



Diagnostic Exercise

From the Latin Comparative Pathology Group and the Davis-Thompson Foundation:

Dictyocaulosis in cattle

Adriana L. Stigger¹, David Driemeier², Ana L. Schild³, Rafael P. Lima², Fernanda F. Perosa²,
Irene Guterra¹, Allanis M. Silveira¹

¹Laboratory of Veterinary Pathology, Centro Universitário da Região da Campanha (Urcamp), Alegrete, RS, Brazil

²Setor de Patologia Veterinária, Faculdade de Medicina Veterinária, Universidade Federal do Rio Grande do Sul (UFRGS), Porto Alegre, RS, Brazil.

³Laboratório Regional de Diagnóstico (LRD), Faculdade de Veterinária, Universidade Federal de Pelotas (UFPel).

*Corresponding author: Corresponding author: astigger@hotmail.com

Clinical History:

In October (spring), on a farm in southern Brazil (29° 47' 02" S 55° 47' 28" W), 84 10 to 12 months old mixed breed calves out of a herd of 120 presented anorexia, weight loss, cough, respiratory distress, dyspnea, and serous nasal discharge. Some calves had open-mouth breathing, with extended necks and altered stance (with their forelimbs apart, in an "air-hunger" posture). Mortality was reportedly "high", though the exact number of deaths was not mentioned.

Gross Lesions:

Two of the dead calves were submitted for necropsy (Fig. 1). They exhibited rough hair coats and were in poor body condition. The lungs were heavy and mottled, with multifocal raised white soft areas (normal or compensatory emphysematous areas) alternated with red prominent firm (pneumonia) or depressed soft (atelectasis) red areas. (Fig.2). The cut surface of the lungs had a similar appearance (Fig.3). Large amount of foam filled the lower portion of the trachea and the large bronchi. Amid this foam were numerous white threadlike parasites, 4 to 8 cm long, consistent with *Dictyocaulus viviparus* (Fig.4).

ANSWERS

Follow-up Questions:

- *Morphologic diagnosis*
- *Etiologic diagnosis*
- *Diferencial diagnoses*

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Figure 1. Ten-month-old calf that died in an outbreak of respiratory disease. It had a rough hair coat and was in poor body condition.



Figure 2. Calf. The lung was heavy, red, and firm, with multifocal raised white emphysematous areas in the caudal lobes (arrows) and red consolidated areas in the cranial lobes.



Figure 3. Calf. Lung. Cut surface. Consolidated pneumonic red areas (P) alternated with normal areas (N). There was also marked interstitial edema (*).

Microscopic Description:

Numerous adult nematodes consistent with *D. viviparus* are inside the bronchi (Fig. 5). These nematodes measure 350 µm in diameter, have a smooth eosinophilic cuticle of approximately 5 µm, coelomyarian musculature, lateral cords and pseudocoelom. The reproductive organ contains eggs with embryos and larvae, and a multinucleated intestine. Within the lumen of the bronchioles, there is extensive inflammatory infiltrate consisting of eosinophils and neutrophils mixed with mucus, sometimes surrounding nematode larvae (Fig. 6). In some regions of the lung, the eosinophilic infiltrate



Figure 4. Calf. Bifurcation of the trachea into the major bronchi. Numerous white threadlike 4- to 8-cm long nematodes (consistent with *Dictyocaulus viviparus*) amidst abundant white foam.

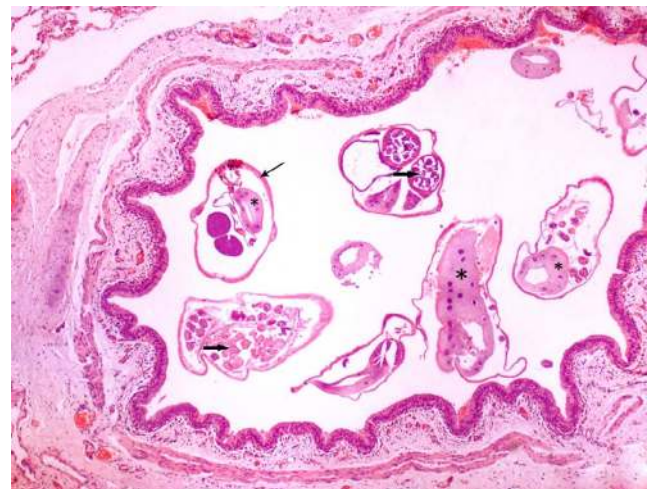


Figure 5. Calf. Lung. A bronchus contains multiple adult nematodes consistent with *Dictyocaulus viviparus*. Parasite histological sections measure 350 µm in diameter, have a smooth eosinophilic cuticle of approximately 5 µm, platymyarian musculature (thin arrow), and pseudocoelom. A reproductive organ contains eggs with embryos and larvae (thick arrow), and a multinucleated intestine (asterisks).

