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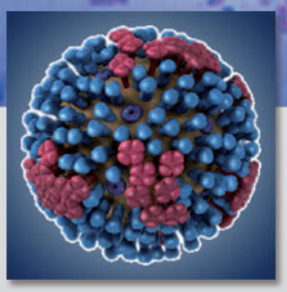
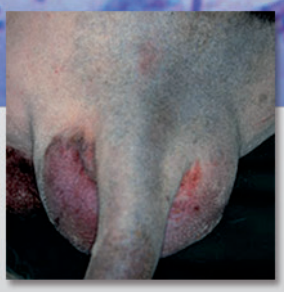


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Fecava Symposium*

Feline intestinal protozoa - zoonotic importance or not?

Michael R. Lappin[1]

SUMMARY

Of the intestinal protozoa of cats, the coccidians *Toxoplasma gondii* and *Cryptosporidium* species and the flagellate *Giardia* species have the most zoonotic risk. It is known that most *Cryptosporidium* species in humans, dogs or cats are host adapted. The cat and dog genotypes, *C. canis* and *C. felis*, are less common than *C. hominus* and *C. parvum* in people and are unlikely to cause illness in people. *Giardia* species can be characterized into genetic assemblages that appear to be mainly be host-specific, with cats and dogs usually being infected by assemblages C, D and F. However, assemblages A and B, which are associated with illness in humans, are occasionally identified in faeces of cats and dogs. *Toxoplasma gondii* oocysts require an incubation period (1 – 3 days) before being infectious and cats only shed an average of 7-10 days. Thus, most people are exposed to *T. gondii* in a contaminated environment or by ingesting undercooked meat. In contrast, *Cryptosporidium* species oocysts and *Giardia* species cysts are immediately infectious when passed by the host. Host-adapted *Cryptosporidium* species and *Giardia* species are common in cats but if individual cats are healthy with normal faeces, there appears to be little zoonotic risk to people.

Key words. *Cryptosporidium*, *Giardia*, *Toxoplasma*, cat, assemblage

* This paper was presented at the FECAVA Symposium on Zoonosis on 7 November 2014. Eur J Comp An Pract 25(1), Spring 2015, p4-7
Go to http://www.fecava.org/ejcap to see the online presentation of this paper.

Cryptosporidium

Cryptosporidium spp. inhabit the respiratory and intestinal epithelium of many vertebrates, including birds, mammals, reptiles and fish. Once thought to be a commensal, *Cryptosporidium* spp. are now known to cause gastrointestinal tract disease in several mammalian species, including cats, calves dogs, humans and rodents. The organisms have an enteric life cycle similar to that of other coccidians that culminates in the production of thin-walled, autoinfective oocysts and thick-walled, environmentally resistant oocysts that are passed in faeces (Scorza and Tangstrongsup, 2010). Oocysts (4-6 µm in diameter) are passed sporulated and are immediately infectious to other hosts.

Multiple species of *Cryptosporidium* spp. exist, including *Cryptosporidium parvum, C. hominis, C. felis* and *C. canis*. Although some *Cryptosporidium* infect multiple animal species, others have a limited host range. However, strains that infect both pets and people cannot be differentiated by light microscopy from those that infect only pets, so all *Cryptosporidium* spp. should be considered potentially zoonotic. The most common *Cryptosporidium* spp. isolated from dogs and cats are the host-adapted *C. canis* and *C. felis*, respectively. However, zoonotic species have also been amplified (Sotiriadou et al, 2013). The relative risk of *C. felis* to humans has been discussed in many manuscripts to date (Ballweber et al, 2010; Bowman and Lucio-Forster, 2009; Lucio-Forster et al, 2010; Thompson et al, 2008; Xiao and Fayer, 2008).

[1] Michael R. Lappin DVM PhD DACVIM. 300 West Drake Road, Fort Collins, Colorado State University, Colorado 80523, USA.
E-Mail mlappin@colostate.edu

The prevalence of *Cryptosporidium* spp. oocysts in dog and cat faeces approximates that of *Giardia* (5 – 20%). *Cryptosporidium parvum* infection of humans after exposure to infected calves has been recognized for years. Human infection associated with contact with infected dogs and cats has been reported but illness associated with *Cryptosporidium* species of dogs and cats is thought to be unusual. While DNA of *C. felis* or *C. canis* has been amplified from the faeces of some humans, there is no evidence suggesting the agents cause disease or are spread to other humans. Most humans have infection with *C. hominus*, the host-adapted strain or *C. parvum*. Person-to-person contact with oocysts byfaecal-oral contamination and ingestion of contaminated water are the most likely routes of exposure.

The small size (approximately 4-6 µm in diameter) of *Cryptosporidium* spp. oocysts leads to difficulty in diagnosis after faecal flotation; fluorescent antibody staining or acid-fast staining is more sensitive. Multiple enzyme-linked immunosorbent assays (ELISA) for the detection of *C. parvum* antigen in faeces are commercially available but do not accurately detect *C. felis* or *C. canis*. PCR is the most sensitive test to date but is not standardized among laboratories (Scorza and Tangstrongsup, 2010).

No drug has been shown to eliminate *Cryptosporidium* spp. from the gastrointestinal tract (Scorza and Tangstrongsup, 2010). Thus, the major goal for management of a dog or cat with diarrhoea and *Cryptosporidium* species infection is to resolve the diarrhoea, not affect a therapeutic cure. Avoiding exposure is the most effective prevention. Routine disinfectants require extremely long contact with the organism to be effective. Drying, freeze thawing and steam cleaning can inactivate the organism. Surface water collected in the field for drinking should be boiled or filtered.

Giardia

Giardia spp. (flagellate), *Entamoeba histolytica* (amoeba) and *Balantidium coli* (ciliate) are enteric protozoans that can be transmitted to humans by contact with faeces; the cysts do not require an incubation period to become infectious. *Entamoeba histolytica* infection is extremely rare in dogs and cats; *B. coli* infection is rare in dogs and has not been reported in cats.

Giardia spp. infection of dogs and cats is common and can be detected in faeces of normal dogs and cats and in those with small-bowel diarrhoea (Scorza and Tangstrongsup, 2010; Epe et al, 2010). Stools of dogs or cats with *Giardia* may also contain mucous. Because the organism is immediately infectious when passed as cysts in stool, direct zoonotic transfer is possible. Genetic studies have detected multiple *Giardia* spp., and most dogs and cats are infected with the host-adapted assemblages C, D and F (Scorza et al, 2012). However, assemblages A and B are also detected in feline faeces, suggested that cats can harbour the zoonotic assemblages (Jaros et al, 2011; Overgaauw et al, 2009; Scorza et al, 2012; Sotiriadou et al, 2013). As is the case with *Cryptosporidium*, because determining zoonotic strains of *Giardia* spp. by microscopic examination is not possible, assume that diarrhoeic faeces from all dogs and cats infected with *Giardia* spp. are a potential human health risk until the stools are normal. Genotyping is available commercially in some countries (www.dlab.colostate.edu).

Healthy pets (http://www.cdc.gov/healthypets/index.html) are generally not considered significant human health risks to HIV-infected people by the Centers for Disease Control in the United States. Dogs and cats with normal stools are unlikely to be sources of human *Giardia* infection. However, because clinical signs induced by *Giardia* spp. can be intermittent and since some *Giardia* spp. may be zoonotic, treatment of healthy infected dogs or cats should be considered with each owner. Treatment of healthy dogs or cats is controversial because all of the drugs can potentially cause side-effects, animals with normal stools are not considered human health risks, treatment is unlikely to eliminate infection, and re-infection can occur within days. For example, in a recent study of naturally infected dogs, > 75% of treated dogs were still *Giardia* infected when rechecked 34 days after treatment. If treatment deemed indicated by the clinician and owner, many clinicians currently recommend that a 5-day course of fenbendazole be administered for apparently healthy dogs or cats that test positive for *Giardia*. But the Companion Animal Parasite Council (www.capcvet.org) states that repeated treatment is not indicated in healthy pets. The Center for Disease Control in the United States maintains a website providing pet owners about how to deal with *Giardia* infection in their pets (http://www.cdc.gov/parasites/giardia/giardia-and-pets.html).

Toxoplasma

Toxoplasma gondii is a coccidian that is one of the most prevalent parasites infecting warm-blooded vertebrates around the world (Lappin 2010; Vollaire et al, 2005). Only cats complete the sexual phase in the gastrointestinal tract and pass environmentally resistant oocysts in faeces. Sporozoites develop in oocysts after 1 to 5 days of exposure to oxygen and appropriate environmental temperature and humidity. Sporozoites can penetrate the intestinal tract of cats or intermediate hosts and disseminate in blood or lymph as tachyzoites during active infection. *Toxoplasma gondii* can penetrate most mammalian cells and will replicate asexually within infected cells until the cell is destroyed. If an appropriate immune response occurs, replication of tachyzoites is attenuated and slowly dividing bradyzoites develop that persist in within cysts in extra-intestinal tissues. Tissue cysts form readily in the CNS, muscles and visceral organs. Live bradyzoites may persist in tissues for the life of the host.

Toxoplasma gondii seroprevalence rates vary by the lifestyle of the cat. In general, increasing prevalence correlates with increasing age from risk of exposure over time and with cats allowed outdoors as these cats are most likely to contract infected intermediate hosts. In a recent study of clinically ill cats, *T. gondii* antibodies in 31.6% of the 12,628 cats tested (Vollaire et al, 2005). While *T. gondii* infection is common in cats, the oocyst shedding period is generally < 21 days. Thus, detection of *T. gondii* oocysts in feline faeces is uncommon (usually < 1%).

Infection of warm-blooded vertebrates occurs following ingestion of any of the three life stages (sporozoite, tachyzoites, bradyzoites) of *T. gondii* or transplacentally. It is also possible that cats are infected lactationally. Most cats are not coprophagic and so most are infected by ingesting *T. gondii* bradyzoites during carnivorous feeding; oocysts are shed in faeces from 3 to 21 days. Sporulated oocysts can survive in the environment for months to years and are resistant to most disinfectants. Transmission of *T. gondii* is most efficient when cats consume tissue cysts (carnivorism) and when intermediate hosts consume oocysts (faecal-oral transmission).

While *T. gondii* seroprevalence rates are approximately 30% in cats around the world, oocysts are passed for only a short time. Thus, *T. gondii* is not likely to be acquired from individual cats; ingestion of sporulated oocysts in water or soil contaminated by old cat faeces or ingestion of undercooked meat are greater risks. Primary *T. gondii* infection in immunocompetent individuals results in self-limiting fever, malaise and lymphadenopathy that may be not recognized or is misdiagnosed. Primary infection of mothers by *T. gondii* during gestation can lead to clinical toxoplasmosis in the foetus; stillbirth, CNS disease and ocular disease are common clinical manifestations. As T-helper cell counts decline, approximately 10% of people with AIDS develop toxoplasmic encephalitis from activation of bradyzoites in tissue cysts. *Toxoplasma gondii* infection of rodents changes the behaviour of the prey species making it less averse to cats, potentially increasing the likelihood the definitive host (felid) will become infected and potentiate the sexual phase of the organism. Recently, *T. gondii* has been proposed as a cause of behavioural problems in people (Flegr 2013).

People most commonly acquire toxoplasmosis by ingesting sporulated oocysts or tissue cysts or transplacentally. To prevent toxoplasmosis, avoid eating undercooked meats or ingesting sporulated oocysts. Although exposure to cats is epidemiologically associated with acquiring toxoplasmosis some studies, touching individual cats is probably not a common way to acquire toxoplasmosis for the following reasons.

1. Cats generally only shed oocysts for days to several weeks after primary inoculation.
2. Repeat oocyst shedding is rare, even in cats receiving glucocorticoids, cyclosporine or in those infected with feline immunodeficiency virus or feline leukaemia virus.
3. Cats with toxoplasmosis inoculated with tissue cysts 16 months after primary inoculation did not shed oocysts.
4. Cats are very fastidious and usually do not allow faeces to remain on their skin for time periods long enough to lead to oocyst sporulation; the organism was not isolated from the fur of cats shedding millions of oocysts 7 days previously.

However, since some cats will repeat oocyst shedding when exposed a second time, faeces should always be handled carefully. If a faecal sample from a cat is shown to contain oocysts measuring 10 X 12 µm it should be assumed that the organism is *T. gondii*. The faeces should be collected daily until the oocyst shedding period is complete; administration of clindamycin (20 mg/kg, daily) blocked *T. gondii* oocyst shedding in cats when administered prior to

infection and may shorten the oocyst shedding period if started after infection is documented.

Since humans are not commonly infected with *T. gondii* from contact with individual cats, testing healthy cats for toxoplasmosis is not recommended (www.cdc.gov). Faecal examination is an adequate procedure to determine when cats are actively shedding oocysts but cannot predict when a cat has shed oocysts in the past. There is no serologic assay that accurately predicts when a cat shed *T. gondii* oocysts in the past, and most cats that are shedding oocysts are seronegative. Most seropositive cats have completed the oocyst shedding period and are unlikely to repeat shedding; most seronegative cats would shed the organism if infected. If owners are concerned that they may have toxoplasmosis, they should see their physician for testing.

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Conflict of interest

None to report.

Ethical statements

Not applicable to this work

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FECAVA Symposium*

Influenza in cats and dogs – Risk for humans?

Thomas W. Vahlenkamp, DVM, PhD[1]

SUMMARY

Dogs and cats are susceptible to natural influenza virus infections which are transmitted from avian-adapted virus reservoirs to cats and avian and equine-adapted virus reservoirs to dogs. Due to the heterogeneity of influenza viruses in their natural reservoirs of water fowl and the recent natural infections of carnivores with influenza viruses of the subtypes H1 (H1N1), H3 (H3N2, H3N8) and H5 (H5N1, H5N2), contact of dogs and cats with birds, poultry, pigs or horses in areas where there are influenza outbreaks should be avoided to prevent the possible spread of the virus and possible human exposure to influenza. The transmission of influenza A virus from different mammalian and avian species to carnivores may result in viral adaptation; therefore the epidemiological role of infected dogs and cats needs closer attention.

Key words: Influenza, cat, dog, zoonosis

*This paper was presented at the FECAVA Symposium on Zoonosis, on 7 November 2014.
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Introduction

Every year, influenza A viruses in humans cause seasonal infections with estimated thousands of deaths worldwide. Pandemic infections occur rarely, with the latest being the Spanish Flu (H1N1) in 1918, the Asian Flu (H2N2) in 1956, the Hong Kong Flu (H3N2) in 1968, and the Pandemic Flu (H1N1) in 2009. Since 1997, and in particular 2003, Bird Flu (H5N1) has caused severe economic losses and public health concerns because this virus has been shown to be transmitted (although relatively inefficiently) directly from birds to humans. Most recently a new avian influenza virus of subtype H7N9 caused infections in humans. This has emphasised the need for basic research to further elucidate host virus receptor specificities and mechanisms of pathogenesis.

Influenza A viruses are enveloped viruses of the family Orthomyxoviridae and contain a segmented genome. When two viruses infect an individual target cell, reassortant viruses are formed; this is where the progeny viruses contain a genome composition which is a mixture of the two parent viruses. This mechanism described as anti genetic shift contributes significantly to the overwhelming biological variety of influenza A viruses in nature. In addition, single point mutations contribute to viral adaptation to a particular host. This mechanism is described as antigenic drift. As a result of this biological variety and in order to differentiate the different varieties, the virus nomenclature follows this pattern: the virus type / geographic origin / strain number / year of isolation and virus subtype (e.g. A/Fujian/411/2002 [H3N2]).

