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## The Peculiarities of Donkey Respiratory Disease (14-nov.-2001)

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### Summary

Donkeys belong to the Equidae family and as such are similar to equines in many ways. However, there are a number of significant features peculiar to the donkey that practitioners should be aware of when attempting to investigate and treat respiratory disease. These include differences in anatomy, physiology, pharmacokinetics, disease incidence and morbidity with certain infectious diseases. This chapter focuses on the differences as described in the published literature and recorded from cases at The Donkey Sanctuary (Click here to visit the website) in the UK.

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### Introduction

The donkey (*Equus asinus*), or ass, is a member of the Equidae family, a family that also includes the modern horse (*Equus caballus*) and zebra (*Equus zebra*). Hence asinines (donkeys) share many features with other Equidae, such as equines (horses). The genetic closeness of the Equidae family is reflected in the ability to produce viable (non-fertile) offspring such as the mule, hinny and zebrokey from matings between donkeys, horses and zebras. The respiratory system of the donkey is in most respects similar to that of any small horse. There are, however, significant differences of behaviour, physiology and management, which influence the presentation, incidence, and treatment of many aspects of disease. The lack of clinical information on the donkey often means it is treated as if it were a small horse.

Worldwide there are an estimated 44 million donkeys [1]. Of these 96% are found in developing countries contributing significantly to the rural economy. While in Europe the absolute numbers of donkeys are declining, their importance as companion animals is increasing. Similarly in USA the use of donkeys, miniature donkeys and mules as companion animals is well established. Thus there is a need for equine practitioners to treat donkey patients in an informed and sensitive manner. Only those conditions with significant differences from the horse are described in this chapter. The chapter uses information derived from the records kept at the Donkey Sanctuary (Fig. 1) and from literature reviews. There is comparatively little objective data in the literature, reflecting the relative lack of interest in this equid until recently. The longevity of the companion donkey in the UK results in a disproportionate amount of geriatric respiratory disease being diagnosed at The Donkey Sanctuary. Hence dental sinus empyema, chronic fibrosing lung disease, and neoplasia are some of the most frequently encountered respiratory conditions at the Donkey Sanctuary. Where groups of younger animals are encountered it is not unreasonable to expect a higher incidence of infectious respiratory disease. There is a lack of information on performance related respiratory disease in the donkey such as exercise induced pulmonary haemorrhage (EIPH); soft palate

dislocation, laryngeal hemiplegia and these conditions will not be discussed.

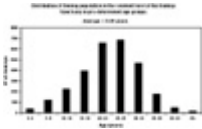


Figure 1. Graph showing the distribution of ages of donkeys at the Donkey Sanctuary. - To view this image in full size go to the IVIS website at [www.ivis.org](http://www.ivis.org) . -

<b>Table 1. Causes of Death or Euthanasia due to Primary Respiratory Conditions Between 1997 - 2001</b>	
Pulmonary Fibrosis	49 (38%)
Chronic Respiratory Disease (no post-mortem)	23 (18%)
Pneumonia	8 (6%)
Chronic Respiratory Failure	5 (4%)
Cor Pulmonale Secondary to Chronic Respiratory Disease	4 (3%)
Pulmonary Neoplasia (primary or secondary)	4 (3%)
Tracheal Stenosis	4 (3%)
Guttural Pouch Empyema	4 (3%)
Chronic Bronchiolitis ("COPD")*	4 (3%)
Pulmonary Oedema	3 (2%)
Pulmonary Haemorrhage	3 (2%)
Chronic Sinusitis	2
Hydatid Cysts	2
Fracture Ribs	1
Pulmonary Abscessation	1
Pulmonary Thromboembolism	1
Ethmoid Haematoma	1
Ruptured Diaphragm	1
Pulmonary Congestion	1
<b>Total Number of Cases</b>	<b>129</b>

\* Now known more correctly as RAO (recurrent airway obstruction).

### **General Considerations for the Clinician**

Presentation - Many UK donkeys are unaccustomed to routine handling or exercise. They are also stoical and un-dramatic in their display of clinical signs. Unlike the horse, coughing with chronic respiratory disease is rarely heard, and is an inconsistent finding with acute respiratory disease. Therefore, respiratory disease is often presented in an advanced and severe form.

Clinical Examination - A full and thorough clinical examination is recommended for any donkey presenting with respiratory signs for a number of reasons. Firstly, chronic respiratory disease is not an uncommon incidental finding in many donkeys in

the UK, as respiratory function is not always tested with regular exercise. Secondly, many systemic or abdominal disorders can produce respiratory signs. For example marked dyspnoea and pyrexia can accompany hyperlipaemia, cyathostomosis and acute pancreatitis. Thirdly, many conditions in the donkey have presenting signs very different from the equivalent conditions in the horse. For example colic may present as dullness, depression and anorexia without rolling, pawing or sweating as commonly observed in the horse. Diarrhoea is often seen only in the most severe or advanced cases of gastrointestinal disease.

Due to the smaller size of the donkey thorax and reduced attenuation of lung sounds through the thoracic wall, the lung sounds are generally more audible in the donkey as compared with the horse. However, in obese donkeys, respiratory sounds are often muffled by a thick layer of subcutaneous fat. Even after dieting, substantial deposits of adipose tissue can remain along the neck and dorsal thorax. The obese donkey is usually unfit and almost any exercise can provoke an alarming degree of tachypnoea. The use of a re-breathing bag will help accentuate subtle sounds.

**Complications** - Any respiratory condition, especially if accompanied by anorexia, can precipitate life-threatening hyperlipaemia and gastric ulceration. Prognosis for advanced or untreated hyperlipaemia is very poor. Therefore, the clinician must be able to diagnose and treat hyperlipaemia to ensure successful treatment of the primary condition [2]. As the stress of hospitalisation and separation from companions can also precipitate a hyperlipaemic crisis, particularly in obese animals, all investigations are best performed in a home environment and using mild sedation as necessary. Post-mortems performed at the Donkey Sanctuary have revealed a high incidence of gastric ulceration, especially in anorexic patients (N.J. Bell unpublished data). Therefore anti-ulcer therapy could be instigated in such cases.

### **Anatomy of the Respiratory Tract**

In contrast to the horse, there are few reports describing the anatomy of the donkey respiratory tract [3-9]. The differences that have been described are subtle and probably of minor clinical significance.

**External Nares** - The external nares and false nostrils closely resemble those of the horse. One notable difference is the position of the nasolacrimal duct opening (Fig. 2). This is positioned dorsally in the false nostril approximately 10 mm from the mucocutaneous junction. The site is usually made conspicuous by the lack of pigment visible within the opening. While it is usually possible to pass a standard equine nasolacrimal cannula into the canal for flushing or administering medication, in some donkeys the ostium is particularly small and a fine gauge cannula is required.



Figure 2. Photograph demonstrating the position of the nasolacrimal duct in the donkey. - To view this image in full size go to the IVIS website at [www.ivis.org](http://www.ivis.org) . -

**Nasal Passages** - The nasal passages are narrow compared with those of a horse and so extra care must be taken during endoscopy and stomach tubing. At the Donkey Sanctuary a 9.5 or 13 mm outer diameter stomach tube, and a 9.8 mm outer diameter endoscope are used.

