited to anteroventral part of the lung as in usual bronchopneumonia. Consolidating pneumonia may be associated, generally in the apical and cardiac lobes. *Pasteurella hemolytica or multocida* are generally isolated from these lobes. There is froth in the trachea and bronchi, and the bronchial lymph nodes are moderately enlarged. Emphysema is also present in the tissue around the aorta, kidney, and pericardial sac.[2 8 14 15 20]

Tracheitis varies from mild to areas with clear to hemorrhagic edema of the mucous membrane. Trachea hemorrhages vary from petechial to massive.

**One should remember the white froth in the trachea and bronchi was serum from the lung that can be mixed to a froth by terminal gasping or even the elastic recoil of the lung after death. The froth may be involved in the animal's death but is not necessarily a factor in the disease process.**

*Histopathologic Changes*

Miscroscopic lesions of pulmonary emphysema vary from a bronchiolitis and alveolitis to those of bronchiolitis obliterans.

Thomas reports that the dominating histopathological response in both experimental and natural occurring BRSV disease was an acute bronchiolitis and alveolitis.[28] Pirie states that calves posted week 7 after one outbreak of positive BRSV did not have interstitial emphysema. Microsopically, these lesions were out of either bronchiolitis obliterans or lymphocytic bronchitis.[20]

Microscopic description by Pirie of a classical BRSV infection has been characterized by large multinucleated giant cells and syncytial cells which are larger and have more nuclei than PI.[3] The syncytia are found 14 and 21 days after infection. The alveolar walls are thickened due to cellular infiltration. There is accumulation within the alveolar air spaces of large numbers of macrophages and neutrophils. The large multinucleated syncytia that are present contain large hyperchromatic elliptical overlapping nuclei varying 2 to 8 in number. With the cytoplasm of the syncytia, eosinophilic inclusion bodies were sometimes seen.[19]

Other findings are as follows: alveolar emphysema and/or edema; interstitial edema; hyperplasia of alveolar epithelium; hyaline membrane development; lumens of the bronchi contain septal cells and desquamated epithelial cells; eosinophils present in some part of lung; neutrophils, macrophages and giant cells cause focal aggregations in alveolar spaces or diffusely scattered; atelectasis observed adjacent to emphysematous areas; hemorrhages from anoxia or rupture of alveolar capillaries; hypertrophy of pulmonary musculature; fluid-filled lymphatic vessels.

It appears that the histopatholog changes of BRSV depend on the duration of infection as well as secondary bacterial and viral infection. Not all cases we have observed have shown microscopic changes, which is in accord with Motean *et al.* outbreak.[16]

*Laboratory Diagnosis*

Besides clinical symptoms, gross pathology, and histopathology, BRSV can be confirmed by virus isolation, serology, and fluorescent antibody technique.[23 28 30] The enzyme-linked immunosorbent assay (ELISA) test is being utilized in human medicine for serological measurement.

As the virus is very difficult to isolate and serology takes considerable amount of time, the FA techniques, although sporadic, offer the greatest potential for rapid diagnosis to the veterinarian and livestock producer. This was first demonstrated on field cases by Jolly and Ditchfield in 1965 and Wellemans in 1976. Welleman explains the difficulty in diagnosis lies in the fact that the BRSV often produces a very rapid immunological response in the animal, so that the maximum titer is attained within a week. Ground lung tissue from the dead animal may not only contain the virus and its antigens but also neutralizing antibodies to the virus, thus impeding its isolation.[30]

We are utilizing direct FA technique with the conjugate being prepared by Dr. Frey and Mr. Rhodes at the Veterinary Science Department, University of Nebraska-Lincoln.[6] The nasal swabs are being fixed immediately after they are taken with acetone. This has increased our positive findings in the laboratory but more so in field cases. This procedure is outlined with the case history report.

The following case history is one of the more interesting pulmonary emphysema outbreaks. The outbreak occurred on December 12, 1980 in a herd of 400 purebred bull calves. From December 15, 1980 to December 23, 1980, we examined 19 of 25 animals that dies in this herd. Seven of eleven bulls that died were from one sire, while eighteen of twenty-five bulls that died were sons or grandsons of the same sire. This examination included gross necropsy, histopathology, bacteriology, and fluorescent antibody techniques. Paired serology and virus isolation were conducted by Dr. Frey of the Veterinary Science Department, University of Nebraska-Lincoln.