Influenza in cats and dogs

Although cats and dogs live as pets in close contact to humans, they have largely been neglected for many years as possible hosts for influenza viruses. Several serological surveys have reported influenza A virus antibodies in dogs

[1] Institute of Virology, Faculty of Veterinary Medicine, University of Leipzig, An den Tierkliniken 29, 04103 Leipzig (Germany).
Email: thomas.vahlenkamp@uni-leipzig.de

and cats, however, in the last century, no positive confirmation by virus detection has been reported [1]. Recent serological surveys in pet cats in the USA [2] have revealed moderate seroprevalences against three circulating human-adapted subtypes (H1N1, H3N2, pandemic H1N1) ranging from 22 to 44 %. However, in contrast, serological surveys in cats and dogs in Europe revealed no [3] or only sporadic seropositive samples attributed to H1N1 pdm [4,5] or equine H3N8 viruses [6].

In the last decade, several avian influenza viruses have caused outbreaks, with some of them associated with severe clinical signs in dogs, cats and large felids. These are listed below.

Influenza H5N1

The hemi-pandemic spread of the highly pathogenic avian influenza virus (HPAIV) of subtype H5N1 has caused severe economic damage for the poultry industry in many countries, including in particular South-East Asia and Africa. Due to the zoonotic nature of this virus, there is a tangible threat for public health in countries where the virus has established endemic infections. Spillover transmissions from infected poultry or wild birds have not only occurred in humans but also in cats, dogs, and mustelids [1,7]. The first reports of an extended host spectrum of this virus came from China where in 2002 a tiger died due to an H5N1 infection. Later on, in 2004, a leopard, a tiger and three domestic cats were reported to have died due to a fatal H5N1 infection [8]. In 2005 an outbreak of H5N1 was reported in a zoo in Thailand where 147 out of 441 tigers died or had to be euthanised due to H5N1 infections. Experimental infections confirmed the susceptibility of cats to this particular avian influenza virus[9].

Cats and dogs can shed virus from the nose without showing apparent signs of disease, and receptors for the avian (H5N1) virus are present not only in the lower part of the respiratory tract of dogs but also in their trachea and nose [10]. So far the transmission of HPAIV H5N1 from infected poultry or wild birds to large felids and domestic cats has been reported in eight countries in Asia and Europe and this caused considerable problems and concerns for both veterinary and public health officials in recent years. Experimentally, the infection could also be transmitted from diseased to naive cats. In dogs, a fatal H5N1 infection has been reported under natural conditions. In general it seems that cats are more susceptible than dogs to this particular virus [11]. These findings warrant special attention with respect to the potential progression of avian influenza H5N1 in dogs and cats. Therefore the contact of dogs and cats with birds and poultry should be avoided in areas with influenza outbreaks to prevent the possible spread of the virus and ultimately human exposure to influenza H5N1 that might have been adapted to mammals.

Influenza H5N2

In China, a novel influenza A virus H5N2 was isolated from a dog with pyrexia and respiratory signs of coughing, sneezing and sniffling [12]. Genome analysis of this virus revealed an ancestral relationship to the swine and turkey influenza viruses. Under experimental conditions, dog-to-dog transmission could be demonstrated which was accompanied with increased body temperatures and mild respiratory syndromes including transient conjunctivitis, sneezing nasal discharge, and mild coughing. In addition, there was virus shedding and seroconversion.

Influenza H3N8

Influenza A virus of equine origin (H3N8) caused influenza outbreaks in greyhounds in Florida. In the USA, up until 2008, canine influenza virus H3N8 infections had been reported in pet dogs from 25 US states [13]. Like other influenza outbreaks, infections were also associated with clinical respiratory signs. Further canine H3N8 isolates were obtained from tissues archived in 2003, 2004 and 2005 [13]. Evidence for a similar, but less expansive outbreak was also reported in the UK [14]. Severe respiratory disease was noticed in a kennel of foxhounds. Whilst a virological agent could not be isolated, antibodies against equine influenza virus H3N8 were detected in the recovered animals. In contrast to the epidemiological situation in the USA, transmission or spread of the virus within the UK, or to other European countries, seems to be rather limited [15].

Experimental infection of SPF dogs with subtype H3N8 resulted in seroconversion and virus excretion without obvious signs of disease. Natural infections with influenza H3N8, however, resulted in serious illness, death, and widespread infection in dogs.

A live recombinant equine herpesvirus-1 vaccine expressing the HA of equine H3N8 reduced the clinical signs and

virus shedding in challenged dogs [16]. Similar results were obtained with a canary poxvirus vector engineered to express equine HA of subtype H3 [17].

Influenza H3N2

In 2007, a canine lineage of influenza A virus of subtype H3N2 was identified as a causative agent of an epidemic outbreak of severe respiratory disease, similar to those seen in US greyhounds, among pet dogs throughout South Korea [18,19]. Genetic analysis revealed an avian origin for these viruses. This was the first time that a low pathogenic avian influenza virus (LPAIV) was found to cause respiratory disease in dogs under natural conditions. Affected breeds included Miniature Schnauzers, Cocker Spaniels, Jindo dogs and Yorkshire terriers.

Influenza-like respiratory signs, such as dyspnoea, were observed among cats as well as in dogs in an animal shelter located in Seoul, South Korea [20]. The affected cats showed 100 % morbidity and 40 % mortality. A virus could be isolated from a lung specimen of a dead cat which had suffered from the respiratory disease. The eight viral genes isolated were almost identical to those of the canine influenza H3N2 virus, suggesting interspecies transmission of the canine influenza H3N2 virus to the cat. Subsequent experimental investigations showed that intranasal infection of domestic cats with canine/Korea/GCVP01/07 (H3N2) resulted in elevated rectal temperatures, nasal virus shedding and severe pulmonary lesions, such as suppurative bronchopneumonia. In addition, airborne interspecies transmission of canine influenza virus H3N2 from dogs to cats was also reported [21].

Influenza H1N1

In Mexico in March/April 2009, a triple reassortant influenza A virus of subtype H1N1 with a genetic lineage from human, classical swine, and Eurasian swine influenza viruses emerged. Within a few months infections were also found in cats and dogs [22,23]. Natural infection of dogs with the H1N1 pdm virus revealed that the virus caused respiratory signs with viral shedding in nasal secretions. Dogs were clinically recovered after three to four days. Experimental infection in a 10-week-old beagle revealed that the H1N1 pdm virus caused elevated temperatures associated with mild respiratory signs. Transmission of the virus from the experimentally infected animal to naïve contact dogs proved to be relatively inefficient [24]. Similarly to the clinical signs shown in dogs, naturally infected cats exhibited depression, loss of appetite, and respiratory signs with a clinical improvement within one week [25]. Experimental H1N1 pdm virus infection in cats resulted in mild to moderate disease with the most common signs being elevated temperature, laboured breathing, loss of appetite and protruding nictitating membranes.

Conclusion

So far interspecies transmissions of influenza viruses from cats or dogs to humans have not been reported. However, during the last decade several influenza viruses have been described in companion animals. This is of particular interest as influenza virus infections in cats and dogs were historically generally unnoticed. Due to the heterogeneity of influenza viruses in their natural reservoirs and the recent documented infections in carnivores, the contact of dogs and cats in areas with influenza outbreaks in e.g. birds, poultry, pigs or horses should be avoided to prevent possible spread of the virus and subsequent human exposure to influenza.

Infectivity and transmissibility studies in dogs using the human seasonal H3N2, pandemic (pdm) H1N1 (2009) and B influenza viruses revealed that dogs may be hosts for human seasonal H3N2 and H1N1 pdm influenza viruses [26]. As the transmission of influenza A virus to carnivores from different mammalian and avian species may allow for viral adaptation, the epidemiological role of infected dogs and cats needs closer attention.

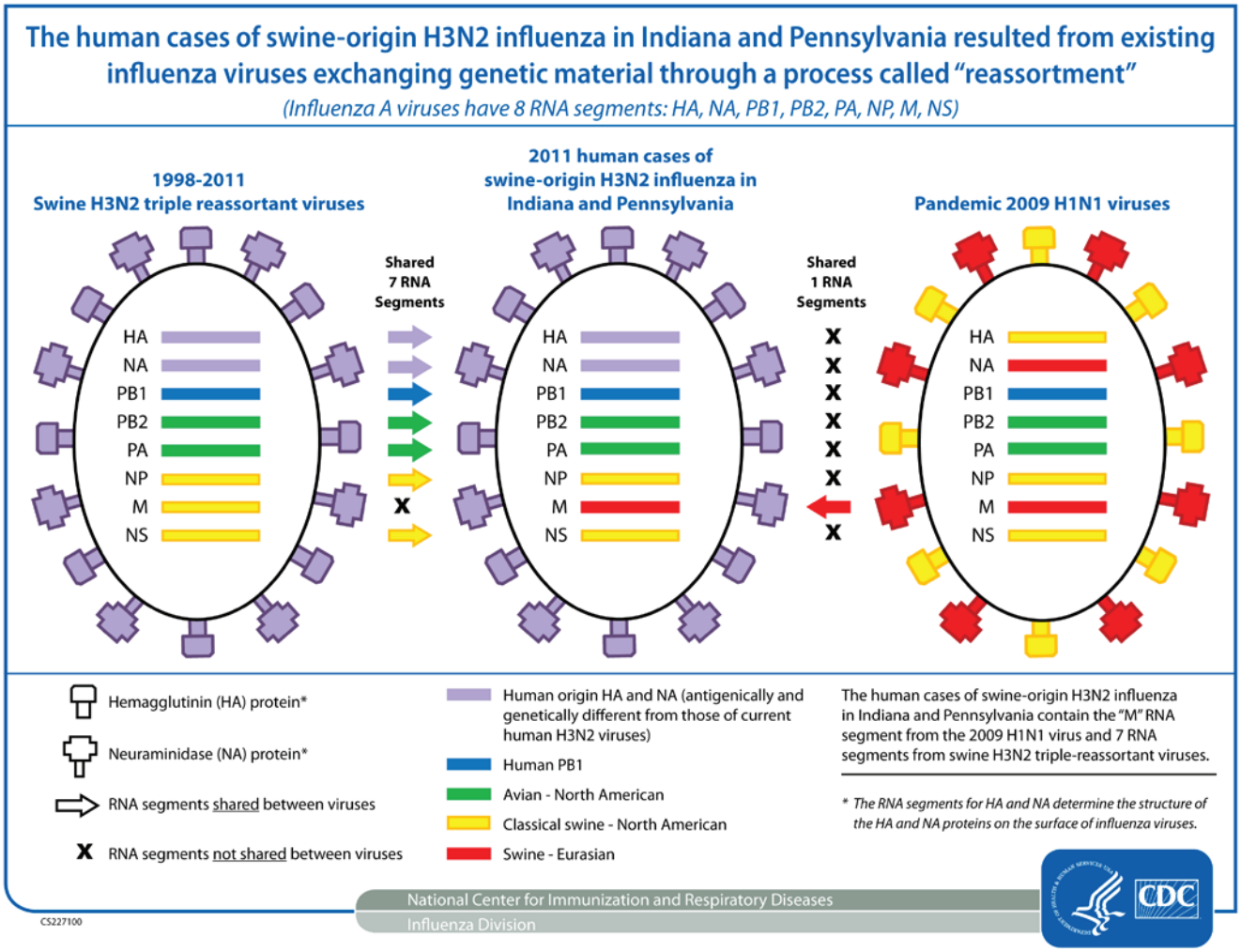

Fig 1: This diagram depicts how the human cases of swine-origin H3N2 influenza virus (USA, September 2011) resulted from the reassortment of two different influenza viruses. The diagram shows three influenza viruses placed side by side, with eight colour-coded RNA segments inside of each virus. Note: All influenza viruses contain 8 RNA segments. These RNA segments are labelled HA (haemagglutinin), NA (neuraminidase), PB1, PB2, PA, NP, M and NS. (©CDC/ Douglas Jordan, MA)

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FECAVA Lecture*

Perineal hernia – is a staged approach best?

R.A.S. (Dick) White[1]

SUMMARY

Perineal hernia (PH), or the failure of the pelvic diaphragm to support the rectum during defaecation leading to abnormal rectal faecal accumulation is a commonly presented clinical entity in small animal practice (Figures 1 & 2).

The underlying aetiology for this disease still remains unclear but older, male entire dogs predominate.

PH requires surgical management and the prognosis correlates primarily with:

- Surgical technique selected
- Increasing experience with technique

The prognosis has not been shown to correlate with concurrent castration but discussion still surrounds the benefit of performing PH as a part of staged process to address concurrent disease (e.g. prostatic disease, bladder retroflexion).

* This paper is based on the FECAVA Lecture delivered at the 20th EuroCongress in Munich, Germany.
Eur J Comp An Pract (2015), Winter 25(1); p13-p17
Go to http://www.fecava.org/ejcap to see the online presentation of this paper.

Anatomy and Physiology of the Pelvic Diaphragm

The rectum is supported within the pelvic canal by a number of striated muscular and fascial structures that contribute to anorectal and rectal function during defaecation is entirely dependent on the capacity of these muscles to compress the pelvic contents.

They include:

- the external anal sphincter,
- the levator ani muscle and,
- the coccygeus muscle (Figure 3)
- The anal sphincter muscles comprise an internal and an external component. The fascia of the outer layer of striated muscle is closely applied to the levator and coccygeus muscles.
- The levator ani m. is a triangular shaped muscle that lies in close contact with the lateral rectal wall. It originates on the medial iliac shaft and pelvic symphysis and inserts on through its tendon ventral to the Co7 vertebra. There is also an intimate attachment on its medial surface to the pelvic fascia and by this to the anal sphincter m. Contraction causes angulation

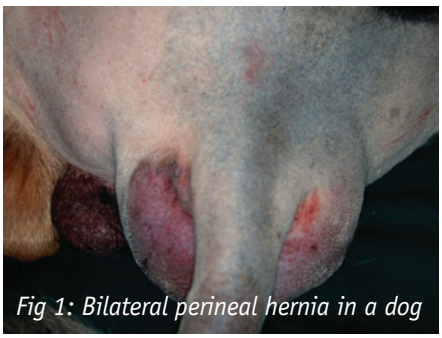

Fig 1: Bilateral perineal hernia in a dog

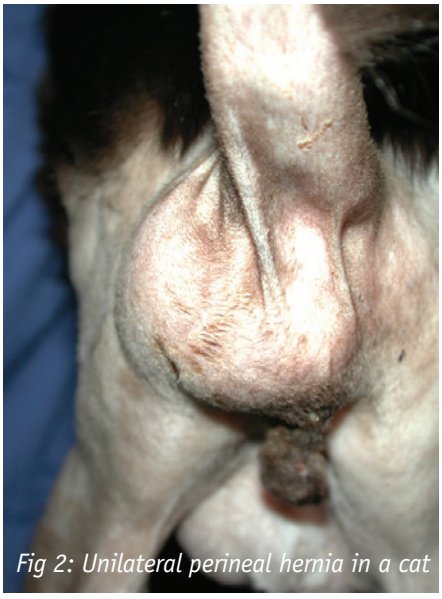

Fig 2: Unilateral perineal hernia in a cat

[1] Dick White Referrals, Station Farm, London Road, Six Mile Bottom, Cambridgeshire CB8 0UH, UK. Email dw@dwr.co.uk

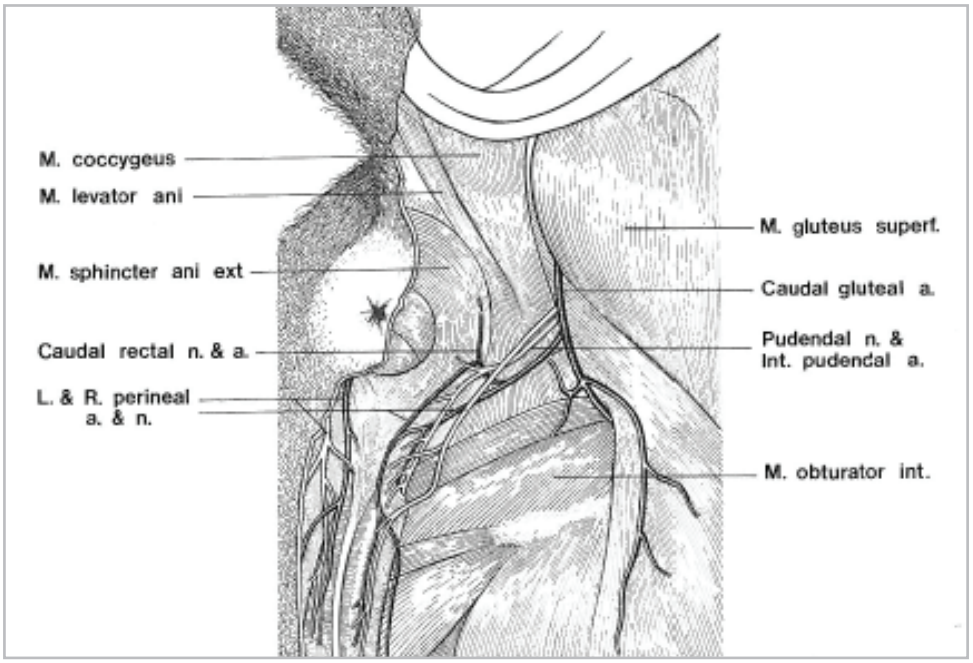

Fig. 3: Anatomic relationship of the muscles of the pelvic diaphragm.

of the tail at Co 6/7, rectal compression and presses the tail against rectum during defaecation.