**Pharynx** - Lindsay and others [7] described in detail the nasopharyngeal region of 10 donkeys, and drew comparisons with horse and pony specimens. In the donkeys the pharyngeal airway was more constricted in its middle, and more flared dorsally and ventrally than in the horse (Fig. 3). The pharyngeal recess is a diverticulum of the pharyngeal mucosa and lies between the guttural pouch openings. In the donkey it forms a deep membranous pouch 2 - 3 cm in diameter extending 4 - 6 cm caudally from a constricted orifice, through which it communicates through the nasopharynx. The mucosa of the pharyngeal recess bulges into the airway during the high-pitched inspiratory part of the bray, and may play a role in vocalisation.



Figure 3. Photograph of a sectioned donkey head (midline sagittal section) demonstrating the pharyngeal anatomy. - To view this image in full size go to the IVIS website at [www.ivis.org](http://www.ivis.org) . -

**Guttural Pouches** - The position of the guttural pouch flaps was described by Fores et al., [10] as being more horizontally placed than in the horse. At the Donkey Sanctuary we have not encountered difficulty catheterising the guttural pouches. The pouches resemble those in the horse with differences in shape being of minor significance. However, surgical access to the

pouches can be difficult given the close proximity of the surgical landmarks in the smaller head.

Larynx - The angulation of the *aditus laryngis* (laryngeal opening) in the donkey was found to be tilted caudally, and the apex of the epiglottis was pulled nearer to the arytenoid cartilages. This angulation can make it hard to observe the trachea through the *rima glottidis* (opening through the vocal folds) from within the pharynx with an endoscope. Radiographic studies confirmed that the air passageway from the nasopharynx through the larynx was more angulated in the donkey. The donkey epiglottis was reported to be shorter and more sharply pointed than that of the horse. The *aditus laryngis* in the donkey has been described as having a transverse diameter greater than its dorsoventral diameter, in comparison to the more oval, elongated opening in the horse.

The laryngeal anatomy of the donkey is described in detail by Lindsay and Clayton [7]. The lateral ventricle (the pouch between the vocal and vestibular folds) is a broad shallow depression in the donkey and the opening cannot be visualised clearly endoscopically. In the donkey a median ventricle was found to be expanded into bilateral pouches. The donkey has large laryngeal saccules (Fig. 4), the mucosal pouches interposed between the thyroarytenoideus muscle and the thyroid cartilage. The saccules open into the laryngeal cavity through small circular orifices near the rostroventral extremity of the vestibular folds. The saccule is lined with mucus secreting respiratory epithelium. The large thyroarytenoideus muscle is divided into vestibular and vocal parts and these are thought to control secretion from the laryngeal saccule.



Figure 4. Photograph demonstrating the laryngeal saccule. - To view this image in full size go to the IVIS website at [www.ivis.org](http://www.ivis.org) . -

The functions of the laryngeal ventricles and saccules are unknown but may play a role in the characteristic vocalisation of the donkey. The bray of the donkey is a sound that communicates over long distances, and is unusual as it consists of both an inspiratory and an expiratory component (most vocalisations are expiratory only). The resonance of the bray is related to the airway anatomy, in particular the pronounced mid nasopharyngeal constriction, the pharyngeal recess and laryngeal saccules.

Trachea - Matthews and others [11] felt that the airway of the donkey was narrower than in a comparably sized pony. At the Donkey Sanctuary a 21.6 mm (size 16) or a 18.6 mm (size 14) endotracheal tube are used for intubation.

Lung and Mediastinum - The pattern of bronchial tree division and lack of external lobe divisions resembles that of the horse. The mediastinum caudal to the heart is thin and easily perforated like that of the horse. Fenestrations are difficult to identify; although addition of fluid to one hemithorax has been shown to enter the opposite hemithorax clearly demonstrating a communication exists or easily develops.

### **Pharmacology**

There are a number of important differences in donkey pharmacokinetics as compared with the horse to be considered with any medical treatment or anaesthetic. Some drugs are metabolised more rapidly, notably phenylbutazone [12], and should be administered more frequently to achieve therapeutic levels in the donkey. At the Donkey Sanctuary long-term doses of 2.2 mg/kg phenylbutazone per donkey twice daily are frequently used without ill effect. With regard to anaesthesia, a number of drugs, notably ketamine, guaphenesin, and etorphine, show differences in metabolism and distribution. When considering drug dosages for geriatric patients, as with any species, it is good practice to adjust drug dosages according to hepatic and renal function as indicated by laboratory results. Mathews and others [11] published the most recent review article on donkey pharmacokinetics.

### **Physiological Pulmonary Function Peculiarities**

The most recent paper to study the physiological differences of cardiorespiratory function in the donkey is by Delvaux and others [13]. These works compared the results of a number of tests in donkeys to those in horses and their findings may be summarised as follows:

1. Haematological parameters were similar to those ascertained by French et al., (Appendix 1).
2. Arterial blood gas measurement did not differ from those obtained in the horse.
3. Anatomical differences found by endoscopy of the larynx were equivalent to those described previously.

In addition partial collapse of the pharynx was observed in 43% of cases. The donkeys exhibited forceful bouts of

coughing during the introduction of the endoscope through the larynx. To minimise this, xylocaine was used to desensitise the larynx. (At the Donkey Sanctuary we have also found xylocaine useful to permit the advancement of the endoscope into the trachea).

4. Tracheal wash cytology in the donkey showed some differences from the horse. There was an increased percentage of epithelial cells, neutrophils and eosinophils in the donkey and a reduction in the percentage of macrophages.
5. ECGs and phonocardiographs were obtained and the donkey's heart rates (49 - 59±12) were found to be slightly elevated in comparison to the horse [35-40].  
In addition only 39% of the donkeys had a detectable arrhythmia associated with changes in vagal tone during respiration. This is less frequent than in horses at rest. On the other hand no second degree A-V blocks were observed. It is considered that these findings may be related to a lower resting vagal tone in the donkey.
6. Echocardiography was found to be easy to perform in a donkey, which readily stood still without sedation. The measurements obtained of heart size and chamber diameter were comparable to those found in a small pony of similar size. This reflects that the size of the heart is proportional to the size of the animal.
7. Thoracic radiography (using exposures of 67 - 75 KV, +13 - 22.5 mA) demonstrated that a donkeys lungs were similar in appearance radiographically to those of ponies or foals. It was noticeable that a higher percentage of donkeys showed spondylosis of the vertebrae.
8. The parameters of mechanical ventilation were measured in 10 donkeys, using a facemask for airflow output and an oesophageal catheter for pleural pressure measurements. (A number of donkeys found the test stressful and were dropped from the study). In general, donkeys showed results more similar to that of farm animals, such as bovines, than horses. Compared with horses, donkeys had: increased respiratory frequency; increased total pulmonary resistance; increased variation in maximal pleural pressure; increased specific volume of air ventilated per minute and a reduced dynamic compliance. It is possible that the laryngeal conformation contributes in part to the increased total pulmonary resistance the backwards pointing larynx represents a bottleneck obstruction to the passage of air. The writers hypothesize that the differences in the mechanical ventilation parameters measured reflected differences in reaction to danger; donkeys tend to stand and confront danger while horses tend to demonstrate a flight response.