All calves showed evidence of pulmonary emphysema, except one which died of apparent bloat. None of the lungs of the pulmonary emphysema calves collapsed when the thoracic cavity was opened. This emphysema varied from small pinpoint areas in the diaphragmatic lobe to several boluses. Lobular demarcation was acentuated in all cases, causing reddish-pink "meat-like" appearing parenchyma. Fourteen animals showed consolidation in the apical and cardiac lobes. Lymph node involvement was present in all cases; this varied from slight to moderate enlargement.

Froth was present in the trachea in six cases. Fifteen

© Copyright American Association of Bovine Practitioners; open access distribution.

animals showed various types of hemorrhage. This varied from petechial to 2 animals whose tracheas contained free blood. Hemoglobin inhibition occurred in 4 cases. Tracheal edema occurred in 6 cases, 3 of which measured 4 cm involvement.

Hemorrhage occurred in the larynx of all calves except 2. This varied from petechia to hemoglobin inhibition which occurred in 4 animals.

Fifteen animals had inflamed turbinates, two had hemorrhages, and two were clear. One animal with inflamed turbinates also had a purulent discharge.

Ulcers were noticed on the buccal cavity of 1 animal.

Emphysema was present around the aorta in 4 cases, kidney and esophagus in 3 cases, and pericardial sac in 2 cases.

Twelve animals had increased fluid in the pericardial sac; in 3 animals, this was clear and in 9 it was blood tinged. Nine animals had hemorrhage on the heart; 3 were petechial, 4 were ecchymotic, and 2 were paint-brush hemorrhages.

*Bacteriology:*

All lungs were routinely cultured on 5% sheep blood and 5% bovine blood chocolate agar.

*Pasteurella multocida* was isolated from 11 animals; *P. hemolytica* from 1 animal; *E. coli* from 3 animals; and 2 had *Corynebacterium pyogenes.* Four animals did not have significant bacteria.

*Light Microscopy:*

Tissues were fixed in 10% buffered formalin and processed using standard techniques, and stained with hemotoxylin and eosin.

Lung samples were obtained from the dorsal border of the diaphragmatic lobe approximately 15-20 cm behind the apical lobe. Nasal turbinate sections were obtained from the anterior and posterior one-third of the ventral turbinate.

Microscopic lesions observed varied from animal to animal. These changes were alveolar emphysema, alveolar edema, interstitial edema, hyperplasia of alveolar epithelium, hyaline membrane development, eosinophils, hemorrhages and diffusely scattered neutrophils and macrophages.

Changes in the nasal concha showed loss of respiratory cilia and vacuolation of the epithelial cells in the respiratory mucosa. The lamina propria contained erectile veins engorged with blood, edema in the lamina propria and foci of lymphocytes. Severity of these lesions increased in the posterior one-third of the turbinates.

*Fluorescent Microscopy:*

Fluorescent antibody tests for bovine respiratory synctial virus were made on various areas of the nasal turbinates. In this herd, nasal swabs taken from live animals showing symptoms of atypical interstitial pneumonia did not reveal fluorescing cells. On those animals necropsied, the heads were split by mid-line sagittal sections to reveal the nasal turbinate areas. Swabs were taken from 12cm, 20 cm, and 25 cm from the nasal opening (Table 2). Smears were made on glass slides and fixed in acetone for 10 minutes. A direct bovine respiratory syncytial virus conjugate with a dilution of 1:60 was added to the fixed nasal smears. Slides were placed in a humid chamber fan incubated in a $CO^2$ incubator for 20 minutes. Excess conjugate was removed by using distilled water, then rinsed in PBS for 10 minutes & a final rinse in distilled water. Fixed slides were allowed to air dry, FA mounting fluid was added to the slides and were coverslipped. Slides were observed on a Zeiss microscope with a halogen light source. We were able to demonstrate fluorescence on 15 animals at the area 21-25 cm from the nares, while only 1 fluoresced at 1-12 cm. (Table 2)