- The coccygeus m. is a rectangular shaped muscle which lies caudal and lateral to the levator ani. It originates on the tendon on spine of ischium and inserts on the transverse processes of Co 2–5. Contraction results in rectal compression and presses the tail ventrally against rectum during defaecation.

During defaecation waves of rectal peristalsis propel the faecal bolus towards the anus. Voluntary contraction of the diaphragm and abdominal muscles results in an increase in intra-abdominal pressure whilst a further pressure change within the rectum results from contraction of the muscles of the pelvic diaphragm which compresses the rectum in a dorsoventral direction.

Pathophysiology

The single major consequence of PH is the increase in the rectal capacity as the lateral support is progressively lost (Figure 4 & 5). Unilateral rectal enlargement is termed sacculation whilst the bilateral disease is termed dilatation. Rectal diverticulation is almost never encountered. Herniated tissues usually include pelvic and peritoneal fat but occasionally loops of small intestine may become involved. The most serious complication of all is retroflexion of the bladder (+/- prostate gland) (Figures 6 & 7).
PH is encountered almost exclusively in the male dog

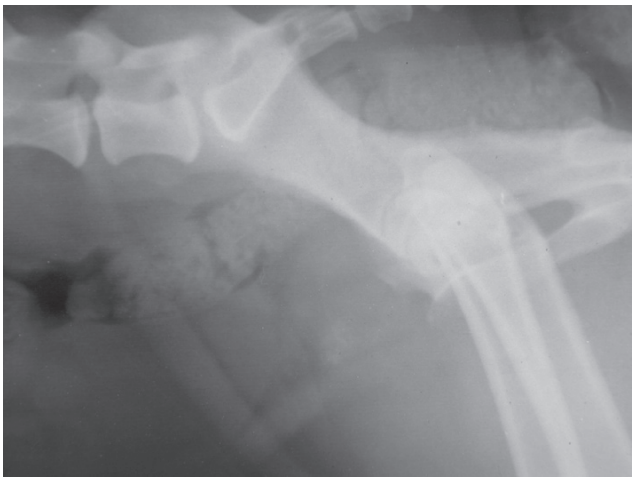

Fig. 4: Rectal accumulation of faeces in perineal hernia.

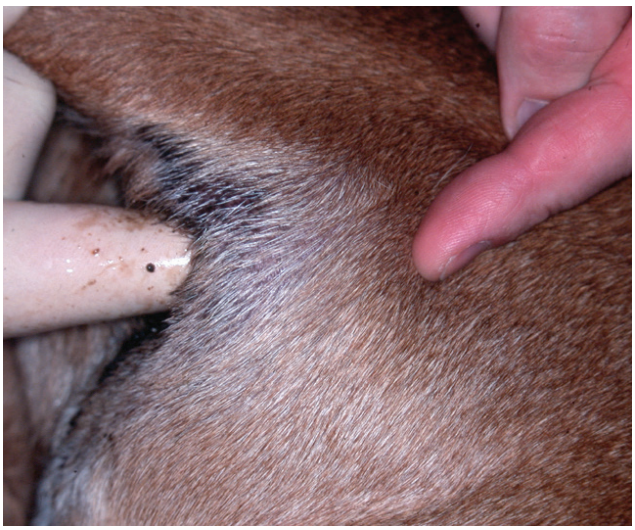

Fig. 5: Perineal 'touch' technique for confirmation of perineal hernia.

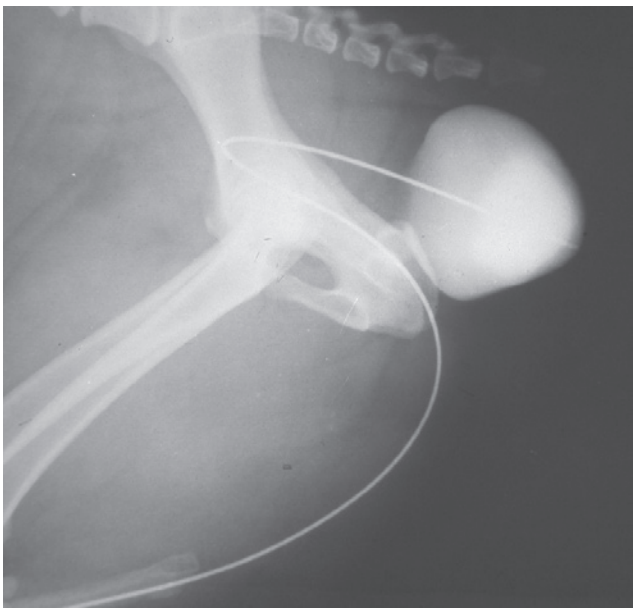

Fig. 6: Bladder retroflexion highlighted on cystography.

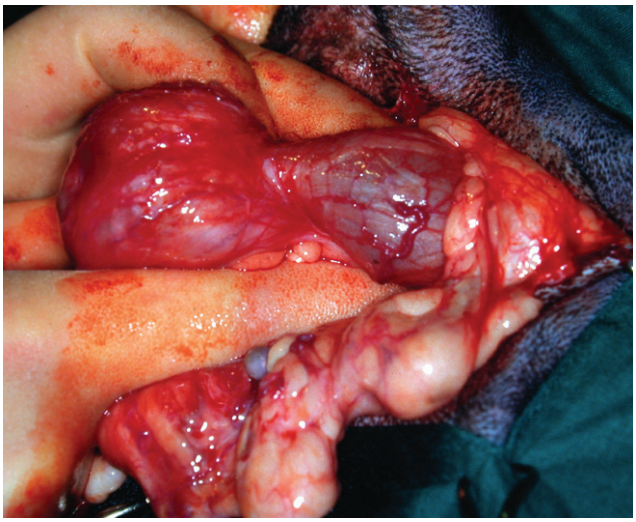

Fig. 7: Retroflexion of bladder and prostate during surgical repair of perineal hernia.

although it is occasionally seen in the bitch and still less frequently in cats.

Aetiopathogenesis

Key features of the incidence of PH include:

- Age 7 - 9 yrs
- Gender male (>99%)
- Side R > L
- Breed Collie, Boxer, Yorkie, Dachshund, Old English Sheepdog

The aetiology of PH remains unclear but is associated with the degenerative changes which occur in the muscles of the pelvic diaphragm resulting from changing androgen receptor / secretion from the ageing testicle. There is little evidence to support the suggestion that the condition is encountered more frequently in docked breeds. Various theories for the aetiopathogenesis of PH have been put forward including:

1. **Myopathy** - Reduced numbers of androgen receptors are found in the muscles of the pelvic diaphragm in dogs with PH; decreased dihydrotestosterone levels in the ageing dog exacerbate this inadequate trophic androgen influence.
2. **Prostatomegaly** - prostatic disease is common in perineal hernia and the consequent defaecatory tenesmus increases intra-abdominal pressure which may lead to weakening of the pelvic diaphragm.
3. **Relaxin** - A 'Relaxin-like' substance secreted by the ageing prostate inhibits collagen synthesis and enhances its breakdown. Receptors for 'Relaxin' are found in pelvic muscles and atrophy is limited to pelvic diaphragm[1]
4. **Lower Bowel Inflammatory Disease** - is a consistent feature in some breeds with perineal hernia e.g. German shepherd dogs.

Management

Apart from rare asymptomatic cases, PH is always managed surgically. Several techniques have been described for PH repair and these include:

1. **Re-apposition**
   - *a.* ***'Conventional' or dorsal repair:*** the muscles of the pelvic diaphragm (coccygeus/levator ani and anal sphincter) are re-apposed and sutured to the anal sphincter muscle (Figures 8 - 11)
2. **Muscle Augmentation**
   - *a.* ***Transposition of the internal obturator muscle:*** the internal obturator muscle is tenotomised and sutured to the anal sphincter (Figures 12 - 14). The long term success rate for this technique on its own is unknown.
   - *b.* ***Superficial gluteal m. transposition:*** the tendon of the superficial gluteal muscle is resected and the muscle transposed over the deficient diaphragm before suturing to the anal sphincter.
   - *c.* ***Semitendinosis m. transposition:*** the distal insertion of the semitendinosis muscle is resected allowing the muscle to be used for ventral support of the pelvic diaphragm.
3. **Reinforcements Concepts**
   - *a.* ***Polypropylene mesh implants*** have been investigated but there are few reports of successful management using this technique.
   - *b.* ***Porcine SIS***
   - *c.* ***Fascia lata***

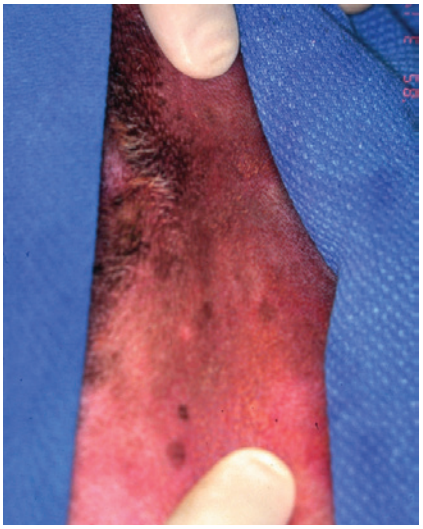

Fig. 8: Dorsal perineal hernia repair i) landmarks for incision – base of tail to tuber ischium.

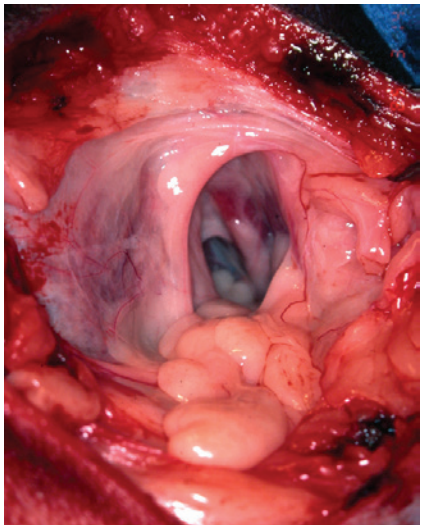

Fig. 9: Dorsal perineal hernia repair ii) separation of levator and coccygeus muscles from external anal sphincter permits rectal enlargement and herniation of pelvic fat

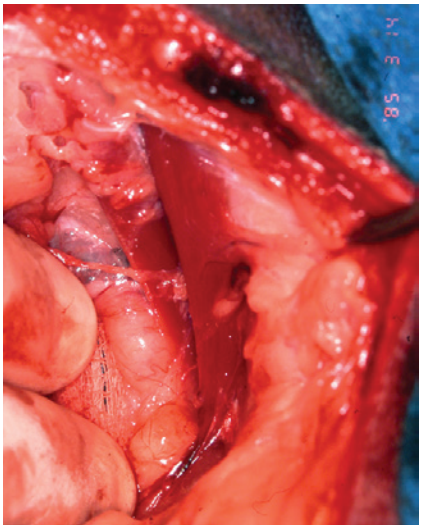

Fig. 10: Dorsal perineal hernia repair iii) Dissection of fascia permits accurate visualisation of the levator ani (medial) and coccygeus (lateral) muscles.

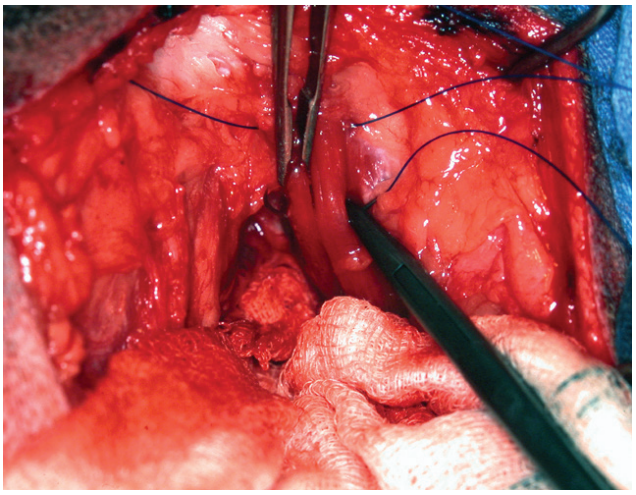

Fig. 11: Dorsal perineal hernia repair iv) Sutures placed through levator and coccygeus muscles for attachment to fascia of anal sphincter muscle.

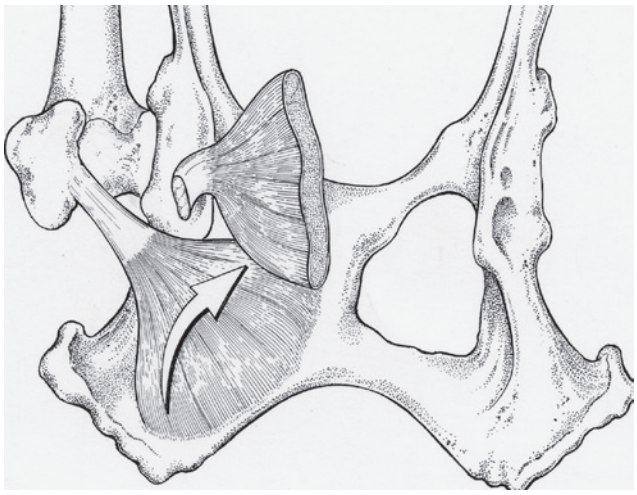

Fig. 12: Internal obturator muscle transposition i) representation of relocation following transection of tendon.