## **Surgery**

Devoicing - The authors should emphasise that they do not believe devoicing to be an ethically acceptable procedure in the donkey or mule. However, the operation of muting or devoicing donkeys and mules was performed on large numbers of animals during wartime, when it was felt that the penetrating bray would betray troops. Today it may be requested by pet owners concerned about perceived nuisance.

The operation is described in precise detail by Stewart [14] and Phillip [15]. For anyone interested in veterinary history the articles make compulsive reading. The operations were performed using chloroform anaesthetic and casting the animal. The technique involved removal of the vocal cord and the complications included haemorrhage and post-operative stenosis requiring tracheotomy. The wounds were left to heal by second intention and the animals were fit for full work in 3 weeks. In the donkey and mule the different shape of the ventricle (as discussed under anatomy) made the operation more difficult. For further information the authors advise referral to the papers by Stewart [14] and Phillip [15].

In the USA devoicing has been tried using electrocautery via the nostrils. The partial healing of the arytenoids results in reduced airway diameter. Removal of vocal folds and ventricles using an endoscopically guided laser is the preferred method currently used in horses (N. Matthews, personal communication).

## **Respiratory Conditions**

### **Infectious Respiratory Disease**

The donkey is reported as having a different susceptibility to a number of diseases as compared with the horse, some of which are endemic throughout the world e.g., African Horse Sickness, Equine Viral Arteritis, Equine Infectious Anaemia and Glanders [16-19]. Of these, Herpes infection, Influenza, and *Dictyocaulus arnfieldii*, where the clinical signs relate to the respiratory tract, will be mentioned.

The authors are unaware of any reports describing any significant differences in the clinical features of other respiratory conditions, such as Strangles or Equine viral arteritis, in the donkey as compared with the horse. These conditions and others will be reviewed as more literature becomes available on the subject.

Herpesvirus - It appears that donkeys exposed to equine herpesvirus 1 (EHV-1) and 4 show clinical signs similar to those seen in the horse, and similar precautions should be taken in the event of a suspected outbreak [20,21]. In one experimental study EHV-1 infection caused abortion in a pregnant donkey mare, with characteristic EHV-1 lesions in the foal [22]. In another study where local donkeys were infected with EHV-1, results indicated that the donkeys were less susceptible to EHV than horses, [23]. The abortion strain of EHV-1 has been recovered from a donkey with respiratory diseases [24].

As well as equine herpesvirus, donkeys are known to suffer from three other herpesviruses named Asinine herpesvirus 1, 2 and 3 (AHV). The herpesvirus isolated from coital exanthema lesions in a donkey were found to differ from EHV-1, 2 and 3 and were temporarily referred to as EHV-5 or AHV-1 [25]. Two distinct herpesviruses were isolated from donkeys, firstly from circulating leucocytes and secondly from the nasopharynx after steroid administration, and these were named AHV-2 and 3 [26]. There is more published information on the nucleotide sequences of asinine herpesvirus, as compared with equine herpesvirus [26-28], but less on the clinical signs relating to AHV in the donkey, although AHV is known to be pathogenic in donkeys [29-30]. From nucleotide sequencing studies it has been deduced that asinine herpesvirus 3 is closely related to equine herpesvirus 1, and may be the progenitor of EHV-1, which has become host adapted to the horse [27,28,31].

At the Donkey Sanctuary there has been one putative outbreak of AHV infection, with 18 donkeys affected. AHV-3 was isolated from a nasal swab of a donkey with ataxia, anorexia and unilateral facial paralysis. Six donkeys died after showing signs of weakness, anorexia and pyrexia. Twelve donkeys survived after showing milder clinical signs and some serous nasal discharge. Profound leucopaenia was apparent in six animals. Virus neutralizing antibodies to EHV were raised in most animals. Treatment was symptomatic and supportive.

Serological diagnosis of herpesvirus infection (Personal communication, Richard Newton) - The complement fixation test (CFT) for EHV-1, EHV-4, Equine rhinovirus 1 and Equine rhinovirus 2 is, in some cases, complicated by anti-complement activity. The presence of a non-specific, and as yet unidentified, anti-complement factor in the serum produces a positive result even though virus induced complement-fixing antibody is not present. False positives due to anti-complement activity are detected by routinely running the CFT without the addition of antigen. Without antigen the virus induced antibody should not fix complement. Hence a positive control result clearly indicates the presence of another agent that fixes complement and the serum sample is said to have anti-complement activity.

Anti-complement activity has been reported occasionally in horses but is encountered in many of donkey serum samples submitted to the Animal Health Trust, Newmarket. A virus neutralisation test (VN test) is an alternative means of detecting viral antibody titres. The VN test has an important limitation in that virus neutralising antibodies have a much longer half-life than complement fixing antibodies. The longer half-life means antibody levels change more slowly, with background antibody titres making recent rises more difficult to detect. As the VN test is considered inferior to the CFT, it has become obsolete in many laboratories. However, when serum samples have demonstrated anti-complement activity the VN test may be the only means at present of confirming a diagnosis. In this case it may be necessary to contact your laboratory before submitting donkey serum samples for the VN test so that the test can be ordered and prepared. In the future ELISA may be available as an alternative test and may eventually supersede the CFT.

Influenza - Equine influenza in donkeys has been reported in the literature since as early as 1874 [32]. The clinical signs are similar to those seen in the horse with pyrexia, cough, nasal discharge, depression and inappetance being the significant findings. However, in a number of experimental trials and natural outbreaks [33-36] the mortality has been higher and the clinical signs more pronounced in the donkey. The infected donkeys were found to be more likely to develop secondary bacterial bronchopneumonia. Rose and others [36] commented on an outbreak of influenza in a donkey stud and noted that the high mortality may have been attributable to concurrent lungworm infection, although the severity of the disease in an individual was not directly related to the size of the lungworm burden.

If nasopharyngeal swabbing is to be attempted, the smallest foal size swab should be used. In most cases soaking in sterile saline is sufficient lubrication, provided the swab is small.

Due to the higher morbidity and mortality the importance of effective vaccination protocols against influenza in donkeys should be emphasized [37], and steps should be taken to treat any concurrent lungworm infections. It is also recommended that donkeys that contract influenza are treated immediately with antibiotics given the high risk of secondary bacterial bronchopneumonia developing following infection. As anorexia is often a feature of influenza, serum triglycerides should be monitored and hyperlipaemia prevented or treated appropriately.

*Dictyocaulus arnfieldi* (lungworm) - The nematode *Dictyocaulus arnfieldii* is a parasite well adapted to living in the donkey, where it is able to grow to maturity and produce eggs [38-40]. In most instances large lungworm burdens will cause few clinical signs in the donkey. However, the pulmonary changes secondary to lungworm may exacerbate other respiratory diseases, such as influenza [36].