**Table 2. Fluorescent antibody results.**

| Cattle #: | Turbinate Area from Nares (1-12 cm) | (13-20cm) | (21-25cm) |
|---|---|---|---|
| 3174 | - | - | + |
| 3307 | – | + | ++ |
| 3209 | – | – | + |
| 3041 | – | + | + |
| 3129 | – | + | + |
| 3135 | – | – | + |
| 3187 | – | – | + |
| 3191 | – | + | ++ |
| 3270 | – | – | – |
| 3326 | – | – | + |
| 3105 | – | + | + |
| 3154 | + | + | – |
| 3055 | – | – | + |
| 3190 | – | + | ++ |
| 3051 | – | – | – |
| 3205 | – | ++ | + |
| 3072 | – | + | ++ |
| 3038 | – | + | ++ |
| Total positive | 1 | 10 | 15 |

\- *negative*

\+ *few fluorescent cells*

× + *many fluorescent cells*

## Conclusion

Pulmonary emphysema which occurs in calves about 2 months after weaning is a recognized cause of sickness and death in numerous herds in central and western Nebraska. The condition has been recognized as an independent entity for at least 10 years at the North Platte Station Veterinary

© Copyright American Association of Bovine Practitioners; open access distribution.

Science Laboratory. Until the last few years, however, the etiology remained obscure. Veterinarians and livestock producers believe that weather, feeding practices and genetic factors may play a role in the disease etiology. In recent research it has been shown that there is active infection with bovine respiratory syncytial virus (BRSV) in calves at the time of the disease. The virus was isolated early in the disease in most cases studied in the fall of 1979, and affected herds almost invariably seroconverted to an antibody-positive state for BRSV at the time of the outbreak.

**In 1980 fluorescent antibody techniques were used in conjunction with serology and virus isolation in numerous herds. We are now routinely checking respiratory cases for BRSV by Fluorescent antibody techniques.**