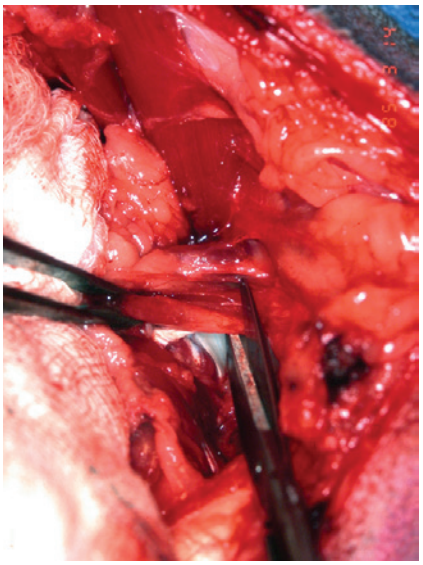

Fig. 13: Internal obturator muscle transposition ii) transection of tendon.

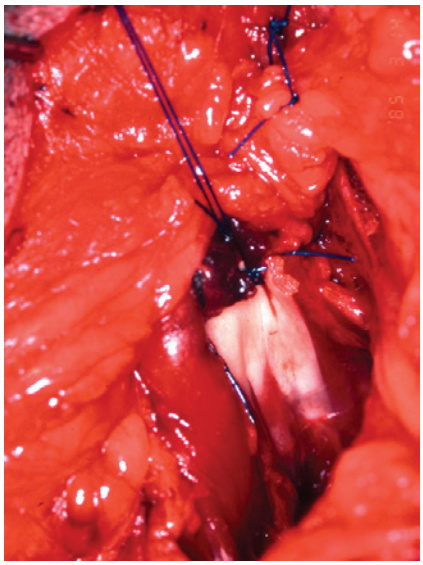

Fig. 14: Internal obturator muscle transposition ii) transected tendon sutured to fascia of anal sphincter muscle to provide ventral support in combination with dorsal repair.

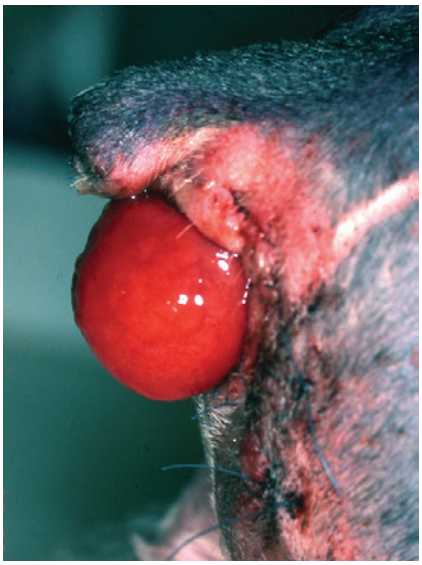

Fig. 15: Rectal prolapse immediately post-operatively for perineal hernia repair.

Outcome

- Techniques 1 & 2a have recurrence rates of approximately 30% whereas a combination of 1 & 2b result in less than 10% recurrence rates[2].
- Increasing surgical experience has been shown to significantly improve outcomes.
- The impact of concurrent castration on recurrence rates is unclear.
- Urinary bladder retroflexion does not adversely influence outcome[3]

Surgical Complications

- **Bladder retroflexion** - herniation of the bladder occurs in 20% of all hernias and is a potentially serious complication. This may lead to bladder rupture, azotaemia and renal failure. Herniating bladders should be decompressed by urethral catheterisation or, if this is not possible, by direct cystocentesis. Adequate repair should preclude the need for cystopexy or vas deferensopexy to prevent further retroflexion.
- **Recurrence** - Owners should be warned that recurrence is a possibility but selection of the most appropriate technique and accurate surgical reduction should limit this to <10% at the most. Repeated herniation after surgery is uncommon but nevertheless a very difficult problem to resolve in some cases. Poor identification of the anatomic components of the hernia is the most common reason for failure.
- **Rectal eversion / prolapse** - is occasionally seen after surgery. Lubrication with lidocaine gel and gentle reduction are usually all that is necessary but some may require temporary purse string sutures or epidural anaesthesia to reduce the tenesmus. (Figure 15)
- **Sciatic paralysis** - is recorded but a very rare but much publicised complication of surgery and may occur where sutures are inadvertently placed lateral to the sacrotuberous ligament.

The Staged Approach to Management of PH

Step 1 involves laparotomy for management of concurrent conditions. It has been suggested that staged management is indicated for 'complicated' perineal hernias including hernia recurrence, major rectal dilatation, concurrent surgical prostatic disease and retroflexed bladder[4]. However, a more rational approach to separate 'cause' of PH from 'effects' of PH; those conditions that are causes may benefit from a staged approach.

Causes:

- Surgical prostatic disease (e.g. gross enlargement due to BPH, large cysts, abscesses or prostatitis) should be regarded as potential causes of PH and therefore may benefit from a staged approach.
- Inflammatory bowel disease should be regarded as potential causes of PH and therefore may benefit from a staged approach.

Effects:

- Bladder retroflexion is an effect of PH and the indications for staged cystopexy are less compelling.
- Rectal prolapse is most commonly an effect of PH.

Step 2 the definitive hernia repair is performed following a 7 day intervention.

Conclusions for Staged Management

- Resolving the conditions that are likely to cause recurrence of PH before definitive surgery may be a good idea.
- The rationale for trying to resolve conditions that are the consequences of PH is more open to debate.

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Reprint paper*

Dyspnoea in cats: is it the heart?
Differentiating between feline pleural effusions of cardiac and non-cardiac origin using pleural fluid NT-proBNP concentrations

Karen Humm[1], Melanie J Hezzell, Julia Sargent, David J. Connolly and Adrian Boswood

SUMMARY

Objective: To assess whether pleural fluid and urine amino terminal proB-type natriuretic peptide (NT-proBNP) can distinguish cardiac from non-cardiac causes of pleural effusion.

Methods: Blood, urine and pleural fluid were prospectively collected from cats presenting with pleural effusion categorised as cardiac or non-cardiac in origin. NT-ProBNP concentrations were measured using a feline-specific enzyme-linked immunosorbent assay. Groups were statistically compared and receiver operating characteristic curves constructed to determine cut-offs to distinguish cardiac from non-cardiac pleural effusion in plasma, pleural fluid and urine.

Results: Forty cats with pleural effusion (22 cardiac and 18 non-cardiac) were studied. NT-proBNP concentrations in plasma and pleural fluid were strongly correlated. Plasma ($P<0.001$) and pleural fluid ($P<0.001$) NT-proBNP concentrations and urinary NT-proBNT/creatinine ratios ($P=0.035$) were significantly higher in the cardiac group. After receiver operating characteristic curve analysis, a plasma NT-proBNP cut-off of 214.3 pmol/mL was suggested [sensitivity=86.4% (95% CI: 66.7 to 95.3%), specificity=88.9% (95% CI: 67.2 to 96.9%)] and a pleural fluid NT-proBNP cut-off of 322.3 pmol/mL was suggested [sensitivity=100% (95% CI: 85.1 to 100%), specificity=94.4% (95% CI: 74.2 to 99.0%)]. No cut-off with adequate sensitivity and specificity for urinary NT-proBNP/creatinine ratios was suggested.

Clinical significance: Measurement of NT-proBNP in pleural fluid distinguishes cardiac from non-cardiac causes of pleural effusion in cats.

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Introduction

Cats frequently present to veterinary practitioners with dyspnoea secondary to pleural effusion. Disease processes causing pleural fluid accumulation include neoplasia, cardiac disease, pyothorax and feline infectious peritonitis (Waddell & King 2007). Various tests are available to aid the clinician in elucidating the underlying cause, including cytological examination of the pleural fluid, fluid culture and echocardiography (Beatty & Barrs 2010). In humans, the measurement of amino terminal proB-type natriuretic peptide (NT-proBNP) in pleural fluid has been found to

[1] Veterinary Clinical Sciences, The Royal Veterinary College, North Mymms, Hatfield Herts. AL9 7TA, United Kingdom. Email: khumm@rvc.ac.uk

be useful in distinguishing between cardiogenic and non-cardiogenic causes of pleural effusion (Porcel et al. 2004; Kolditz et al. 2006; Janda & Swiston 2010). NT-proBNP has also been measured in the urine of human patients with heart failure and has been found to correlate well with plasma concentrations (Cortés et al. 2007). Serum or plasma measurements of NT-proBNP have been shown to be beneficial in aiding identification of cardiac disease in cats (Connolly et al. 2009; Fox et al. 2009) and this has led to the development of a commercially available test for feline NT-proBNP (Vetsign Feline Cardiopet proBNP; IDEXX Laboratories).

Performing venepuncture on a cat in respiratory distress due to pleural fluid can be challenging and the stress it causes can be deleterious. As therapeutic thoracocentesis is mandatory in such cases and the collection of voided urine involves no contact with the animal, the value of NT-proBNP measurement in these fluids over serum is clear if they were to be diagnostic of cardiac disease.

The aims of this study were to determine whether NT-proBNP was detectable in the pleural fluid and urine of cats presenting with pleural effusion arising from a variety of causes, to assess whether pleural fluid and urine NT-proBNP correlate with plasma NT-proBNP and finally to determine whether pleural fluid and urine NT-proBNP concentrations could distinguish cardiac from non-cardiac causes of pleural effusion. The null hypothesis was that pleural and urinary NT-proBNP would not distinguish cardiac from non-cardiac causes of pleural effusion.

Materials and methods

The study was approved by the institutional ethics and welfare committee of the Royal Veterinary College (RVC) and informed owner consent was obtained. Cats with pleural effusion requiring thoracocentesis consecutively presenting to the RVC as first opinion emergencies or as referral cases were prospectively recruited to the study. Forty cats were recruited as this number was deemed to be possible within a 2-year period given the historical frequency of cats presenting with pleural fluid to the hospital. The signalment of the cats and their final diagnoses were recorded. Body condition score and systolic blood pressure were also recorded. All cats received a thorough physical examination and appropriate diagnostic tests, including echocardiography (interpreted by a board-certified veterinary cardiologist), in order to determine the cause of the pleural effusion. Cats were placed in one of two groups, those having a cardiac cause and those having a noncardiac cause of pleural effusion. Cats were classified as having a cardiac cause of their pleural effusion by the attending cardiologist after full analysis of their echocardiogram, history, physical examination and results of any other diagnostic tests performed.

Pleural fluid samples were obtained at the time of therapeutic or diagnostic thoracocentesis. Plasma samples were obtained when venepuncture was required for diagnostic samples. Urine samples were obtained either when diagnostic cystocentesis was performed or a free catch sample was obtained from the litter tray (filled with plastic beads) as soon as micturition was noted. One millilitre samples of pleural fluid and urine were collected and centrifuged at 3000 g for 5 minutes within 15 minutes of collection. The supernatant was transferred into commercially available tubes containing a proprietary protease inhibitor (PI tube) (Cardiopet proBNP specimen tubes; IDEXX Laboratories). Two millilitre samples of blood were collected into K3-EDTA treated tubes. Within 15 minutes of collection the samples were centrifuged at 3000 g for 5 minutes, separated, and 1 mL aliquots of plasma were transferred to PI tubes. Protease inhibited samples of pleural fluid, urine and plasma were stored at –80°C for batched analysis.

Samples were allowed to thaw at room temperature and plasma NT-proBNP (pmol/L) was measured within 1 hour of thawing using a commercially available enzyme-linked immunosorbent assay (ELISA) (Vetsign Feline Cardiopet proBNP; IDEXX Laboratories) previously validated for use with feline plasma (Connolly et al. 2008). The kit was used precisely according to the manufacturer's instructions. The kit incorporates two immunoaffinity- purified sheep antibodies specific for feline NT-proBNP. The plate consists of the capture antibody anti-NT-proBNP (1 to 20) bound to the wells of the plate. The tracer comprises the detection antibody, anti-NT-proBNP (60 to 80) conjugated to horseradish peroxidase. Incubation time was 5 hours at room temperature (mean temperature=22°C) and sample volume per well was 30 µL in all samples tested. Photometry was performed using a Wallac Victor 2 1420 Multilabel Counter. The lower and upper limits of detection used were those reported in the manufacturers' instructions, 24 and 1500 pmol/L, respectively. Values of NT-proBNP less than the lower limit of detection or greater than the upper limit of detection of the assay

were assigned values of 24 or 1500 pmol/mL, respectively. All samples were assayed in duplicate and the mean of the two values used. Following initial analyses, samples were pooled from a number of cats with measurements at the lower, middle and upper ends of the range of assay detection. These pooled samples were subsequently used for validation purposes. Case acquisition was not completed before the expiration of assay test kits obtained in house; samples collected subsequently (54 of 109) were assayed by a commercial laboratory (IDEXX Laboratories). One millilitre samples of urine were also spun as described above and the creatinine concentration in the supernatant was measured by a commercial laboratory (Diagnostic Laboratories, Royal Veterinary College). The ratio of urinary NT-proBNP to creatinine was calculated.

Statistical analysis

Statistical analysis was performed using commercially available software (SPSS 20; IBM). Precision and reproducibility of measurements of NT-proBNP in pleural fluid and urine were assessed by calculation of intra- and inter-assay coefficients of variation (CV), respectively, in samples of low, medium and high NT-proBNP concentrations. Data were examined graphically for normality of distribution. Group-wise comparisons were performed using Mann-Whitney tests, Wilcoxon-signed rank tests or Fisher's exact test, as appropriate. Correlations were assessed using Spearman's correlation coefficient. Receiver operating characteristic (ROC) curves were constructed to determine separate cut-offs to distinguish cardiac from non-cardiac causes of pleural effusion if significant differences between groups were detected. The areas under the ROC curves were compared using the method described by Hanley & McNeil (1983). The positive and negative likelihood ratios of these cut-offs were calculated for this population. The positive likelihood ratio is the ratio of true positives to false positives and the negative likelihood ratio is the ratio of true negatives to false negatives. A positive likelihood ratio greater than 5 is considered a reasonable diagnostic test for ruling in a condition, and a negative likelihood ratio less than 0.2 is considered a reasonable diagnostic test for ruling out a condition.

Results

Forty cats with pleural effusion were enrolled in the study between February 2011 and June 2012. All cats that presented with a pleural effusion and that had thoracocentesis and venepuncture were recruited when the study personnel were available for sample collection and processing. Pleural fluid and plasma samples were obtained in all cases with urine samples obtained in 28 of the 40 cats (65%). The study population consisted of 26 domestic short-haired cats, five domestic long-haired cats, two Maine coon and two Burmese cats, and one each of the following breeds: Bengal, Birman, Persian, rag doll and Siamese. Twentyfour cats were male neutered, 12 were female neutered, 3 were male entire and 1 was female entire. The median age of the cats was 9.5 years (range: 3 months to 16 years 4 months). Median body condition score was 4 of 9 (range: 2 to 7) although it was not recorded in four cats. Median blood pressure was 120 mmHg (range: 60 to 180 mmHg) with measurements not recorded for five cats. The cause of pleural effusion was determined as cardiac in 22 of the 40 cats. The echocardiographic diagnoses (as described by Bonagura (2010)) were hypertrophic cardiomyopathy (n=11), feline unclassified cardiomyopathy (n=5), restrictive cardiomyopathy (n=2), dilated cardiomyopathy (n=2) and arrhythmogenic right ventricular cardiomyopathy (n=2). The cause of pleural effusion was found to be non-cardiac in 18 cats, with underlying causes being neoplasia (n=8), idiopathic chylothorax (n=3), pyothorax (n=2) and traumatic chylothorax, aspiration pneumonia and feline infectious peritonitis (n=1 each). In two cats no cause for the pleural effusion could be determined; these cats were assigned to the non-cardiac group as echocardiographic findings were not suggestive of a cardiac cause of the effusion.