The life cycle involves ingestion of third stage larvae that migrate from the gastrointestinal tract to the lungs via the blood and lymphatics. Larvae infiltrate the alveoli causing a low-grade inflammation. Adult worms grow up to 8 cm within the bronchi and can cause blockage of smaller airways [41]. The eggs produced by adults in the airways are coughed up, swallowed and passed out in the faeces. The prepatent period is a minimum of two months.

Diagnosis of the parasite in the donkey relies upon finding larvae in faeces using the modified Baermann technique (Appendix 2). By contrast infection in the horse stimulates severe tissue reaction and mature egg laying adults are rarely found. In the horse lungworm infection is diagnosed on the basis of a history of chronic coughing and co-grazing with an

infected donkey. Bronchioalveolar lavage or tracheal wash may retrieve numerous eosinophils and larval stages, [42]. A response to therapy will also confirm the diagnosis.

The treatment of choice is a single dose of ivermectin at 200 µg/kg per os [43]. Recently moxidectin has been shown to have efficacy against lungworm at 0.4 mg/kg [44]. Less effective options include mebendazole 15 - 20 mg/kg SID for five days [45], two doses of thiabendazole at 440 mg/kg with a one-day interval between doses or febendazole at 15 mg/kg [46]. The parasite can be hard to eliminate from paddocks due to prolonged survival in cool, moist conditions. Pasture management involving harrowing, ploughing or crop rotation may reduce numbers of larvae. The larvae will not survive severe frost. However once eradicated, the long prepatent period of lungworm means reinfestation is slow, especially if ivermectin or moxidectin is being used as anthelmintic for the control of the intestinal nematodes.

### **Conditions of the Upper Respiratory Tract**

In the donkey population at the Donkey Sanctuary the conditions of the upper respiratory tract encountered include false nostril atheroma, ethmoid haematomas, sinus disease, palate ulceration/penetration/oedema secondary to dental overgrowth, laryngeal paralysis secondary to hepatic failure, guttural pouch empyema/chondroids and tracheal stenosis. There is a lack of information about exercise and performance related upper respiratory tract conditions e.g., soft palate dislocation due to the non-athletic nature of donkeys. Tracheal and sinus diseases are discussed in detail below. Standard diagnostic and therapeutic considerations apply to the other conditions mentioned.

Tracheal Disease - As in the horse, clinical tracheal disease is uncommon in the donkey. Obstruction may be caused by collapse of the tracheal rings, compression from external masses or abscesses, or intramural lesions.

Collapsed Trachea (Tracheal Stenosis) - Degenerative lesions of the trachea with dorso-ventral flattening of the tracheal lumen may be seen in aged animals. In a case reported by Mair and Lane [47] a donkey demonstrated sudden onset respiratory distress with severe inspiratory and expiratory dyspnoea. Vibrations were palpable over the ventral cervical trachea. Endoscopy revealed a dorso-ventral tracheal collapse at the thoracic inlet. Radiography confirmed airway narrowing and calcification of tracheal cartilage rings. The donkey was euthanased.

Presentation and Clinical Signs - Tracheal stenosis may go undetected for many years. Presentation is variable depending on the inciting cause. In the uncomplicated and progressive cases a chronic coughing or pronounced "honking" respiratory sound may be the alerting sign. In cases with concurrent lower respiratory tract disease, stertor with audible tracheal fluid may first alert owners to a respiratory tract problem. However in the acute crisis stertor will be loud, the head may be low to the ground, nostrils flared often with mouth breathing. Once mouth breathing is observed the condition is seldom reversible and deterioration is rapid. Autopsies on these cases have demonstrated almost complete occlusion of the tracheal lumen due to tracheal collapse and tracheal mucosal thickening.

Epidemiology - Weakness and collapse of the cartilaginous rings in the trachea is a common incidental finding at post-mortem. Histopathology has revealed mineralisation and osseous metaplasia of the cartilage in the rings. This is likely to be an age related degenerative change, more pronounced in some individuals. The mineralisation presumably alters the properties of the cartilage, resulting in the observed increase in cartilage ring brittleness, reduced cartilage elasticity and a predisposition for ring collapse, either with trauma or with extreme changes of airway pressures, as encountered with dyspnoea due to lower respiratory tract conditions. The extra-thoracic trachea adjacent to the thoracic inlet is the site most predisposed to collapse.

Between 1997 and 2001 there were four cases of tracheal obstruction that led to euthanasia at the Donkey Sanctuary. There was marked dorso-ventral flattening at the thoracic inlet in two cases, at the tracheal bifurcation in one case and in the mid-cervical region in the other case. In one case there was also mineralisation of the tracheal cartilage rings. All cases occurred in aged animals (mean age 33 years). Chronic lung fibrosis causing increased respiratory effort was a significant factor in two cases.

Diagnosis - Diagnosis is based on findings on clinical examination, which include stertor and severe dyspnoea combined with a palpably distorted trachea and tracheal vibration. Harsh tracheal noises are usually audible without need of a stethoscope and are referred to the lungs preventing proper auscultation of the lung fields. Endoscopy and radiography can be used to confirm the examination findings and eliminate other causes of luminal obstruction or upper respiratory noise.

Differential Diagnoses - The differential diagnoses should include dyspnoea due to lower respiratory tract disease (see chronic interstitial pneumonia), tracheal intra or extra luminal masses, pharyngeal or laryngeal paralysis producing stertor, food inhalation and nasal reflux.

Treatment - The affected donkey appears to compensate for the obstruction during normal non-athletic activities. An acute crisis may be provoked following exercise, trauma, stress or dyspnoea due to lower respiratory tract disease.

The acute crisis without obvious lower respiratory tract (LRT) involvement should be managed with intravenous corticosteroids e.g., Dexamethasone 0.1 - 0.2 mg/kg IV to limit tracheal mucosa swelling. For more chronic cases clenbuterol 0.8 µg/kg slow IV or oral 0.8 - 3.2 µg/kg bid is the bronchodilator of choice as atropine can cause colic signs. Frusemide 0.5 -

1 mg/kg IV or IM may help reduce the tracheal mucosal swelling, as well as limit the development of pulmonary oedema that might accompany severe dyspnoea. Placement of a tracheal tube will often fail to by-pass a thoracic inlet stenosis and may contribute to increased mucosal oedema and stenosis. Therefore, this procedure should be performed with caution if at all. If this condition is encountered secondary to lower respiratory tract disease, such as chronic interstitial pneumonia then treatment of the lower respiratory tract disease can be sufficient to reduce the stertor to acceptable levels. Effective treatment of acute crises is often not possible and euthanasia is advisable.

Prevention - Given that tracheal ring disease is likely to be a progressive, age-related degenerative disorder, prevention seems impossible. However, as most affected donkeys appear to compensate until mid-old age efforts should be directed at maintaining healthy lungs and minimising the risk of precipitating an acute crisis. Unexpected stresses and tracheal trauma by feeders with high brisket boards should be avoided. Trauma due to kicks from other horses, donkeys or mules is a theoretical risk that may need to be considered when introducing new animals to a group.