completely obstructs the lumen of the bronchioles. Within the alveoli, there is a large amount of pink and homogeneous material (edema fluid), intact and degenerated neutrophils and eosinophils, as well as foamy macrophages. Eggs and larvae of *D. viviparus* are inside alveoli, surrounded by marked

inflammatory infiltration of eosinophils and a few multinucleated giant cells (Fig. 7)

- *Morphologic diagnosis: Acute to subacute, disseminated, severe bronchopneumonia*
- *Etiologic diagnosis: Verminous bronchopneumonia*
- *Differential diagnoses: Interstitial and bronchopneumonias of cattle.*

Comments:

Dictyocaulus viviparus (Table 1) is the only adult nematode that infects the lungs of cattle, although it can also be infected by ascarid larvae. *D. viviparus* causes severe, sometimes fatal, respiratory disease, known as parasitic chertitis or verminous pneumonia (3,4). Although *D. viviparus*

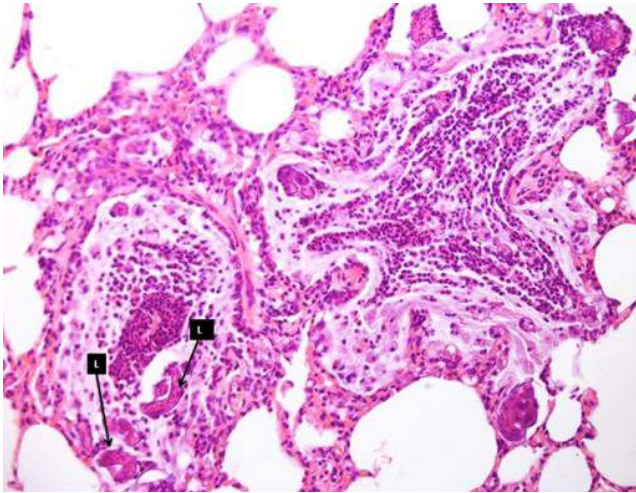


Figure 6. Calf. Lung. A bronchiole is filled with inflammatory exudate (neutrophils and eosinophils) and mucus. Note the nematode larvae (L) amidst the bronchiolar exudate.

Table 1. Taxonomy of *Dictyocaulus viviparus* (8)

Phylum	Nemathelminthes
Class	Nematoda
Order	Strongylida
Superfamily	Trichostrongyloidea
Family	Dictyocaulidae