In pleural fluid samples assayed at the RVC, intra-assay CVs for samples of low (150 pmol/L), medium (609 pmol/L) and high (1332 pmol/L) NT-proBNP concentrations were 5.1, 8.6 and 2.1%, respectively. Inter-assay CVs for samples of low, medium and high NT-proBNP concentrations were 16.5, 10.3 and 10.0%, respectively. In urine, samples assayed by an external laboratory (IDEXX Laboratories), intra-assay CVs for samples of low (77 pmol/L), medium (115 pmol/L) and high (1620 pmol/L) NT-proBNP concentrations were 20.5, 3.9 and 7.6%, respectively. Inter-assay CVs for samples of low and medium NT-proBNP concentrations were 13.6 and 6.9%, respectively. Inter-assay CVs for high NT-proBNP concentrations could not be calculated as all results were greater than 1500 pmol/L. Urinary NT-proBNP was measured by both laboratories in five samples. No evidence of a difference in NT-proBNP measurements between laboratories was detected (P=0.500). Summary statistics are provided in Table 1. No differences in age, breed, sex, body condition score or systolic blood pressure

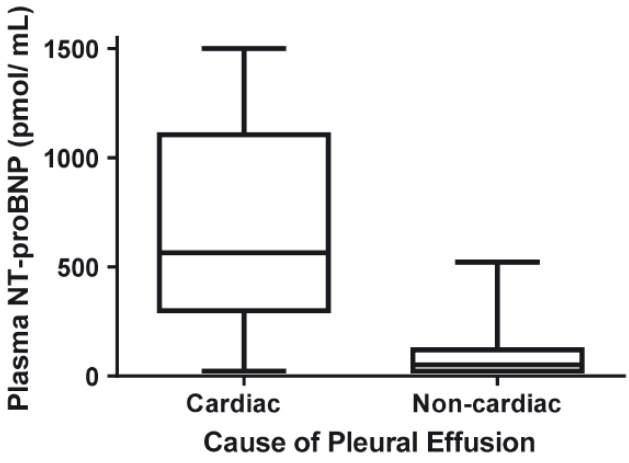
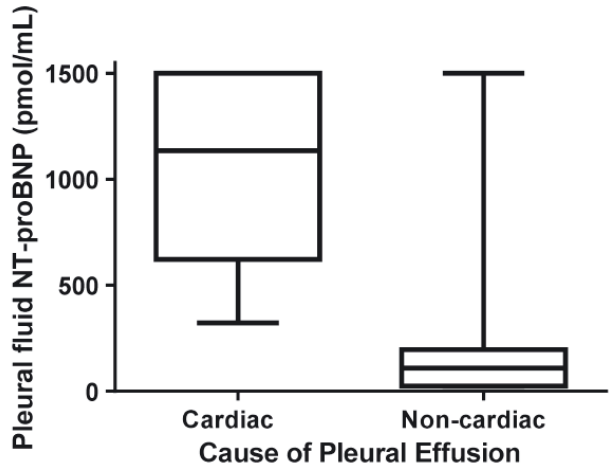


FIG 1. Box and whisker plots of plasma NT-proBNP for cats with cardiac and non-cardiac causes of pleural effusion. The lower and upper boundaries of the box represent first and third quartiles of the data respectively, with the line within the box representing the median. The whiskers represent the complete range of the data. A significant difference was detected between the groups (P<0.001)

were detected between groups. NT-proBNP concentrations were significantly higher in pleural fluid samples than in plasma (P<0.001). NT-proBNP concentrations were strongly correlated in plasma and pleural fluid samples (Rs=0.759, P<0.001). A moderate correlation between plasma NT-proBNP and urinary NT-proBNP/creatinine ratio (UBC) was detected (Rs=0.488, P=0.008). No correlations between plasma NT-proBNP and age (P=0.594), body condition score (P=0.606) or systolic blood pressure (P=0.409) were detected. NT-proBNP concentrations were significantly higher in the cardiac group in plasma (P<0.001) (Fig 1 and Table 1) and pleural fluid (P<0.001) (Fig 2 and Table 1); UBC was also significantly higher in the cardiac group (P=0.035) (Fig 3 and Table 1). Five cats with non-cardiac causes of pleural effusion had plasma NT-proBNP concentrations greater than 100 pmol/mL, two of which had plasma NT-proBNP concentrations greater than 360 pmol/mL.



F*IG 2. Box and whisker plots of pleural fluid NT-proBNP for cats with cardiac and non-cardiac causes of pleural effusion. The lower and upper boundaries of the box represent first and third quartiles of the data respectively, with the line within the box representing the median. The whiskers represent the complete range of the data. A significant difference was detected between the groups (P<0.001)*

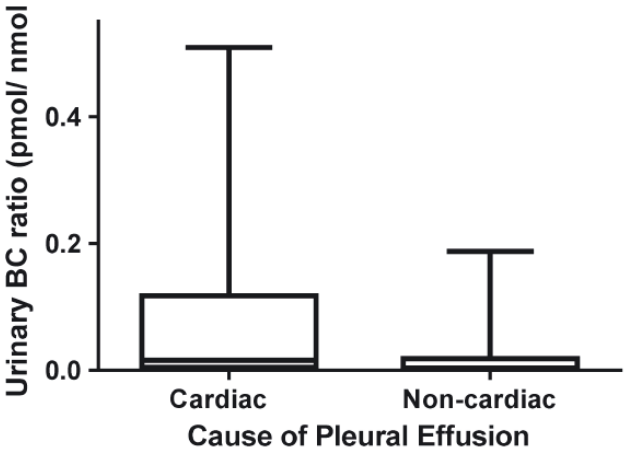


FIG 3. Box and whisker plots of urinary NT-proBNP to creatinine (BC) ratio for cats with cardiac and non-cardiac causes of pleural effusion. The lower and upper boundaries of the box represent first and third quartiles of the data respectively, with the line within the box representing the median. The whiskers represent the complete range of the data. A significant difference was detected between the groups (P=0.035)

Table 1. Differences in variables between the cardiac and non-cardiac groups. The median and interquartile range are shown for continuous variables

Variable	Cardiac group	Non-cardiac group	P
Age (months) (n=40)	119.5 (51.0, 153.8)	108.5 (81.0, 146.5)	0.463
Pedigree breed (yes/no) (n=40)	6/ 22	3/ 15	>0.999
Sex (male/female) (n=40)	16/ 6	11/7	0.509
Body condition score (n=36)	4.5 (4.0, 5.5)	4.0 (3.3, 5.0)	0.219
Systolic blood pressure (mmHg) (n=35)	113.0 (117.5, 133.5)	125.0 (115.0, 137.0)	0.419
Plasma NT-proBNP (pmol/mL) (n=40)	565.5 (300.1, 1104.4)	51.8 (24.0, 120.3)	*<0.001*
Pleural fluid NT-proBNP (pmol/mL) (n=40)	1135.3 (621.5, 1500.0)	111.0 (25.5, 197.1)	*<0.001*
Urinary NT-proBNP to creatinine ratio (pmol/nmol) (n=28)	0.016 (0.005, 0.448)	0.004 (0.001, 0.018)	*0.035*

Continuous variables and median (25th, 75th percentiles) are reported. Significant differences are highlighted in italic text

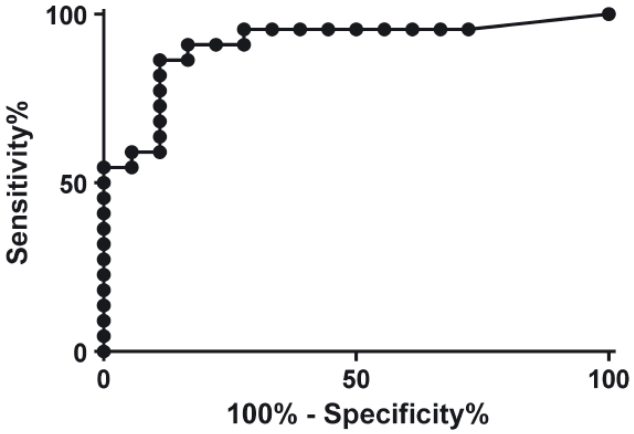


FIG 4. ROC curve for the prediction of cardiac cause of pleural effusion by plasma NT-proBNP. AUC=0.908 (95% CI: 0.810-1.000), P<0.001. NT-proBNP Amino terminal proB-type natriuretic peptide, ROC Receiver operating characteristic, AUC Area under the curve

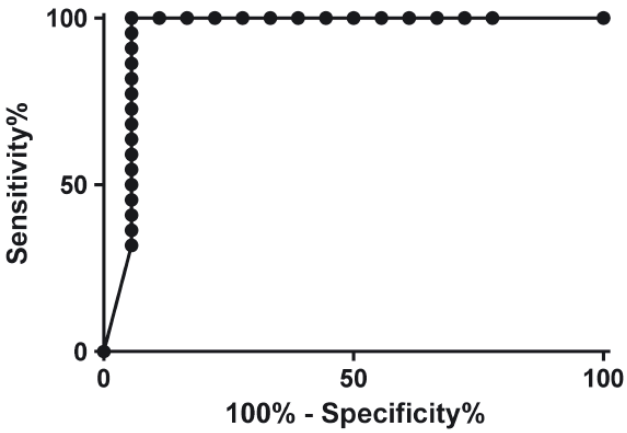


FIG 5. ROC curve for the prediction of cardiac cause of pleural effusion by pleural fluid NT-proBNP. AUC=0.953 (95% CI: 0.863-1.000), P<0.001. ROC Receiver operating characteristic, NT-proBNP Amino terminal proB-type natriuretic peptide, AUC Area under the curve

ROC curves were constructed to determine separate cutoffs to distinguish cardiac from non-cardiac causes of pleural effusion for plasma and pleural fluid NT-proBNP and urinary NT-proBNP to creatinine ratio (Figs 4 and 5). ROC curve analysis for plasma NT-proBNP had an area under the curve (AUC) of 0.908 (95% CI: 0.810 to 1.000), and suggested an optimal cut-off of 214.3 pmol/mL [sensitivity, 86.4% (95% CI: 66.7 to 95.3%), specificity, 88.9% (95% CI: 67.2 to 96.9%)]. ROC curve analysis for pleural fluid NT-proBNP had an AUC of 0.953 (95% CI: 0.863 to 1.000), and suggested an optimal cut-off of 322.3 pmol/mL [sensitivity, 100% (95% CI: 85.1 to 100%), specificity, 94.4% (95% CI: 74.2 to 99.0%)]. There was no evidence of a significant difference between the areas under the curves. No cut-off with adequate sensitivity and specificity for UBC was suggested by ROC curve analysis.

The number of animals with measurements of plasma and pleural fluid NT-proBNP above and below these cut-offs in each group, with associated positive and negative likelihood ratios, are listed in Table 2.

Discussion

The results of this study suggest that pleural fluid NT-proBNP, measured using a feline-specific assay, can distinguish cardiac from non-cardiac causes of pleural effusion in cats with similar accuracy to plasma NT-proBNP concentrations. Thoracocentesis is usually indicated in cats with pleural effusion for diagnostic or therapeutic reasons. Measurement of NT-proBNP in pleural fluid rather than plasma may therefore be preferable, as this might remove the need to perform venepuncture, a procedure which can cause undesirable distress in cats with respiratory compromise.

Both plasma and pleural fluid NT-proBNP concentrations had good sensitivity and specificity for the diagnosis of cardiac disease in this study. The cut-off for plasma NT-proBNP concentration suggested by the ROC curve analysis of 214.3 pmol/L is similar to other reported cut-offs of 220 pmol/L with a sensitivity of 93.9% and specificity of 87.8% (Connolly et al. 2009) and 265 pmol/L with a sensitivity of 90.2% and specificity of 87.9% (Fox et al. 2009).

Table 1. Differences in variables between the cardiac and non-cardiac groups. The median and interquartile range are shown for continuous variables

Variable	**Cardiac group** (Number above cut-off/ number below cut-off)	**Non-cardiac group** (Number above cut-off/ number below cut-off)	**Positive likelihood ratio** (95% CI)	**Negative likelihood ratio** (95% CI)	**P**
Plasma NT-proBNP	19/3	2/16	7.773 (2.082-29.014)	0.153 (0.053-0.445)	<0.001
Pleural fluid NT-proBNP	22/0	1/17	18.000 (2.679-120.918)	0 (N/A)	<0.001

CI Confidence interval

In humans, Kolditz et al. (2006) found that pleural fluid and serum concentrations of NT-proBNP were nearly identical. This led the authors of that study to question whether there was any benefit in measuring pleural fluid NT-proBNP concentrations as in humans venepuncture is generally preferable to thoracocentesis. However, as discussed above, this is not necessarily the case in cats. In contrast, in this study, NT-proBNP measurements were higher in pleural fluid than in plasma. This is interesting as pleural fluid NT-proBNP is hypothesised to be derived from plasma proBNP which has diffused into the pleural space before being cleaved into NT-proBNP and BNP (Zemans et al. 2004). Concentrations might therefore be expected to be lower in pleural fluid rather than higher. The reasons for the higher concentrations found in pleural fluid in this study are unknown.

Analysis of urine NT-proBNP to creatinine ratio in this study showed that it was not a useful indicator of whether cardiac disease was the cause of pleural effusion. This is unlike the findings in humans where urinary NT-proBNP has been shown to be a useful indicator of symptomatic heart failure (Cortés et al. 2007; Jungbauer et al. 2010). This may be because the samples in this study were not processed quickly enough after urine was voided as many samples were collected by free catch and processed only once noted in the cat's litter tray. NT-proBNP concentrations have been shown to decrease significantly faster in non-protease inhibited feline plasma than in samples mixed with PI (Connolly et al. 2011) and it is likely that this is also true for urinary NTproBNP. Other possible explanations include the fact that pleural fluid, plasma and urine samples were all collected at different time points, although the investigators attempted to collect samples as close together in time as possible. It is also possible that feline and human excretion of NT-proBNP differs.

A limitation of this study is that plasma, pleural fluid and urine samples were not collected at the same time point, with the maximum time recorded between collection of samples being 42 hours. Echocardiography was also not performed at the same time as sample acquisition. Another limitation is that NT- proBNP measurements were performed in two separate laboratories, which may have increased the inaccuracy of measurements. This would reduce the probability of detecting differences between groups and correlations between measurements. Finally, cats were assigned to the cardiac or non-cardiac pleural effusion groups based on the opinion of the attending cardiologist. This decision was inherently subjective although echocardiography was performed in all cases and was interpreted by an experienced board-certified cardiologist decreasing the risk of misclassification. These limitations prevent a definitive conclusion that urinary NT-proBNP is not useful in the diagnosis of cardiac disease. However, they did not prevent the demonstration that pleural fluid concentration of NT-proBNP is a sensitive and specific indicator.