Sinus Disease - Problems associated with the sinus region are found in the donkey as in the horse. Standard diagnostic techniques are appropriate, although with the shorter length of the facial crest, it is appropriate to use a small trephine (10 - 13 mm) for access to the rostral and caudal maxillary sinuses. Primary and secondary sinusitis in the donkey has been recorded [48,49].

Dental disorders account for a large number of secondary sinus empyemas especially in the geriatric population. Many donkeys have had little attention paid to their molar arcades throughout their life and severe abnormalities accumulate in later years. In addition, the limited access afforded in smaller and miniature donkeys can make treatment of dental problems technically challenging. Secondary, sinusitis due to fungal infections, sinus cysts and tumours have all been seen at the Donkey Sanctuary.

In these geriatric patients careful clinical examination combined with blood testing should be undertaken before radical dentistry is performed. Intra-oral extraction is preferred combined with sinus flushing. Surgical repulsion or buccotomy techniques may also be appropriate if intra-oral extraction is not possible.

### **Conditions of the Lower Respiratory Tract and Lung**

In the donkey population at The Donkey Sanctuary the conditions of the lower respiratory tract commonly encountered include idiopathic pulmonary fibrosis and heaves. Heaves or recurrent airway obstruction is often encountered in the live donkey and is managed, as in the horse, with a combination of improved air quality and medication including bronchodilators, mucolytics and inflammatory. Nebulization of inhaled drugs is tolerated well in the donkey and similar dosage regimes described for the horse should be used. Pulmonary neoplasia is seen in the geriatric population and is an important differential diagnosis in the aged, dyspnoeic donkey. Other conditions of the lower respiratory tract seen include hydatid cysts, lungworm, influenza, fractured ribs, thromboembolism, pulmonary haemorrhage, abscessation and oedema. Hydatidosis and idiopathic pulmonary fibrosis are discussed below.

Hydatid Cysts - Hydatid cysts are the metacestode of *Echinococcus granulosum*, the tapeworm of domestic carnivores. Donkeys and other equines are infected when grazing pastures contaminated by the faeces of dogs, foxes, cats etc. The donkey and horse act as an intermediate host in which hydatid cysts form containing fertile protocoelocysts. These are generally found in the liver or lung. The cysts grow slowly and can reach 6 - 7 cm in diameter. Most cysts are asymptomatic and are found at post-mortem examination. However, in large numbers, cysts can affect lung and liver function. Premortem diagnosis can be difficult, ultrasonography providing the most effective method [50].

The incidence of hydatid cysts found at post-mortem examination in donkeys at the Donkey Sanctuary is higher in donkeys that originate from Ireland and Wales, highlighting regional differences in distribution. Control is achieved by treating dogs and cats that could potentially defaecate on the grazing pastures using praziquantel at 0.1 mg/kg. Dogs and cats should be prevented from eating raw offal and their faeces removed from pastures [51].

A donkey at the Donkey Sanctuary that demonstrated marked respiratory distress, weight loss and ventral oedema was found to have numerous hydatid cysts in the lungs and liver. Routine haematology and biochemistry revealed a leucocytosis with neutrophilia; hypoalbuminaemia; and raised total proteins, total globulins, alpha 2 and gamma globulin fractions. Terminal hyperlipaemia developed presumably secondary to the liver involvement.

Treatment of affected cases has rarely been attempted. Drainage of cysts can seed daughter cysts and induce anaphylaxis, although success has been reported in treatment of a horse with an isolated retrobulbar cyst [52]. Surgical excision is rarely an option due to the location of cysts.

The prevalence of hydatidosis in donkeys in Central and Northern Jordan has been evaluated [53,54]. Both surveys found no donkeys less than three years of age were infected. In donkeys over four years 36.8% in Central Jordan and 33.3% of donkeys in Northern Jordan were infected. The number of cysts per donkey increased at an average rate of 0.48 cysts per year. In Central Jordan among infected donkeys 57.1% had hepatic infections, 33.3% were hepatic and pulmonary, and 9.5%

were pulmonary. The majority of cysts (67.7%) were non-fertile, immature, miliary cysts while 10.8% were necrotic and calcified. The volume of cystic fluid ranged from 0.1 to 132 ml.

Idiopathic Pulmonary Fibrosis - IPF synonyms: chronic fibrosing pneumonopathy, chronic fibrosing interstitial pneumonia with alveolitis. The sedentary nature of most donkeys and a reduced tendency to cough as compared with the horse means respiratory conditions such as pneumonia are usually advanced by the time they are presented. Chronic fibrosis of the sub-pleural and interstitial lung tissues is a common finding on post-mortem at the Donkey Sanctuary. In the year 2000, at post-mortem sixteen donkeys were found to have some degree of gross pulmonary fibrosis out of 159 thoracic examinations performed. In many instances, the fibrosis was an incidental finding, unrelated to the primary cause of death. The lesions are likely to be produced by a chronic and progressive interstitial pneumonia, the aetiology as yet undetermined. Interstitial pneumonia of unknown aetiology has been described in horses in the USA [55-57]. Three cases of diffuse alveolar damage, with histopathological changes similar to those seen in the donkey, have also been described [58]. However, the chronic interstitial pneumonia appears to be of much greater significance in the donkey, causing more morbidity and mortality than all the other respiratory conditions seen at the Donkey Sanctuary.

Presentation and Clinical Signs - The presentation of idiopathic pulmonary fibrosis (IPF) is variable depending on the stage at which it has been detected. Earlier cases or mildly affected donkeys tend to demonstrate tachypnoea without a large abdominal lift. Very rarely is a cough reported. However, despite the chronic nature of the condition, most donkeys with chronic interstitial fibrosis present with an acute episode of severe dyspnoea and tachypnoea. Usually there is no previous history of respiratory problems although adventitious noises have been noted as incidental findings prior to the onset of clinical disease.

If there is recovery from an acute episode of dyspnoea then a moderate and progressive degree of dyspnoea tends to persist. In severe cases the inspiratory phase of breathing appears to be lengthened with a marked abdominal lift. The restrictive nature of the pulmonary and subpleural fibrosis presumably limits inspiration while having a much smaller impact on expiration. The inspiratory dyspnoea may wax and wane, but never resolves. The presence of a nasal discharge is also quite variable. Stertor may be clearly audible in severe, longstanding cases especially if there is some degree of tracheal collapse, or if significant exudates are present in the airways. The donkey's demeanour appears to be most dependent on the degree of respiratory distress. In the majority of cases the donkey is remarkably bright and alert, with a normal appetite. In more severe cases the donkey adopts a posture that assist pulmonary ventilation with the head lowered and nostrils flared. In this situation there is usually no interest in food and the prognosis is grave.

Clinical Examination - Nasal discharge is inconsistent but when present is usually a tacky, odourless, mucopurulent discharge. Auscultation may reveal a variety of adventitious lung sounds reflecting the great variety of lesions encountered with this condition. Any combination of crackles, wheezes, squeaks or reduced noise may be auscultated. This makes distinguishing this condition from acute bacterial bronchopneumonia using history and clinical examination alone difficult. However, pyrexia (rectal temperature above 37.7°C) that would be expected with bronchopneumonia appears to be an inconsistent finding with IPF.