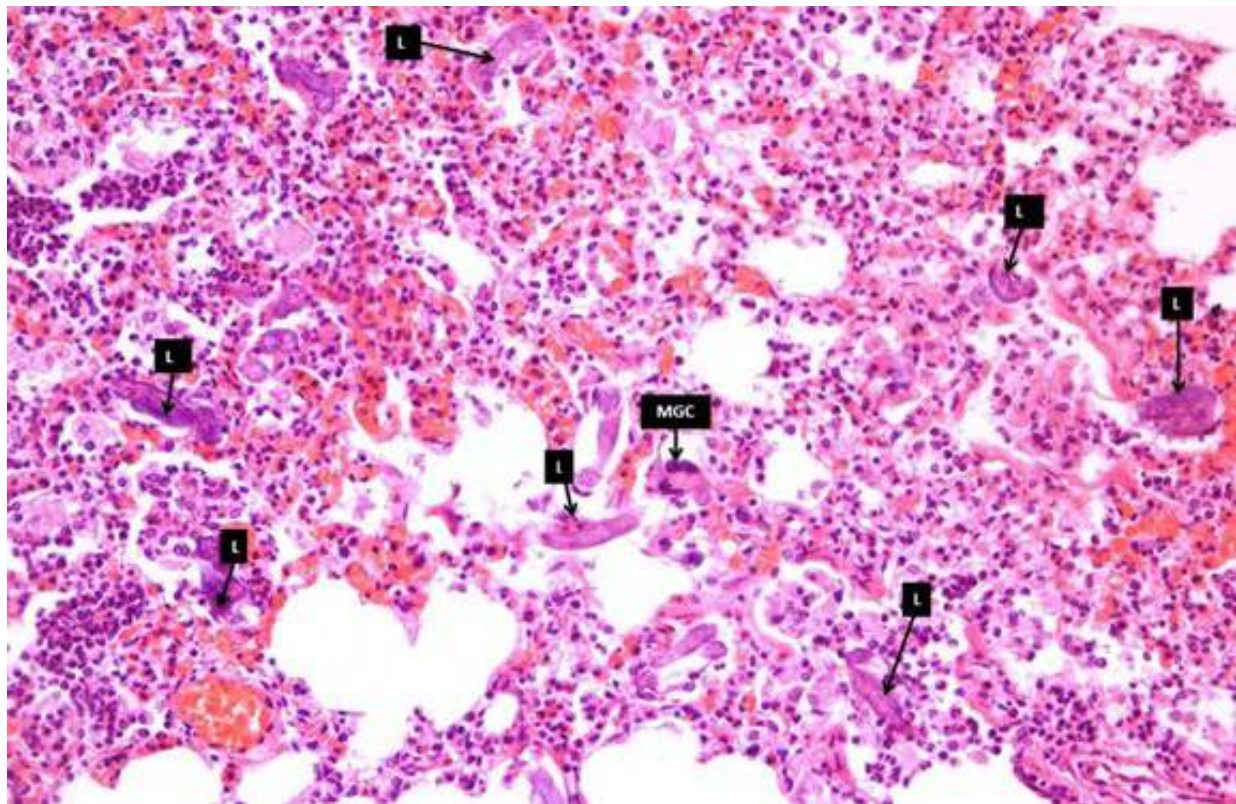


Figure 7. Calf. Lung. A bronchiole is filled with exudate, consisting of mucus, eosinophils and neutrophils. There is proliferation of alveolar macrophages within alveoli, occasionally forming multinuclear giant cells (MGC). Numerous *D. viviparus* larvae (L) are within the alveolar exudate.

is a cosmopolitan parasite capable of infecting cattle of any age, parasitic bronchitis and pneumonia typically occur as primary infection in immature (12 months old or younger) dairy calves during their first season at pasture (1), as well as in cattle grazing in colder climate or during colder months of the year. In the southern hemisphere, cases mainly arise during the autumn and winter but can also occur in the spring.

The life cycle of *D. viviparus* (Fig.8) is as follows: Adult lungworms reside and lay eggs in the bronchi and bronchioles of the bovine host. Males measure approximately 4.0-5.5 cm, while females are 6-8 cm long. (1). The females are ovo-viviparous and lay embryonated eggs, which hatch almost immediately, liberating L1 larvae. These L1 larvae are coughed up, swallowed, and shed in the feces (2). In the external environment, the larvae reach the infective L3 stage within five days or longer, depending on the climate conditions (moisture and temperature) (8). To leave the fecal pat and reach the herbage, the L3 larvae rely on their motility or



Figure 9. Fungus *Pilobolus* sp. on manure showing ripe, black sporangia on top of a fluid filled vesicle, which provides the propelling force when it bursts, supported by a sporangiophore (stalk). From Forbes A. Lungworm in cattle: epidemiology, pathology and immunobiology. *Livestock*; 2018; 23(2). Reproduced with permission.