One case that was classified as having a non-cardiogenic pleural effusion had markedly elevated pleural fluid (>1500 pmol/L) and plasma (522 pmol/L) NT-proBNP concentrations compared to all the other non-cardiogenic cases. This cat had immunemediated haemolytic anaemia and the underlying cause for the pleural fluid was not determined. The cat had a left atrial to aortic ratio of 1.15 (with <1.5 described as normal by Luis Fuentes (2010)), however, the left atrium appeared enlarged in long axis at 21.5 mm (with a normal value of <16 mm described by Luis Fuentes (2010)) and the left ventricular internal diameter in diastole was 21.3mm (upper range of 20 mm reported by Luis Fuentes (2010)) indicating moderate dilation. The attending cardiologist felt that cardiac disease was unlikely to be the cause of the pleural fluid and so the cat was classified as having noncardiogenic pleural fluid. The echocardiographic results in this case were equivocal and it is possible that the cat was misclassified. This represents a potential limitation of the gold standard applied, but it is also possible that the cat's underlying disease process or therapy administered could have resulted in the elevated NT-proBNP concentrations.
General practitioners are able to diagnose cardiac disease in cats with significantly improved accuracy and confidence when the results of plasma NT-proBNP measurements are available (Singletary et al. 2012). It is therefore likely that measurement of NT-proBNP in pleural fluid would similarly assist practitioners. This study analysed samples from clinical cases rather than from animals in which disease was induced, making the results relevant to practicing veterinarians. The diagnosis of cardiogenic pleural effusion on the basis of NT-proBNP measurements should be accompanied by careful assessment of the patient's full clinical history and physical examination to decide whether further investigation of potential exacerbating factors is necessary.

The intra- and inter-assay CV for pleural fluid were acceptable for medium and high concentration samples,

but the inter-assay CV was higher than desirable at 16.7% for low concentration samples. This suggests that the ELISA is less consistent at lower concentrations of pleural fluid NT-proBNP, but concentrations at this level are distant from the cut-off reported making this finding clinically less significant.

In conclusion, the results of this study suggest that NTproBNP can be measured in feline pleural fluid with good precision and acceptable reproducibility. Measurement of NTproBNP in plasma or pleural fluid, but not urine, allows differentiation of cardiac from non-cardiac causes of pleural effusion with similar accuracy in cats. These findings should be validated by prospective testing of the suggested cut-off values in a separate population.

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Conflict of interest

None of the authors of this article has a financial or personal relationship with other people or organisations that could inappropriately influence or bias the content of the paper.

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Reprint paper*

Bloody diarrhoea: what exactly is going on?
Endoscopically visualized lesions, histologic findings and bacterial invasion in the gastrointestinal mucosa of dogs with acute haemorrhagic diarrhoea syndrome

Stefan Unterer[1], Kathrin Busch, Miriam Leipig, Walter Hermanns, Georg Wolf, Reinhard Straubinger, Ralf Mueller and Katrin Hartmann

SUMMARY

Background: Aetiology of haemorrhagic gastroenteritis (HGE) syndrome in dogs is unknown and histopathologic and microbial investigations have only been performed post mortem.

Objective: To identify characteristic intra vitam endoscopic and histologic mucosal lesions, as well as bacterial species, within the mucosa of dogs with HGE.

Animals: Ten dogs diagnosed with HGE were included. Eleven dogs with gastroduodenoscopy and different intestinal diseases were used as controls for microbial changes. Dogs pre-treated with antibiotics or diagnosed with any disease known to cause bloody diarrhoea were excluded from the study.

Methods: In this prospective study, gastrointestinal biopsies were collected from 10 dogs with HGE. Endoscopic and histologic changes were assessed according to WSAVA guidelines. Biopsies from the stomach, duodenum, ileum and colon were investigated by histology and by immunohistochemistry for the presence of *Clostridium* spp. and parvovirus. The first duodenal biopsy taken with a sterile forceps was submitted for bacterial culture.

Results: Acute mucosal lesions were only found in the intestines, not in the stomach. *Clostridium* spp., identified as *Clostridium perfringens* in 6/9 cases, were detected on the small intestinal mucosa in all dogs with HGE, either by culture or immunohistopathology. In the control group, *C. perfringens* could only be cultured in one of 11 dogs.

Conclusions and Clinical Importance: The results of this study demonstrate an apparent association between *C. perfringens* and the occurrence of acute haemorrhagic diarrhoea. The term "HGE," which implies the involvement of the stomach, should be renamed as "acute haemorrhagic diarrhoea syndrome."

Key words: Acute haemorrhagic diarrhoea syndrome; Bloody diarrhoea; *Clostridium perfringens*; Haemorrhagic gastroenteritis.

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Go to http://www.fecava.org/ejcap to see the online presentation of this paper.

[1] Clinic of Small Animal Medicine, LMU Munich, Veterinaerstrasse 13, 80539 Munich, Germany;
e-mail: s.unterer@medizinische-kleintierklinik.de

Abbreviations:

AHDS	acute haemorrhagic diarrhoea syndrome
CBC	complete blood count
CPE	*Clostridium perfringens* enterotoxin
CRT	capillary refill time
ELISA	enzyme-linked immunosorbent assay
HGE	haemorrhagic gastroenteritis
WSAVA	World Small Animal Veterinary Association

Introduction

A clinical syndrome in dogs, characterized by the acute onset of bloody diarrhoea and vomiting, is well known to veterinary practitioners.[1,2] Over the last 40 years, allergic, hereditary, autoimmune and infectious disorders have been proposed as causes for this syndrome. However, the exact pathogenesis remains unknown.[1,3–7] In the first description of a large group of dogs with acute haemorrhagic diarrhoea, the name "haemorrhagic gastroenteritis" (HGE) was used.[1] This terminology has since been questioned, as the intestinal histology of affected dogs investigated at necropsy showed no evidence of an inflammatory reaction in a previous study.[6] The principal intestinal lesions of dogs with HGE at necropsy were described as superficial mucosal haemorrhagic necroses.[5,8,9] An additional histologic finding of the intestinal lesions identified by histopathology was the adherence of large Gram-positive bacilli, identified as *Clostridium perfringens*, to the necrotic mucosal surfaces.[7–9] However, all of the histologic and microbiological evaluations of gastrointestinal tissues from dogs with HGE, thus far, had been performed post mortem, which complicates the interpretation of abnormal findings. Autolysis of the gastrointestinal tract begins rapidly and post mortem degenerative changes can be observed as early as 90 minutes after death. Necrosis and autolysis of mammalian cells present very similar morphological appearances, which can cause difficulties in assessment.[10] Because all types of *C. perfringens* can normally inhabit the intestines of most animals, cultures of this microorganism from the intestinal contents of these animals has no diagnostic value, especially not cultures taken post mortem.[11–14] So far, no studies on the macroscopic appearance or on histologic changes performed intra vitam in dogs with HGE exist.

An association between a clostridial infection and acute haemorrhagic diarrhoea is suspected, as 8/27 dogs with acute haemorrhagic diarrhoea had positive faecal ELISA results for *C. perfringens* enterotoxin (CPE), 7/27 for *C. difficile* toxin A, and 1/27 for both toxins.[5] However, *C. perfringens* can be cultured from the faecal specimens of more than 80% of diarrheic and non-diarrhoeic dogs, and CPE can also be detected in up to 14% of non-diarrhoeic, healthy dogs.[11,15] *C. difficile* can even be found in up to 23% of healthy, non-diarrhoeic dogs.[15–17] In addition, the moderate-to-poor sensitivity and specificity of commercial ELISA, which was used in this previous study, have been reported by comparison with the gold standard of cytotoxicity assays.[18]

Thus, the aim of this study was to describe endoscopically identifiable gross lesions of the gastrointestinal tract and histologic findings in prospectively collected gastric and intestinal biopsy samples from dogs with HGE. A second goal was to potentially identify bacteria in these biopsy samples. These investigations should provide new insight into the pathogenesis and role of bacteria in this syndrome.

Materials and Methods

Patients

This study was conducted according to German animal welfare laws. Each owner was informed of the purposes of the study. Between August 2010 and December 2012, 10 dogs with acute haemorrhagic diarrhoea without an identifiable cause, whose owners agreed to have endoscopy performed, were presented to the emergency service of the Clinic of Small Animal Medicine, LMU University of Munich, Germany. The inclusion criterion was an acute onset of haemorrhagic diarrhoea (<3 days since presentation). Patients pre-treated with antibiotics and having haemorrhagic diarrhoea caused by a disease aetiology unrelated to HGE were excluded from this study. Exclusion diagnoses included nonsteroidal anti-inflammatory or corticosteroid toxicosis, hypoadrenocorticism, inflammatory bowel disease, severe hepatitis, hepatic neoplasia or hepatic failure, acute and chronic renal failure, pancreatitis, anticoagulant toxicosis, gastrointestinal neoplasia or foreign bodies and enteric infection with parvovirus, *Giardia* spp. or endoparasites. To rule out these possible causes of haemorrhagic diarrhoea, all dogs underwent a physical examination, abdominal ultrasound examination, CBC, serum biochemistry profile, serum bile acid concentrations, clotting profile and faecal examination for nematode and protozoan parasites (29.5% sodium nitrate flotation solution,a Giardia antigen ELISAb) and for parvovirosis (antigen ELISAc). In addition, the presence of parvovirus was excluded by immunohistochemistry (IHC).

Immunolabeling of the mucosal tissue was performed through indirect immunostaining using mouse anti-canine/-feline parvovirus (MCA 206d) as primary and peroxidase-labelled rabbit anti-mouse immunoglobulins (P0s6e) as secondary antibodies.

Treatment was standardized for all dogs and included fluid therapy (crystalloids; the fluid amount depended on dehydration, maintenance demands and on-going losses) and anti-emetics (maropitantf 1 mg/kg SC q24h, on days 1 and 2). Analgesics were administered dependent on clinical judgement of abdominal pain. The clinical assessment was performed by calculating the "canine HGE activity index."[2] This index includes the parameters attitude, appetite, vomiting, stool consistency, stool frequency and dehydration. Each parameter was scored (0 = normal, 1 = mild, 2 = moderate, 3 = severe) and the sum of scores yielded a total cumulative score (maximum = 18).
Eleven dogs that presented during the same time period and in whom gastroduodenoscopy was performed for other reasons than haemorrhagic diarrhoea served as a control group for microbiological evaluations. Endoscopic biopsies in these dogs were used to assess the significance of microbial findings. All samples, including endoscopic biopsies, were collected in a manner identical to that in the HGE patients. Dogs pre-treated with antibiotics were excluded from the control population.

Endoscopy

As early, after initial presentation, as anaesthesia was considered safe (good pulse quality, heart rate 70–140/min, respiratory rate <25/min, CRT <1 second, rectal temperature 37.5–39.5°C), endoscopy of the upper and lower intestinal tracts of dogs with HGE was performed and evaluated according to the WSAVA International Gastrointestinal Standardization Guidelines.19 The flexible endoscope Olympus GIF Type 160g was used for dogs <20 kg and the Olympus PCF Type 140 Lg was used for dogs >20 kg. Each endoscopic parameter (hyperaemia/vascularity, discoloration, oedema, friability, texture changes, haemorrhage and erosions/ulcers) was scored as 0 = normal, 1 = mild, 2 = moderate or 3 = severe by two of the authors performing the endoscopies (SU & KB).
At least 6 biopsy specimens were collected from the stomach and each intestinal segment and then submitted for histology. Because of the size of the dog and the duration of the anaesthesia, endoscopy of the ileum and colon was not possible in every patient. In some of these cases, biopsies were obtained blindly. In addition, the first duodenal biopsy was taken with a sterile single-use biopsy forceps,h transferred into a sterile tube without transport medium and immediately submitted for bacterial culture.

Histologic Examination of Biopsies

Tissue samples were fixed in neutral buffered 10% formaldehyde for 24 hours, embedded in paraffin and plastic, sectioned and stained with the haematoxylin and eosin and Giemsa. Two pathologists (ML and WH) performed the histologic examination of all tissues.
The characterization of the histologic changes in the endoscopic biopsy samples was performed according to the histopathologic standards established by the WSAVA Gastrointestinal Standardization Group.[20]

Immunohistochemistry for *C. perfringens*

Immunohistochemistry was performed according to standard protocols for indirect IHC assays. All incubations were completed at room temperature. Following manual deparaffinisation and rehydration, 4-µm-thick tissue sections mounted on positive-charged glass slides were treated with 1% hydrogen peroxide to quench endogenous peroxidase activity and washed in a bath of Tris-buffered saline (TBS, 0.5 M, pH 7.6). After incubation with goat normal serum (1 : 10 dilution)i for 30 minutes, the slides were incubated with polyclonal anti-*Clostridium* spp. antibody (polyclonal rabbit antibody against *C. perfringens*, *C. sordellii*, *C. novyi*, *C. septicum* and *C. chauvoei*; 1 : 100 dilution; No. 2119-2701)d as the primary antibody for 60 minutes. Subsequently, the slides were washed in baths of TBS, incubated with peroxidase-labelled goat anti-rabbit immunoglobulin (1 : 100 dilution; No. P0448)e as the secondary antibody for 1 hour and washed again in a bath of TBS. The binding of peroxidase coupled to the secondary antibodies was visualized by the reaction of H2O2 and 3'3'-diaminobenzidinetetrahydrochloride j as chromogens. Slides were counterstained with Mayer's haematoxylin.
For clostridial IHC, positive controls included a clostridial suspension (isolated from dogs with HGE) injected into swine muscle and processed routinely (fixed in formalin and embedded in paraffin).

Bacterial Culture

Within 30 minutes after taking the biopsy, samples were plated onto agar plates using sterile tweezers. Nutrient agar with 5% defibrinated sheep blood for aerobic and Schaedler agar with 5% sheep blood for anaerobic cultivation were used.k Plates were incubated at 38°C. Colony growth was monitored for 3 days. Every colony

type was sampled and differentiated by mass spectrometry using MALDI-TOF.l Microbial identification by MALDI-TOF is considered a very specific method comparable to conventional diagnostics.[21]

Statistical Evaluation

The numbers of positive bacterial cultures and positive IHC in dogs with HGE and control dogs, respectively, were compared with a Fisher Exact test. Improvement of the HGE index from day 1 to day 3 was compared with a repeated measures ANOVA. For one dog that was sent home because of almost complete remission, the value of day 2 was carried forward to day 3 to allow appropriate statistical analysis. For all tests, a $P < 0.05$ was considered significant.

Results

Study Population

Dogs with HGE included 5 females and 5 males, of which 3 and 2 were neutered, respectively. The median age and weight were 5 years (range 1–10) and 11.0 kg (range 2.9–30.5), respectively. Breeds included mixed breeds (n = 3), Yorkshire Terrier (n = 2) and one each of Jack-Russell Terrier, Labrador retriever, Dachshund, West Highland white terrier and Miniature Australian Shepherd. The median duration of clinical signs until presentation was 12 hours (range 6–36). All 10 dogs also exhibited vomiting (6/10 bloody vomiting) and haemorrhagic diarrhoea. In general, a rapid improvement of clinical signs was observed during the first 48 hours (median HGE score at presentation: 12 [range 6–17]; after 48 hours: 5 [range 0–10]). The improvement from day 1 to day 2 and day 1 to day 3 was highly significant ($P < 0.01$ and $P < 0.001$, respectively). All dogs recovered and were discharged. The median duration of hospitalization was 3 days (range 2–6).

Control Group

Dogs included 2 females and 9 males, of which 2 each were neutered in both groups. The median age and weight was 10 years (range 1–13) and 27.7 kg (range 5.0–62.5), respectively. Breeds included mixed breeds (n = 3) and one each of Yorkshire Terrier, Beagle, Jack-Russell Terrier, Rhodesian Ridgeback, Leonberger, Malinois, Magyar Viszla and Dalmatian. Final diagnoses included gastrointestinal foreign body (n = 3), adverse food reaction (n = 2) and one each of esophagitis, gastritis, inflammatory bowel disease, ulcerative colitis and hypertrophic pylorus.