Diagnosis and Differential Diagnosis - Severe dyspnoea due to conditions other than IPF is uncommon at the Donkey Sanctuary. However, cases of tracheal collapse, inhalation pneumonia, bronchopneumonia, pulmonary abscessation, pulmonary oedema and pleural effusion are encountered occasionally. Dyspnoea may also develop secondary to other conditions. Acute cyathostomosis or other acute protein losing conditions may be associated with dyspnoea presumably because of acute pulmonary oedema following massive falls in plasma albumin. Sudden internal haemorrhage either through trauma or neoplasia can also cause acute dyspnoea. Some degree of dyspnoea usually accompanies hyperlipaemia in the donkey. In these cases a full and thorough clinical examination accompanied with blood biochemistry and haematology is usually sufficient to exclude most differential diagnoses. Radiography will demonstrate an interstitial pattern. The extent of pulmonary oedema and pleural effusions may also be assessed by radiography. Assessment of the cardiac silhouette may reveal right-sided enlargement, especially in long-standing cases of chronic respiratory disease. This finding would indicate the development of secondary pulmonary hypertension and hence a predisposition for the development of cor pulmonale. Thoracic ultrasound may be used to confirm the radiographic changes. Tracheal washes are beneficial for diagnosing concurrent conditions such as bronchitis or Dictyocaulus infection. However, a diagnosis of bacterial bronchitis or lungworm infection should not be made without first excluding IPF. Lung biopsy could be a useful technique but this has not been attempted on any cases at the Donkey Sanctuary.

Gross Pathology - There is a wide spectrum and patterns of interstitial, subpleural and pleural fibrosis with mild and severe fibrosis generally appearing more frequently than intermediate levels of fibrosis. However, the two poles of the spectrum appear to be over-represented at post-mortem, with mild and severe lesions arising more frequently than moderately affected cases.

**Table 2. Differential diagnoses for dyspnoea in the donkey**

**Respiratory conditions**

Interstitial pneumonia  
 Bronchopneumonia - primary bacterial or secondary inhalation pneumonia following choke  
 Tracheal collapse  
 Pleural effusions or pleural haemorrhage e.g., neoplastic  
 Pulmonary oedema - secondary to acute protein losing conditions such as cyathostomosis.  
 Anaphylaxis or post-surgery.

Pulmonary neoplasia  
 Pulmonary abscessation  
 Pulmonary hydatidosis  
 Chronic pleural adhesions  
 Fractured ribs  
 Diaphragmatic rupture  
 Guttural pouch disease producing pharyngeal compression  
 Pharyngeal and laryngeal paralysis (laryngeal paralysis has been observed with hepatic disease)

**Non-respiratory conditions**

Colic including pancreatitis and gastrointestinal tract impactions producing abdominal distension  
 Hyperlipaemia  
 Anaemia or hypovolaemia

**Table 3. A comparison of gross pathology in mildly affected and severely affected fibrotic lungs in donkeys**

Gross Observation	Mildly Affected Lungs	Severely Affected Lungs
<b>Weight</b>	Normal	Heavy
<b>Palpation</b>	A few firm nodules palpable	Dorsal lobes consolidated
<b>Pleura</b>	No lesions although white fibrotic pattern visible through pleura, affecting dorsal lobes more severely	Often multiple fibrinous or connective tissue plaques on the dorsal lobes. Pattern of sub-pleural fibrosis visible through the pleura is described below
<b>Fibrotic Pattern</b>	Focal, discreet areas of fibrosis	Large areas (whole lobules) diffusely fibrosed. In donkeys that have recovered from the acute crisis, the subpleural fibrosis is more generalised. Septa of fibrosis are more prominent
<b>Airways</b>	Usually unaffected	In the more acute stages of disease the airways are grossly unaffected. In long-standing disease mucous accumulations may be present in the airways

Mild Fibrotic Lesions - Mild lesions appear grossly as multiple foci of irregular, often vermiform, fibrosis of the visceral pleural. The lungs are normal to palpate and are a normal weight. Foci of sub-pleural fibrosis are usually concentrated over the dorsal lobes, especially that of the caudal lobes. Rarely are focal areas of pleural fibrosis seen affecting the ventral lung lobes. The demarcation between fibrosis and normal lung is typically sharp. These cases are incidental findings on post-mortem, having no previous history of lung disease. Both right and left lungs are usually affected; occasionally one side is more severely affected than the other.

Severe or Long-standing Fibrotic Lesions - With severe or long-standing cases of IPF, the fibrosis is more generalised. In these cases the lungs feel heavy when removed from the thoracic cavity. On gross inspection the generalised pleural fibrosis means the lungs are very pale and firm to palpate (Fig. 5a). Circles of pinker lung, 1 - 3cm in diameter, may be visible through the zone of subpleural fibrosis. The consolidated lung can act as a cast for the thorax, retaining the rib impression

marks normally lost once the lung is removed from the thoracic cavity. Subpleural fibrosis along these impression marks accentuates the pattern. There may be some weak fibrous adhesions between visceral and parietal pleura in severe cases, again normally seen attached to the dorsal lobes. The accessory lobe is occasionally adhered to the right lobes. Sometimes plaques of fibrous tissue with a ragged edges are present, the remains of what presumably were once pleural adhesions. The pattern of collapse and consolidation is variable, sometimes regional but often generalised.



Figure 5a. Photograph of lungs from a donkey with severe pulmonary fibrosis. - To view this image in full size go to the IVIS website at [www.ivis.org](http://www.ivis.org) . -

On cut section, septa of interlobular fibrosis are usually noted extending from thick subpleural fibrosis, connecting with fibrosis in the region of the large airways. The subpleural fibrosis can be extremely thick, up to 20 mm thick dorsally in some instances. Other areas of peri-bronchiolar interstitial fibrosis will be evident without septa of fibrosis to the pleura. If the interstitial fibrosis is generalized a subtle marbling affect is produced with areas of pale consolidated lung surrounding pinker areas of normal lung. The contents of the airway lumen can vary from normal amounts of frothy secretions to mucopurulent material. In one case a large thrombus was located in the lumen of the caudal bronchus originating from a smaller airway. A small quantity of clear, stringy mucus in the trachea, bronchi and bronchioles is a frequent finding.

Apart from adhesions, the parietal pleura remains remarkably unaffected even with the most dramatic visceral pleura fibrosis. Likewise, the pleural fluid is usually normal in volume and on gross inspection.

Histopathology - Even with an acute presentation of dyspnoea the fibrous tissue seen on histological examination is generally mature. Depending on the extent of the disease the fibrosis may be well demarcated, separated from normal lung parenchyma by a thin region of alveolar septal fibrosis. In other cases the septal fibrosis is more diffuse which may reflect a later stage of disease. The multifocal nature of the condition is seen histologically as complete fibrosis of entire lobules in some areas.