### References


1. Breukink, H.J. 1978. *Enzootic bronchopneumonia in young cattle, a constant challenge to the clinician.* Respiratory Disease in Cattle. The Hague, Netherlands. pp., 522-524. - 2. Bryson, D.G., J.B. McFerran, H.J. Ball and S.D. Neill. 1978. *Observations on outbreaks of respiratory disease in housed calves. Epidemiological, clinical and microbiological findings.* Vet. Rec. (Nov. 25.) pp. 485-489. -3. Bryson, D.G., J.B. McFerran, H.J. Ball and S.D. Neill. 1979. *Observations on outbreaks of respiratory disease in calves associated with parainfluenza type 3 virus and respiratory syncytial virus infection.* Vet. Rec. (Jan 20.) pp. 45-49. - 4. Cheville, N.F. 1976. *Cell Pathology.* The Iowa State University Press, Ames. p. 339. - 5. Fenner, F. 1976. *The classification and nomenclature of viruses.* Arch. Virol. 51:141-149. - 6. Frey M. Personal communication. - 7. Hall, C.B., A.E. Kopelman, R.G. Douglas, J.M. Gieman, M.P. Meagher. 1979. *Neonatal respiratory syncytial virus infection.* New England J. Med. (Febr. 22.) 300:393-396. - 8. Hibbs, C.M. 1978. *Pulmonary emphysema in newly weaned calves.* Proc. 11th Annual Convention of the Amer. Asso. of Bovine Practitioners. pp. 125-129. - 9. Holzhauer, C. 1978. *Bovine respiratory syncytial virus infection and symptoms of atypical interstitial pneumonia.* Respiratory Diseases of Cattle. The Hague, Netherlands. pp. 216-223. - 10. Holzhauer, C. and A.C.S.M. Wertenbroek. 1979. *Results obtained using an attenuated bovine syncytial virus vaccine against bronchopneumonia yearlings.* Tijdschr. Diergeneesk. deel. 104:674-678. - 11. King, J.M., F.S. Hsu, C.B. Hong and R.C.T. Lee. 1976. An atlas of general pathology with special reference to swine diseases. Joint Commission on Rural Reconstruction. Republic of China. (May). - 12. Knight, V. 1973. *Viral and Mycoplasmal Infections of the Respiratory Tract.* Lea & Febiger, Philadelphia. pp. 131-140. - 13. Leipold, H.W., M.M. Kaye and O.M. Radostits. 1973. *Interstitial emphysema in a neonatal calf.* VM/SAC (Sept.) pp. 1040-1043. - 14. Mohanty, S.B., A.L. Ingling and M.G. Lillie. 1975. *Experimentally induced respiratory syncytial viral infection in calves.* Amer. J. Vet. Res. (April) 36:417-419. - 15. Monlux, W.S. and A. W. Monlux. 1972. *Atlas of Meat Inspection Pathology.* Agriculture Handbook No. 367. ARS/USDA. (May) p. 160. - 16. Moteane, M., L.A. Babink and B. Schiefer. 1978. *Studies on the occurrence and significance of bovine respiratory syncytial virus in Saskatchewan.* Can J. Comp. Med. (April) 42:246-248. - 17. Odegaard, O.E. and J. Krogsrud. 1977. *A field outbreak caused by bovine respiratory syncytial virus.* Acta. Vet. Scand. 18:429-431. - 18. Pierson, R.E. and R.A. Kainer. 1980. *Clinical classification of pneumonias in cattle.* Bovine Practitioner. (Nov.) No. 15, pp. 73-76. - 19. Pirie, H.M. 1978. *Some pulmonary lesions of calves and their significance.* Respiratory Diseases of Cattle. The Hague, Netherlands. pp. 391-405. - 20. Pirie, H.M., L. Petrie, C.R. Pringle, E.M. Allan and G.J. Kennedy. 1981. *Acute fatal pneumonia in calves due to respiratory syncytial virus.* Vet. Rec. (May 9) pp. 411-416. - 21. Rosenquist, B.D. 1974. *Isolation of respiratory syncytial virus from calves with acute respiratory disease.* J. Inf. Dis. (Aug.) 130:177-181. -22. Rossi, C.R. and G.K. Kiesel. 1974. *Serological evidence for the association of bovine respiratory syncytial virus with respiratory tract disease in Alabama cattle.* Infection and Immunity. *(Aug.) pp. 293-298. - 23.* Rossi, C.R. and G.K. Kiesel. *1977 Bovine respiratory syncytial virus infection of bovine embryonic lung cultures: A kinetic study by the fluorescent antibody technique.* Am. J. Vet. Res. (Nov.) 38:1901-1904. - 24. Smith, M.H., M.L. Frey and R.E. Dierks. 1974. *Isolation and characterization of a bovine respiratory syncytial virus.* Vet. Rec. 94:599. - 25. Smith, M.H., M.L. Frey and R.E. Dierks. 1975. *Isolation, characterization, and pathogenicity studies of a bovine respiratory syncytial virus.* Arch. Virol. 47:237-247. - 26. Stokes, G.M. et al. 1981. *Lung function after bronchiolitis.* J. Pediatr. 98:871. - 27. Swenson, M.J. 1977. *Duke's Physiology of Domestic Animals.* 9th edition. Comstock Publishing Associates, Ithaca. p. 310. - 28. Thomas, L. H. and E. J. Scott. 1981. *Diagnosis of respiratory syncytial virus infection in the bovine respiratory tract by immunofluorescence.* Vet. Rec. (May 16) 108:432-435. - 29. Van Der Maaten, M., W. Hubbert, A. Booth, J. Bryner and P. Estes. 1973. *Isolations of bovine syncytial virus from maternal and fetal blood.* Amer. J. Vet. Res. 34:341-343.- 30. Wellemans, G. 1977. *Laboratory diagnosis methods for bovine respiratory syncytial virus.* Vet. Sci. Commun. 1:179-189. - 31. Welliver, R.C., *et al.* 1980. *The appearance of cell-bound IgE in respiratory tract epithelium after respiratory-syncytial-virus infection.* New England J. Med. 303:1198. -31. *Woods, G.T. 1974. Bovine parvovirus 1, bovine syncytial virus, and bovine respiratory syncytial virus and their infections.* Advances in Vet. Sci. and Comp. Med. 18:273-286.


*Editor's Note: Dr. Merwin Frey's paper will appear in the 1982 Bovine Practitioner.*

© Copyright American Association of Bovine Practitioners; open access distribution.