Endoscopy

In all dogs, an endoscopy was performed within the first 12 hours of presentation. In 6/10 dogs, the mucosa of the oesophagus was assessed as normal; 4/10 dogs showed signs of mild hyperaemia. The stomach was macroscopically normal in 7/10 dogs; 3/10 showed mild changes characterized by mild-to-moderate oedema and hyperaemia. Gastric erosions or ulcerations could not be detected in any dog, although 6/10 dogs were presented with haemorrhagic vomiting. Duodenoscopy was performed in 10/10, colonoscopy in 8/10 and ileoscopy in 6/10 dogs. The most important macroscopic findings in the intestinal tract, which could be observed in every dog, included hyperaemia, increased mucosal friability, haemorrhage and erosions.

Table 1. Endoscopically visualized lesions and histologic changes in the gastrointestinal mucosa of dogs with acute haemorrhagic diarrhoea syndrome.

Endoscopic Changes	**Duodenum (n = 10)**				**Ileum (n = 6)**				**Colon (n = 8)**			
Severity	**No**	**Mild**	**Mod.**	**Severe**	**No**	**Mild**	**Mod.**	**Severe**	**Norm.**	**Mild**	**Mod**	**Severe**
Hyperemia	1	0	7	2	1	1	4	0	0	4	4	0
Friability	0	3	5	2	0	2	4	0	1	5	2	0
Haemorrhage	3	3	2	2	0	3	3	0	2	2	3	1
Erosion/ulcers	3	3	2	2	1	2	3	0	2	2	4	0

Histologic Changes	**Duodenum (n = 10; ns = 64)**				**Ileum (n = 8; ns = 29)**				**Colon (n = 9; ns = 55)**			
Severity	**Norm.**	**Mild**	**Mod.**	**Severe**	**Norm.**	**Mild**	**Mod.**	**Severe**	**Norm.**	**Mild**	**Mod**	**Severe**
Epith. injury	2	3	3	2	2	4	2	0	0	3	2	4
L. p. neutroph	2	6	1	1	1	3	3	1	0	4	5	0
Vill. stunting	4	2	2	2	1	2	4	1				

n: number of dogs; ns: number of adequate samples per location in total; epith. Injury: epithelial injury; L. p. neutroph.: Lamina propria neutrophils; vill. stunting: villous stunting; norm.: no changes; mod.: moderate changes.

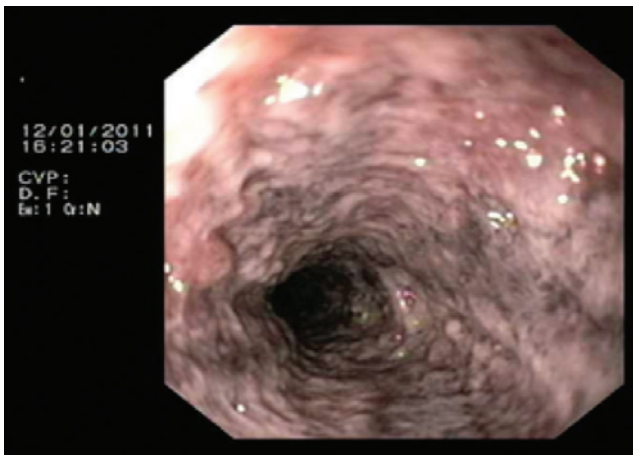

Fig 1. Dog #4. Endoscopic appearance of the duodenum, showing oedema, hyperaemia, increased vascularity and erosions.

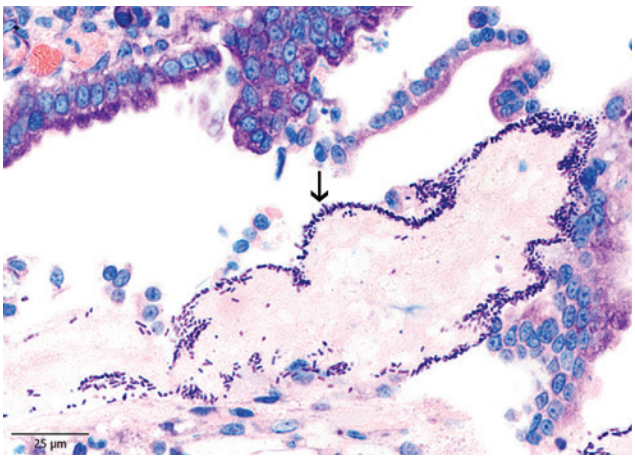

Fig 2. Giemsa staining of a duodenal section collected from Dog #2. A dense layer of large rod-shaped bacteria adherent to a necrotic villous tip (black arrow).

Compared with the gross lesions in the macroscopic colon, the lesions in the duodenum appeared to be more prominent (Table 1, Fig 1).

Histology

Lesions were restricted to the small and large intestine and were not found in the stomach. Neither relevant signs of acute inflammation nor destruction of the mucosal epithelial surface could be detected in gastric biopsy samples. The most important histologic changes in the intestine included acute mucosal necrosis (10/10) and neutrophilic infiltration (10/10). Histologic lesions reflecting acute mucosal intestinal destruction in the intestine are presented in Table 1. A prominent feature of the small intestinal lesions was the adherence of large rod-shaped bacteria to the necrotic mucosal surfaces, which could be observed in 9/10 dogs (6/10 in duodenal, 5/8 in ileal biopsies; Table 2, Fig 2). The bacteria built a dense layer on the surface of the necrotic lesions. Bacteria adhering to the duodenal mucosa were not detected by histology in any dog of the control group. There was a significant difference in the microscopic detection rate of clostridial-type bacteria in duodenal biopsies between patients (6/10) and control dogs (0/11) (P = 0.004) (Table 3).

Table 2. Detection of Clostridium spp. *in small intestinal biopsy samples by histology and culture (all confirmed as* C. perfringens *by mass spectrometry using MALDI-TOF) in the 10 individual patients with acute haemorrhagic diarrhoea syndrome. The first duodenal biopsy was taken with a sterile forceps and submitted for bacterial culture.*

Dog Number	1	2	3	4	5	6	7	8	9	10
Histology duodenum	+	+	-	+	-	-	+	+	-	+
Culture duodenum	NE	+	+	+	+	-	-	+	-	+
Histology ileum	-	+	-	NE	+	+	NE	+	+	-
Either histology or culture	+	+	+	+	+	+	+	+	+	+

+ : visualization of large rod-shaped bacteria/culture of *C. perfringens*; - : no visualization of large rod-shaped bacteria/ no culture of *C. perfringens*; NE : not evaluated

Table 3. Comparison of Clostridium spp. *Detection on the duodenal mucosa between dogs with haemorrhagic gastroenteritis and control dogs.*

	Patient Group		Control Group		
	Positive	Negative	Positive	Negative	P-value
Culture duodenum	6	3	1	10	.017
Histology ileum	6	4	0	11	.004
Either histology or culture	8	2	1	11	.002

All cultured *Clostridium* spp. were identified as *C. perfringens* by mass spectrometry using MALDI-TOF.

Immunohistochemistry

The histologically detected bacteria were identified as clostridial antigen positive by immunohistochemical staining (Fig 3).

Fig 3. Immunohistochemical staining of a duodenal section collected from Dog #2. Many rod-shaped bacteria present on the necrotic villous tissue were strongly positive for a polyclonal chicken antibody raised against Clostridium spp.

Bacterial Culture

A bacterial culture from duodenal biopsies was performed in 9 dogs with HGE and 11 control dogs. In 6/9 dogs with HGE, *C. perfringens* could be cultured. In 1/11 control dogs, a growth of *C. perfringens* was observed. There was a significant difference in the number of positive *C. perfringens* cultures from duodenal biopsies between patient and control group (P = 0.016) (Table 3).

Discussion

Results of this study demonstrate an association between clostridial overgrowth and HGE. Necrotic epithelial lesions were restricted to the small and large intestine. Biopsy samples from dogs with HGE showed a lack of gastric mucosal involvement in this study. In addition to profuse, watery-mucoid, bloody diarrhoea, many dogs with HGE demonstrate acute vomiting.[1] Therefore, it was assumed that the stomach is involved in this disorder. Because the clinical signs could most likely be explained by an acute inflammation of the entire gastrointestinal tract, the name "HGE" was given to this syndrome.[1]

A very early report about the microscopic evaluation of the gastrointestinal mucosa from dogs with acute haemorrhagic diarrhoea (obtained post mortem) mentioned that "no evidence of an inflammatory reaction" could be detected.6 A description of 2 other cases with pre-acute haemorrhagic enteritis and their necropsy results was published 6 years later. One dog died within 15 minutes after admission, whereas the second dog was found dead, surrounded by pools of bloody diarrhoea and vomitus, at home by his owners before being presented for necropsy. Microscopic changes in the gastric mucosa could only be detected in the second dog. Gastric lesions were described as superficial epithelial necrosis and it remains unclear whether autolytic changes that were present at the time of necropsy caused difficulties in the assessment of the post mortem lesions.[8] In all other reports on the histologic changes of the gastrointestinal tract in necropsied dogs with acute haemorrhagic diarrhoea, descriptions concerning gastric changes are lacking or the results are conflicting. One explanation is that all previously reported histopathologic evaluations were based on necropsied dogs and post mortem changes can modify the microscopic appearance of the gastrointestinal mucosa of animals very rapidly after their deaths. In pigs, significant epithelial loss of the stomach begins to occur only 90 minutes after death.[10] Gastric necrosis observed in necropsied dogs with haemorrhagic diarrhoea could therefore represent autolytic morphological changes. This study, for the first time, evaluated the gastrointestinal histopathologic changes from samples collected intra vitam and immediately fixed in formalin. The results clearly show no signs of significant acute inflammation or destruction of the gastric mucosal epithelial surface. This finding was surprising because all of the dogs exhibited vomiting, even haemorrhagic vomiting in 6/10 cases, indicative of stomach involvement. Because (1) no acute gastric lesions could be detected macroscopically or histologically, (2) the prominent lesions found in the intestines may themselves cause vomiting and (3) the presence of blood in the vomitus could be explained by the duodenal erosions, it is likely that lesions associated with this syndrome are generally restricted to the intestines and that the stomach is not primarily involved in the disease process.

Some reports have mentioned that lesions in dogs with HGE mainly affect the small intestine,[5,9] whereas other reports have stated that these lesions are particularly severe in the colon.[8] In the present patient population, we demonstrated that the entire intestinal tract was affected in dogs with HGE and that mucosal damage is generally more severe in the large intestine. This study confirms, for the first time intra vitam, that intestinal mucosal haemorrhagic necrosis is the principal histologic lesion found in dogs with HGE. In contrast to previous histologic studies,[5,6] some infiltration of neutrophilic granulocytes in the intestinal mucosa was observed; however, in relation to the severe epithelial destruction, the influx of inflammatory cells was disproportionally minor. Acute mucosal destruction, in general, could be explained by ischemia,[22] hyperthermia,[23] acute parvoviral infection,24 and enterotoxins.[25–28] In this study

population, systemic hypoxia and hyperthermia were ruled out by patient evaluation and history. Local thrombosis in the intestine was not likely, as no underlying disease predisposing for thromboembolism could be identified in any of the cases. Parvovirus infection was ruled out by faecal ELISA and IHC. Therefore, enterotoxins are the most logical explanation of the destructive lesions found in the dogs.

Clostridium spp., identified by culture as *C. perfringens* in 6/9 cases, were detected in the small intestinal biopsies of all dogs with HGE either via culture or via IHC of formalin-fixed histologic samples. In contrast, clostridial-type bacteria were not detected by histology or IHC in any dog of the control group and only a mild growth of *C. perfringens* could be observed in a single duodenal biopsy sample. Dogs have highly diverse duodenal microflora that differ markedly among individual dogs.29 Although a high number of *Clostridium* spp. can be a part of the normal colonic flora,[11] *C. perfringens* can only be rarely cultured from the duodenal juice,30 and bacterial layers on the surface of the duodenum are normally not present in healthy dogs. [31] Dogs with parvovirus enteritis show similar clinical signs (acute haemorrhagic diarrhoea) and similar histologic small intestinal lesions (epithelial necrosis) as dogs with HGE. However, no adherence of bacteria to necrotic villous tips is observed in necropsied dogs with parvovirosis.[32] Therefore, the microscopic detection of a large number of clostridial-type bacteria in the small intestine of a majority of the patients in this study was judged as clostridial overgrowth on the mucosal surface. It is known that inflammation drives dysbiosis[33] and dogs with acute non-haemorrhagic diarrhoea have profound alterations in their microbiome. [34] In the presented group of dogs, samples were obtained after the onset of diarrhoea. Thus, it is possible that the clostridial overgrowth is a sequela of the disease rather than the cause.

In one-third of the dogs, *Clostridium* spp. was detected by IHC of the small intestine, but there was no clostridial growth upon duodenal culture. These negative cultures could be attributable to dehydration or the exposure of the anaerobic bacteria to lethal concentrations of oxygen. Alternatively, it is possible that *Clostridium* spp. were not equally distributed or present at each site within the small intestinal tract. In 3/10 cases, clostridial bacteria could be histologically detected in the ileum, but not in the duodenum, which was the only part of the small intestine from which the samples for culture were taken. Enterotoxaemia caused by *C. perfringens* infection causes necrotic enteritis as a result of several toxins in many animal species and humans. It has now been clearly established that certain *C. perfringens* strains are capable of inducing necrotic enteritis in broilers, pigs, lambs, horses, felines and humans.[13,35–38] Currently, the cause of *C. perfringens* overgrowth in the gastrointestinal tracts of humans or other mammals, including dogs, is not always known. It has been proposed that physical stress, decreased immunoreactivity and intestinal hypermotility reduce the normal anaerobic bacterial flora and may predispose the subject to the bacterial overgrowth of *C. perfringens* in the small intestine In the present patient population, no history of previous physical exhaustion was evident and no underlying disorder predisposing for immunosuppression could be found. High-carbohydrate rations and diet changes have also been associated with the overgrowth of *C. perfringens* in different species.[39–41] Owners reported dietary changes at the onset of haemorrhagic diarrhoea in only 8/111 dogs in 1 study.[1] A specific dietary factor could not be identified as the cause for the acute intestinal signs in this study population. Therefore, it seems unlikely that components of the diet are predisposing dogs with acute haemorrhagic diarrhoea to clostridial overgrowth. However, *C. perfringens* is the third most common cause of food-borne illness in humans in the United States,[42] and food poisoning cannot be completely ruled out among the dogs in this study. *C. perfringens* can cause self-resolving enteritis in dogs,[11] and it is possible that haemorrhagic diarrhoea represents the extreme end of a spectrum of infection. Additional research might uncover virulence factors and toxin production caused by *C. perfringens*, as well as host factors predisposing for acute haemorrhagic diarrhoea in dogs.

The following facts suggest a primary pathogenic role for *C. perfringens* in dogs with HGE: (1) the characteristic lesions in the intestine that have been observed in different species after infection with *C. perfringens* could also be identified in the patient population of this study; (2) *Clostridium* spp. (identified in 6/9 patients as *C. perfringens* by mass spectrometry using MALDI-TOF) was detected in the small intestinal biopsy samples of every dog with HGE; (3) the presence of bacteria and epithelial lesions was closely associated; (4) histologic changes were not influenced by autolysis or microbial overgrowth after post mortem changes in the prospectively intra vitam-collected specimens; and (5) no other explanation of the necrosis of the superficial intestinal epithelium could be identified. The rather small number of dogs in the patient group is a

possible limitation of this study. The overwhelming majority of dogs showed the same findings, thus it seems unlikely that the results would have changed with a larger patient group.