Thick layers of fibrosis extend from the pleura along the interlobular septa and alveolar septa. In places the lobular fibrosis is so extensive as to completely obliterate the alveoli. Residual alveolar spaces can be filled with amorphous, acidophilic material. Focal alveolar oedema and islands of normal lung parenchyma are common findings. Within the areas of fibrosis, areas of parenchyma free of fibrotic changes are often affected by passive congestion.

Airway inflammation is usually localised to parts of the collapsed and fibrosed lung. This is unlike the more generalised small airway inflammation typical of heaves in horses. Tracheal collapse, bronchitis, bronchiolitis, emphysema, bronchiectasis, neutrophil luminal plugs and mural necrosis are likely to be the secondary effects of prolonged and severe dyspnoea or a complication of a functionally compromised region of lung. Peribronchial cuffing of the small airways with inflammatory cells is an inconsistent finding. Cellular aggregates are more commonly observed.

The pleura appear largely unaffected histologically. The fibrosis and inflammation appears largely confined to the subpleural region. The occasional pleural tag or plaque of fibrosis is observed, but these findings are conserved to the visceral pleura. The observed inflammatory infiltrates are mild, monocytic and multifocal (aggregates of lymphocytes, plasma cells, macrophages and haemosiderophages). A diffuse monocytic infiltration on the parenchymal surface of the pleural fibrosis has been noted. In places the septal fibrosis is not accompanied by a cellular infiltrate. Rarely are neutrophils observed in the interstitial tissues. Haemosiderophages are a common but inconsistent finding. Presumably severe dyspnoea will result in some pulmonary capillary diathesis.



Figure 5b. Subpleural fibrosis under low power. - To view this image in full size go to the IVIS website at [www.ivis.org](http://www.ivis.org) . -

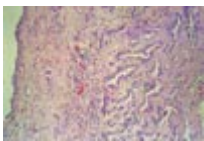


Figure 6. Subpleural fibrosis under high power - To view this image in full size go to the IVIS website at [www.ivis.org](http://www.ivis.org) . -



Figure 7. Subpleural fibrosis under high power - To view this image in full size go to the IVIS website at [www.ivis.org](http://www.ivis.org) . -



Figure 8. Subpleural fibrosis and epithelialisation. - To view this image in full size go to the IVIS website at [www.ivis.org](http://www.ivis.org) . -

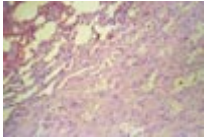


Figure 9. Mature interstitial fibrosis. - To view this image in full size go to the IVIS website at [www.ivis.org](http://www.ivis.org) . -

**Pathogenesis** - Interstitial pneumonias have been classified according to various aetiologies [59] including infectious agents, toxins, chemical damage or allergic reactions. Attempts to isolate organisms from bronchial swabs and lung parenchyma from donkey cases have failed to demonstrate any infectious agent to date. This is consistent with the report in horses [56]. Similar reports of interstitial pneumonia of an uncertain aetiology also appear in human medical literature. As the pathology is always of a chronic nature it is plausible that a fastidious organism present in the acute phases of disease will one day be isolated. However the chronic active nature of the pathology would suggest a progressive and on-going irritation is responsible for the lesions. This may be consistent with an allergen-induced hypersensitivity or repeated toxin-induced damage. Although some young animals are affected the changes are seen more frequently in older donkeys. An association between dental disease and pulmonary fibrosis may be a coincidental finding given both conditions are more prevalent in old age, with dental disease commonly appearing without any evidence of pulmonary fibrosis. However interstitial pneumonia secondary to bacteraemia originating from infected tooth roots cannot be ruled out yet. A response to lungworm infestation has been speculated but there has been no evidence to date to support this.

**Treatment** - Treatment is aimed at optimising the performance of the remaining functional lung in order to maintain the donkey within compensated lung failure.

Treat any bacterial infections whether primary or secondary with antibiotics as indicated by the clinical exam or diagnostic tests. In acute cases there is often little or no response to potentiated sulphonamides, penicillin or ceftiofur. However oxytetracycline 10 - 15 mg/kg slow IV or IM has had some anecdotal success.

Bronchodilators such as clenbuterol 0.8 µg/kg slow IV or orally 0.8 - 3.2 µg/kg bid or etamiphylline camsylate 3 mg/kg IM/SC/PO TID. Dembrexine to loosen the "plug forming" mucous accumulations within collapsed lower airways preventing normal lung aeration.

Prednisolone 1 mg/kg can be used to reduce any lower airway or interstitial inflammation and attenuate the fibrotic process. Inhalation therapy with nebulized beclomethasone is an alternative but the rationale for this treatment when there is no evidence of secondary airway disease is questionable. Systemic corticosteroids are probably more practical for long-term therapy and interstitial and alveolar inflammation are likely to be more reliably addressed by the systemic route.

Clean air management is advised to optimise airway function. Avoid housing in dry, dusty conditions that may exacerbate airway irritation and further compromise the lung. Clipping before the onset of the hot summer months may help prevent complications due to heat stress. On hot days ensure there is cool shelter available.

Fluid therapy may be indicated if dehydration and dry mucoid airway secretions are detected, although pulmonary oedema is a potentially fatal consequence of excessive fluid therapy.

**Prognosis** - The first acute episode of dyspnoea proves fatal in majority of cases at the Donkey Sanctuary. Death or euthanasia usually occurs within two weeks of onset of signs. Donkeys that recover from an acute crisis or have chronic and progressive lesions often continue in a state of compensated lung failure for many years. However, eventually the subpleural fibrosis results in a severe restrictive state, too extreme for compensation.

Secondary bacterial bronchitis is a common feature of IPF. If diagnosed in its early stages, antibiotics and mucolytics may be used to prevent accumulation of inflammatory exudates in the airways of an already severely compromised lung. Recurrent bronchitis in a chronically dyspnoeic donkey may indicate poor environmental management or may be a factor of end-stage lung disease.

**Cor Pulmonale** - A further consequence of chronic respiratory disease with extensive pulmonary fibrosis may be cor pulmonale. In this condition there is enlargement and thickening of primarily the right ventricle, due to the increased load on the heart. Clinical signs include the development of a pronounced jugular pulse, rising heart rate, abnormal cardiac rhythms and murmurs. At post-mortem the heart is enlarged and globose in shape due to hypertrophy and dilation of the right ventricle, and to a lesser extent, the left ventricle. At the Sanctuary this condition has been seen in elderly donkey and given the progressive nature of the problem the prognosis is grave and probably warrants euthanasia.

## Pulmonary Neoplasia

Neoplasia is an uncommon but important contribution to the differential diagnosis of chronic lung disease, especially in elderly donkeys. The tumours may be primary or secondary to metastatic spread, with clinical signs attributable to the primary site and/or the respiratory system. Between 1997 and 2001, eight cases of thoracic neoplasia were found at post-mortem. The mean age of the affected donkeys was 29 years.

There were five cases of adenocarcinoma, mean age at death 28 years. Three of these cases had no previous clinical signs relating to the respiratory system. Of these, one tumour was a cholangioadenocarcinoma of the liver with secondary metastases to the lung, two showed multiple nodular growths on the pleura, lung surface and peritoneum. The two further cases exhibited dyspnoea, tachypnoea and abnormal respiratory noise. There was hyperglobulinaemia, hypoalbuminaemia and mild anaemia. One case had a primary mass within the right lung measuring 20 cm by 15 cm. The other had extensive destruction of the lung and multiple foci of abnormal tissue.