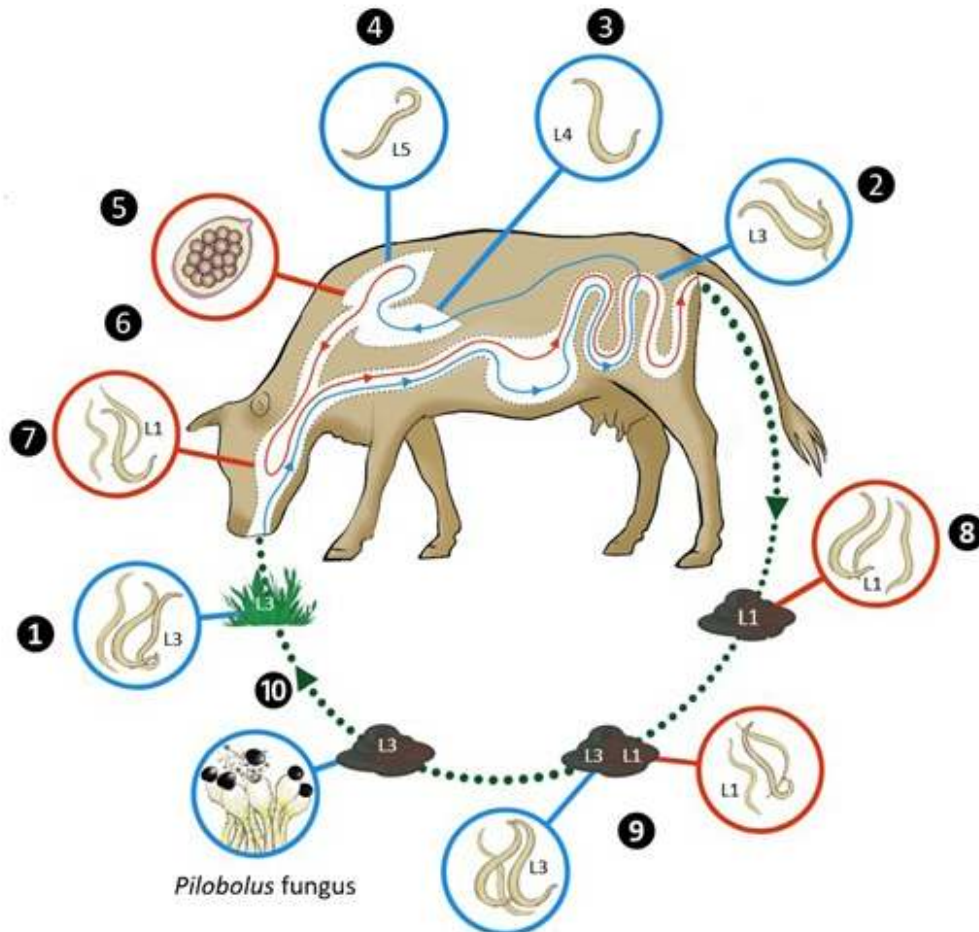


Figure 8. The life cycle of *Dictyocaulus viviparus*. 1. Cattle ingest the L3 larvae with the pasture; 2. L3 larvae cross the intestinal wall and reach the mesenteric lymph nodes, where they molt to L4; 3. L4 larvae reach the pulmonary alveoli through the blood or lymphatic circulation; 4. L4 undergo the final molt to L5 in the bronchioles; 5. Young adults ascend through the bronchi, mature into adults, and produce eggs; 6. The eggs hatch almost immediately, releasing L1 larvae; 7. L1 larvae migrate to the trachea and are swallowed; 8. L1 larvae are shed in the feces 9. L1 to L3 larvae develop in manure; 10. L3 larvae move through the pasture on their own or with the help of the *Pilobolus* fungus (Drawing by Mario Assis Neto, Courtesy Prof. Claudio Barros)

the help of the fungus *Pilobolus* (3) (Fig.9). The larvae climb onto the sporangium of the fungus, which fills with water and bursts, propelling the spore and the lungworm larvae for distances of up to 3 meters (3). Larvae survive best in cold, moist environments, particularly when long herbage or free water stabilizes the microenvironment. Under optimal conditions, larvae can persist in the pasture for more than a year (2). When ingested by a suitable host, the L3 larvae penetrate the intestinal mucosa and reach the mesenteric lymph nodes, where they molt into L4 larvae (1). These larvae then travel via the lymphatic or blood circulation to the lungs and pass through the lung capillaries into the alveoli approximately one week after infection (8). The final molt takes place in the bronchioles a few days later, after which the young adults move up the bronchi and mature. The prepatent period is around 3-4 weeks (1,8). During this period, the parasites undergo remarkably rapid growth, reaching the lungs with a few millimeters in length and developing into adult worms that measure up to 8 cm long in 3-4 weeks. The cycle begins anew when L1 larvae and eggs are excreted onto pasture, hatch, develop into infective larval stages (L3), and are ingested by another host.