The remaining three thoracic neoplasia were:

1. Primary mediastinal carcinoma with multiple metastases.
2. Mandibular sarcoma with metastases to the lung.
3. Pericardial mesothelioma causing terminal intrathoracic haemorrhage.

Clinical signs were variable consisting of dullness, colic, persistent pyrexia and dyspnoea.

The variable clinical signs and biochemical findings mirror those found in cases of thoracic neoplasia in the horse [60]. Pre-mortem diagnosis may be achieved via radiology and ultrasonography combined with thoracocentesis or lung biopsy, although the likely value of intensive work-ups may need to be balanced in the geriatric donkey by the stress of investigations.

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## Appendixes

- Appendix 1 - Physiological, haematological and biochemical values for donkeys - [PRINT](#)
- Appendix 2 - Modified Baermann Technique - [PRINT](#)

### Appendix 1 - [PRINT](#)

Physiological, haematological and biochemical values for donkeys (and ponies where they differ significantly)							
	Range				Range		
	Median	5%	95%		Median	5%	95%
Temperature (°F)	98.8	97.2	100	Mean Corpuscular Volume - fl	64	57	79
Pony	100 - 101			Pony	36 ± 5		
Young Donkey	99.6	97.8	102.1	Young Donkey	54	49	70.5

				Young Horse	35.5 ± 7.2		
<b>Temperature (°C)</b>	37.1	36.2	37.8	<b>Mean Corpuscular Haem. - pg</b>	21.9	18.9	28.6
Pony	37.8 - 38.3			Pony	12.1 ± 2.2		
Young Donkey	37.6	36.6	38.9	Young Donkey	18.5	16.4	25
<b>Pulse (beats/min)</b>	44	36	68	<b>Mean Corpuscular Haem Conc. - g/dl</b>	34.8	31.4	39.1
Pony	36 - 40			Young Donkey	35	25.3	54.0
Young Donkey	60	44	80				
<b>Respiration (breaths/min)</b>	20	12	44	<b>Creatinine - mmol/l</b>	75	53	141
Pony	10 - 14			Pony	134		
Young Donkey	28	16	48	Young Donkey	83	61	107
<b>Packed Cell Volume - litre/litre</b>	0.33	0.25	0.38	<b>Creatine Phosphokinase - IU/l</b>	40	15	149
Pony	0.40±0.055			Young Donkey	41	21	180
Young Donkey	0.34	0.27	0.43				
<b>Haemoglobin - g/dl</b>	11.6	9	15.3	<b>Total Bilirubin - mmol/l</b>	2.7	1.4	7.7
Pony	13.6 ± 1.6			Pony	10.3 ± 3.4		
Young Donkey	11.6	9.3	15	Young Donkey	2.7	1.4	6.1
<b>Neutrophils - %</b>	50.5	28	78	<b>Urea - mmol/l</b>	3.9	1.9	7.6
Young Donkey	45	25	71	Young Donkey	3.7	1.5	6.1
<b>Neutrophil count - 10<sup>9</sup>/l</b>	5.0	2.2	13.3	<b>Triglycerides- - mmol/l</b>	1	0.2	4.3
Young Donkey	6.3	3.3	14	Young Donkey	0.7	0.2	2.0
<b>Lymphocytes - %</b>	43	17	65	<b>Total Protein - g/l</b>	70	58	82
Young Donkey	50	16	67.5	Young Donkey	64	53	78

<b>Lymphocyte count - 10<sup>9</sup>/l</b>	4.2	1.8	7.8	<b>Albumin - g/l</b>	28	20	34
Young Donkey	6.2	2.5	14	Young Donkey	28	21	32
<b>Eosinophils - %</b>	4	1	10	<b>Total Globulins - g/l</b>	40	29	53
Young Donkey	2	0	10	Young Donkey	34	23	50
<b>Eosinophil count - 10<sup>9</sup>/l</b>	0.38	0.09	1.15	<b>γ-Glutamyl Transferase - IU/l</b>	17	8	49
Young Donkey	0.30	0	1.63	Young Donkey	16	8	39
<b>Basophils - %</b>	0	0	0.08	<b>Glutamate Dehydrogenase - IU/l</b>	1.6	0.4	8
Young Donkey	0	0	0	Young Donkey	1.2	0.4	3.9
<b>Basophil count - 10<sup>9</sup>/l</b>	0	0	0.5	<b>Immunoglobulin T - IU/l</b>	1	1	4
Young Donkey	0	0	0	Young Donkey	1	1	6
<b>Monocytes - %</b>	1	0	5	<b>Aspartate aminotransferase - IU/l</b>	109	59	199
Young Donkey	1	0	6	Young Donkey	103	63	172
<b>Monocyte count - 10<sup>9</sup>/l</b>	0.13	0	0.80	<b>Glutathione peroxidase - IU/l</b>	12.1	4.5	51
Young Donkey	0.16	0	0.81				
<b>White Blood Cell Count - 10<sup>9</sup>/l</b>	10.2	6.1	16.1	<b>Alkaline phosphatase - IU/l</b>	265	150	563
				Pony	70 ± 18		
Young Donkey	13.5	7.8	21.9	Young Donkey	305	212	576
<b>Red Blood Cell count - 10<sup>12</sup>/l</b>	5.5	4	7.3	<b>Sorbitol dehydrogenase - IU/l</b>	0.3	0	1.5
Pony	8 - 12						
Young Donkey	6.3	5	8.1	Young Donkey	0.4	0.1	1

Adapted from French and Patrick [61]

## Appendix 2 - PRINT

### Modified Baermann Technique

The Baermann apparatus consists of a large funnel supported upright by a stand. Rubber tubing is connected to the bottom of the funnel and a pipette tip inserted in this. A clip is fitted to the rubber tubing.

1. Fold a piece of paper into four and write the sample identity on it. Place the paper in the funnel, opening it out to line the funnel. Close the clip tightly. Add a small amount of water to wet the paper, check that there is no leakage from the pipette tip.
2. Place 50 g of broken up faeces in the filter paper. (The amount of faeces, packed down, that reaches to the 60 ml mark of the 100 ml plastic beaker is approximately equal to 50 g).
3. Pour on enough water to cover the faeces, mix it gently, taking care not to tear the paper.
4. Leave it to stand for at least 12 hours.
5. Open the clip and allow about 10 ml to run into a centrifuge tube. Note, if this is added directly to a microscope slide then a qualitative result can be gained.
6. Centrifuge for two minutes at 1,500 r.p.m. or leave to stand for 2 hours.

Using a pipette, take a small amount of fluid from the very bottom of the tube, where any larvae will have settled, and spread it out on a microscope slide. Examine using low power and count all the lungworm larvae seen on the slide. The identity of the lungworm can be checked using a higher power. The number counted is the number of larvae per 50 g of faeces.

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