There are two ways in which *D. viviparus* infection can persist from one year to the next on farms (3): (i) in pastures (vegetation and soil) as L3 larvae and (ii) in animals carrying the inhibited L4 larval stage, which develops into adult lungworms in the spring. If in the spring cattle are released onto a pasture grazed by infected cattle during the previous year, they may become infected by ingesting L3 larvae. Alternatively, they can become infected if they graze alongside carrier cattle and acquire larvae deposited in the herbage after activation of inhibited L4, which develop into egg-laying adults (5).

The clinical and pathologic manifestations of dictyocaulosis depend on the stage of infection, the host's immunity level, and the number of invading larvae (6). The different

phases of *D. viviparus* infection are known as penetration, prepatent, patent, and postpatent (1,2,8,9). Their main characteristics are summarized in Table 2. The case presented here fits into the category of patent phase.

Ultrastructural investigation of the lungs of cattle experimentally infected with *D. viviparus* by transmission and scanning electron microscopy (7) show substantial loss of ciliated epithelial cells in the bronchi, severely affecting mucociliary clearance. Bronchiolitis, peribronchiolitis, and cell-rich mucus produce bronchial obstruction and consolidation of the associated alveoli. Inflammatory cells thicken the alveolar walls and there is an increase in connective tissue. The flat type I pneumocytes are damaged and replaced by cuboidal type II pneumocytes. This latter change, often described in the past as "epithelialization" (4), inhibits the gas exchange between blood and alveoli.

In bovine dictyocaulosis, bronchial ciliated epithelial cells become detached and are replaced by microvilli-bearing undifferentiated cells (7), making the host more prone to other pathogens in the airways. Consequently, calves infected with *D. viviparus* are more susceptible to infection by respiratory syncytial virus (RSV) and bovine herpesvirus type 1 (BoHV-1) infection (3).

Another clinical manifestation, known as reinfection syndrome, occurs when partially immune adult cattle on endemically infected farms have access to pastures contaminated with large numbers of infective larvae. In these herds, there is a high prevalence of coughing, tachypnea, and lethargy approximately 2 weeks after exposure to the parasite. Some animals may also develop fatal dyspnea without exhibiting coughing (1).

The clinical diagnosis of dictyocaulosis (6) can be established based on respiratory signs and epidemiological data such as time of the year, age of the animals, temperatures, and rainfall. Laboratory diagnosis relies on detecting larvae in the feces using the Baermann technique, which determines

Table 2. Chronology of clinical signs and pathology of the various phases of *Dictyocaulus viviparus* infection (1).

Phase/Duration	Clinical signs	Pathology
Penetration/days 1-7 PI	Not seen	Not significant; no adult worms or larvae in airways
Prepatent/days 7-25 PI	Cough; increased breathing rate; death is infrequent unless secondary disease occurs	Pulmonary edema; hyaline membranes; alveolar epithelial hyperplasia; interstitial emphysema. No adult worms but possibly larvae in airways.
Patent/days 25-55 PI	Coughing; increased breathing rate; labored breathing; decreased feed/water intake; weight loss; deaths frequent, unless treated	Bronchitis and bronchiolitis; pneumonia with consolidation and collapse of lung lobes; secondary bacterial pneumonia in some cases. Pulmonary edema; hyaline membranes; alveolar epithelial hyperplasia ("alveolar epithelialization"); interstitial emphysema. Many adult worms, larvae, and eggs in airways.
Postpatent/days 55-90 PI	Gradual amelioration of clinical signs seen in the patent phase; death due to acute complicating lesions, such as secondary bacterial infection, pulmonary edema, and emphysema, may occur	Gradual resolution of lesions of the patent phase. Permanent lung injury; bronchiolitis obliterans; bronchiectasis; chronic bronchopneumonia in a few cases. Few or no adult worms in the airways; no larvae in the airways.

PI= postinfection

the number of larvae per gram of feces. Necropsy is indicated when there are several animals with clinical signs in the herd and no larvae are found in the feces. In such cases, immature parasites can be observed in the bronchial tree (6).

Differential diagnoses should include bronchopneumonias from Bovine Respiratory Disease syndrome (BRD) caused by bacteria such as *Mannheimia haemolytica* and viruses such as bovine syncytial virus and parainfluenza virus 3.

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