Dogs with different intestinal disorders, in which intestinal biopsies were diagnostically indicated, were used as controls for microbial changes. To evaluate if clostridial overgrowth is a sequela of the disease rather than the cause, it would have been more informative to include dogs with known causes of haemorrhagic diarrhoea as control dogs.

Another limitation was the fact that ileoscopy was not routinely performed in dogs of the control group. Therefore, data for comparison of microscopic detection of *Clostridium* spp. in the ileum were not available.

Conclusion

In conclusion, this report describes endoscopically visualized changes, histologically confirmed lesions and microbial changes intra vitam in dogs diagnosed with the so-called HGE syndrome. The results of this study demonstrate an apparent association between *C. perfringens* and the occurrence of acute haemorrhagic diarrhoea. The mucosal lesions were restricted to the large and small intestines. Therefore, the term "HGE," which is often used in the current literature and implies the involvement of the stomach, is misleading. We suggest that the syndrome should be renamed "acute haemorrhagic diarrhoea syndrome."

Acknowledgment

Conflict of Interest Declaration: Authors disclose no conflict of interest.

Footnotes

a Natriumnitratflotationslösung; Janssen-Cilag, Neuss, Germany
b ProSpecT Giardia Microplate Assay; Remel Inc, Lenexa, KS
c Snap Parvo Test; IDEXX Laboratories Inc, Westbrook, ME
d AbD Serotec, Duesseldorf, Germany
e Dako, Glostrup, Denmark
f Cerenia; Pfizer Pharma GmbH, Hamburg, Germany
g Olympus Flexible Medizinische Endoskopie, Hamburg, Germany
h Wieser GmbH Medizintechnik & Geräte, Egenhofen, Germany
i No. 08642921, MP Biomedicals, Illkirch, France
j No. 4170, Biotrend Chemikalien, Köln, Germany
k Anaerocult P and Anaerocult C, Merck, Darmstadt, Germany
l Microflex LT, Flex Anaysis Software, Bruker-Daltonics, Bremen, Germany

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Commissioned paper

The Colourful receptionist©

Brian Faulkner[1]

SUMMARY

Every veterinary practice must consistently deliver four outcomes if they wish to be effective and sustainable. These are clinical resolution, client satisfaction, financial resolution and team harmony and happiness. Furthermore every employee needs to contribute to each of these outcomes in one way or another. In the author's experience however, veterinary practices tend to be either 'reactive' or 'proactive' with respect to how they go about training their staff to think about and achieve all of these outcomes. A previous article in EJCAP (2014), Winter 24(4); p46-p54 focused on how the author's Colourful Consultation© model is used to proactively pursue these four outcomes in the consultation room. The focus of this article is to describe the author's Colourful Receptionist© model has been designed to enable a proactive approach to coaching and training members of staff who work 'front of house'.

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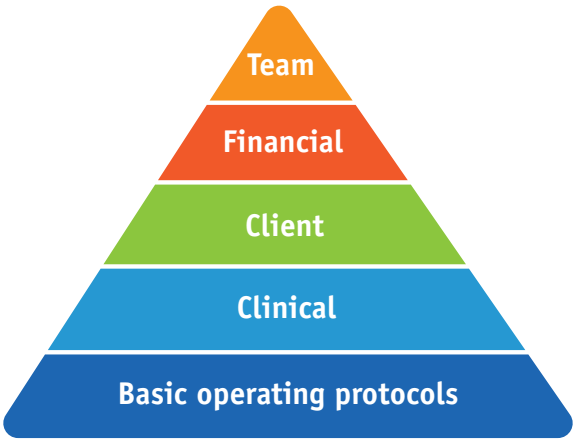
The Colourful Receptionist© model



Level 1 – Basic operating systems

The model starts by taking a proactive approach ensuring that members of staff are able to master the basic operating systems of their veterinary practice.

All too often, new receptionists can get flustered as they struggle with the systems such as these, on top of the avalanche of technical queries or client demands. Receptionists who have mastered these basic skills are able to free their minds so that they can concentrate on dealing with technical queries as well applying the more subtle inter-personal skills enabling them to be more effective.

These Basic operating systems are captured using the acronym RECEPTION which stands for the following:

R	Rule number 1: Book an appointment!
E	Easy to understand and follow directions
C	Computer system basics
E	Emails received /sent /address, faxes and scans
P	Payments: cards, cash and cheques
T	Telephones: understanding and using your telephone system
I	Information about the practice and headline prices
O	Ordering products from wholesalers
N	Notes, memos and directing queries

[1] Brian Faulkner BSc (Hons), BVM&S, CertGP(SAM), CertGP(BPS), MBA, MSc(Psych), MRCVS. 22 High Street, Wickham Market Woodbridge, Suffolk IP13 0HE, United Kingdom, E-mail brian@vetpsych.com

Rule number 1: Book an appointment

The 'default' answer to any query about a patient should be to recommend an appointment. It is a common mistake to assume that clients will ask for appointments if they feel they need one. Giving over-the-counter or over-the-phone advice is risky as it potentially harms the reputation of the practice as well as exposing it to claims of negligence if the advice is ineffective or results in harm. It is also bad for business.

Easy to understand and follow directions

It is important however to be able to give clear and succinct directions in cases of an emergency or when clients or visitors are visiting the premises for the first time. Inaccurate, vague or overly presumptive directions are frustrating and give a bad impression. Remember that not everyone will drive to the surgery and directions need to be tailored for the various modes of transport relevant to your local transport system. It is also important to be aware of how accurate your address is on Google Maps or popular Sat Nav systems and point this out to visitors.

Computer system basics

The computer system is essential to the smooth running of our practice as it contains all the information about our client accounts, their pets and their clinical notes. The computer system is comprised of hardware and software. Hardware refers to the physical equipment such as user terminals, printers and scanners. Software refers to the programmes which are run on the PCs such as the practice management system and Microsoft Office©. Whilst no-one expects veterinary receptionists to become IT experts, it is important to know some do's and don'ts which will help keep the system working well and what to do when things don't work. Shutting a terminal down safely, waiting for a few minutes and re-booting it is a simple and often effective first step.

Email

Email is now a most common form of written communication in veterinary practice. Email is used to send and receive general queries about the practice, online registrations, lab reports and clinical histories. Everyone working 'front of house' should know the practice email address, be able to access the practice email account, check it regularly, reply to, forward and file email correspondence.

Payments: cards, cash and cheques

It is important to know how to take a payment using the credit and debit card machine both in person and over the phone and why some types of payment are preferred to others. It is also important to be aware of the policies regarding when payment is expected (at the time of consultation) as well as any exceptions (such as charity accounts, account clients and perhaps at the time of euthanasia.)

Telephones: understanding and using your telephone system

The telephone is today the most common mode of receiving enquiries and booking appointments. Whilst everyone can pick up and speak into a telephone in a private context, it is essential to know how to use the practice's telephone system effectively and efficiently in order to avoid stress and frustration to clients by cutting them off or losing them 'in the system'. Furthermore, someone who is flustered with technology and struggling with the equipment itself is less able to pay attention to the more subtle aspects of client communication.

Information about the practice and headline prices

It is important to know the basic information about the practice, its basic history and who does what in order to not only sound knowledgeable and competent to clients and suppliers but also to clarify misunderstandings and assumptions clients make about the practice, its services and members of staff. It is also important to know what procedures can and cannot be 'quoted' for without seeing the patient. It is useful to have a one-page price list (see below) of items that can be 'quoted' to clients. Queries about the price of all other services require an appointment in order to provide an estimate.

Ordering from suppliers

Veterinary practices consume and receive requests for both medical and non-medical supplies every day. Veterinary receptionists are often involved in taking orders for products that may need to be ordered in to meet that demand. All practices need a system that ensures that the right amount of the right products arrive at the right branch at the right time and are dispensed at the right dose for the right patients belonging to

Consultation					
Full Consultation	€xx	Euthanasia - dog	€xx	Clip claws	€xx
Second consultation	€xx	Euthanasia - cat	€xx	Express anal glands	€xx

Cremations					
No ashes returned		**Ashes to scatter**		**Ashes in casket with name plate**	
Dog – 0-10kg	€xx	Dog – 0-10kg	€xx	Dog – 0-10kg	€xx
Dog – 10-40kg	€xx	Dog – 10-40kg	€xx	Dog – 10-40kg	€xx
Dog – 40+kg	€xx	Dog – 40+kg	€xx	Dog – 40+kg	€xx
Cat – all sizes	€xx	Cat – all sizes	€xx	Cat – all sizes	€xx

Vaccinations					
Dog vaccines		**Cat vaccines**		**Rabbits**	
Puppy course	€xx	Kitten course (RCP)	€xx	Myxo	€xx
Booster	€xx	Kitten course (RCPFelv)	€xx	VHD	€xx
Kennel cough (alone)	€xx	Cat booster (RCP)	€xx	Combined	€xx
Kennel cough (booster)	€xx	Cat booster (RCPFelv)	€xx		

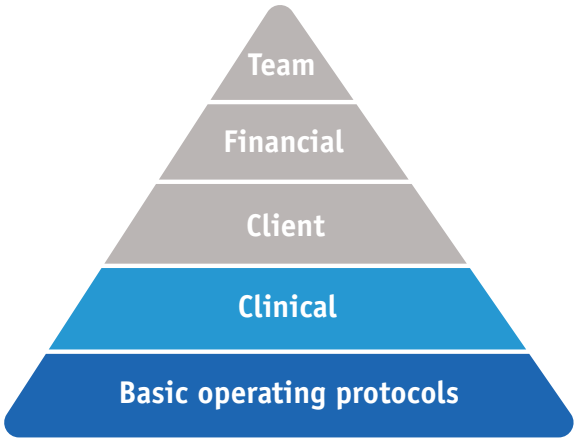
Neutering					
Bitch spays		**Dog castrate**		**Cats**	
0-10kg	€xx	0-10kg	€xx	Cat spay	€xx
10-25kg	€xx	10-25kg	€xx	Cat castrate	€xx
25-40kg	€xx	25-40kg	€xx		
40+kg	€xx	40+kg	€xx	Pre-anaesthetic bloods	€xx

the right clients – or are set aside in the right place ready for collection. Everyone working front of house needs to understand and be able to use that system as well as knowing which products are ordered from which wholesaler(s).

Notes, memos and directing queries

Effective and efficient communication within the practice is essential to maintaining and improving standards of veterinary work, customer service, financial performance as well as team harmony and morale. Internal communication can take the form of face-to-face explanations, notices, notes, memos, internal emails, in-trays, meetings and the minutes created from those meetings. It is important to know how and where to pass on information to colleagues as well as implement, action or respond to it.

Level 2 – Clinical resolution



The second level of the Colourful Receptionist© model looks at how receptionists contribute to 'Clinical resolution'. Whilst there is no intention to make them clinical experts, a proactive philosophy to clinical resolution recognises the importance of understanding the

basics of preventative healthcare. It also ensures that any clinical issues get to see the right vet, in the right place at the right time having been prepared using the right information. The preventative healthcare side of clinical resolution will be discussed using the following acronym; PREVENT IT.

The acronym PREVENT IT which stands for the following;

P	Preventative healthcare plans
R	Reproductive management and neutering
E	Ectoparasites (fleas, ticks, mites and lice)
V	Vaccinations
E	Endoparasites (worms)
N	Nutrition
T	Teeth
I	Insurance and Identification
T	Training and behaviour

Preventative healthcare plans

Many practice offer a Preventative Healthcare Plan whereby pet owners can spread the cost of preventative healthcare products and services as well as receiving a discount compared to the 'pay as you go' arrangement. Veterinary staff who work front of house are integral to promoting, administrating and operating these plans and it is therefore essential that they understand the terms and conditions as well as how to set them up and use them.

Reproductive management and neutering

Managing and preventing conditions associated with the reproductive system in dogs and cats is a common feature of veterinary work. Whilst it is not expected that veterinary receptionists know the full details of these conditions, it is useful to understand the basics of normal form and function of the reproductive system in the core species the practice deals with, as well as the meaning of core terms such as pyometra or phantom pregnancy. Finally, receptionists should be aware of – and mention the value of – the various services the practice provides to prevent and resolve these issues. They should also help the amateur breeder to become aware of services, such as ovulation checks, pregnancy diagnosis and herpes vaccinations for example.

Ectoparasites (fleas, ticks, mites and lice)

External parasites are a cause of distress to both the pets and their owners. Since there is a wide range of ectoparasiticides available from veterinary practice as well as pet shops, it is important that veterinary receptionists know and can explain the features and benefits of the various products sold at their practice. Understanding the value of these products is particularly important given that many veterinary products tend to be more expensive than retail products.

Vaccinations

Dogs, cats and rabbits are routinely vaccinated against various infectious diseases. Often veterinary receptionists are the client's first point of contact regarding these diseases, the vaccines available as well as how and when they are administered. Giving veterinary receptionists a simple 'one-page' guide about these diseases and vaccines helps them answer basic queries which increase the chances that clients engage with these protocols.

Endoparasites (worms)

As with ectoparasites and vaccinations, giving veterinary receptionists a simple 'one-page' knowledge about roundworms, tapeworms and lungworms helps them answer the basic queries about these parasites as well as how and when to prevent them. It is also useful that they understand and can explain the features and benefits of the various wormers products sold at their practice compared to retail products.

Nutrition

Most veterinary practices sell pet food. Again giving veterinary receptionists a simple 'one-page' knowledge about how providing a nutritionally balanced diet relates to a healthy lifestyle. It is imperative that they understand and can explain the features and benefits of the brand sold at their practice.

Teeth

Therapeutic and preventative dental healthcare in pets is a major feature of veterinary work. Veterinary receptionists can help promote good dental care by being aware of the various dental products that the practice sells as well as a basic understanding of the features and benefits of each.

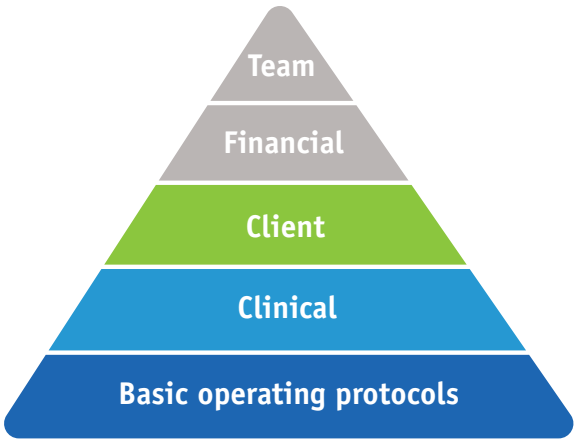
Identification and Insurance

Microchipping pets is already compulsory or is becoming compulsory in some EU states. Veterinary receptionists are often involved in completing paper work as well as online registration. Insurance administration, such as issuing cover notes and filling in claim forms is an increasingly common feature of veterinary receptionist work. It is therefore important that they are familiar with the protocols relating to both these services.

Training and behaviour

The aim of this protocol is to make veterinary receptionists aware of two aspects relating to pet behaviour; the 'critical socialisation period' of puppyhood and the fact that many if not most 'behavioural' conditions in dogs and cats have an underlying medical cause. In other words, when clients engage veterinary receptionists / veterinary practices asking about behavioural issues, the default rule still applies; offer an appointment so that the vet can examine for and rule out medical conditions.

Level 3 - Client satisfaction



Every veterinary practice requires a population of registered and 'active' clients and patients to survive and thrive. Developing and maintaining this population requires a quality of client care that ensures they are kept satisfied with the service they receive, are willing to come back to the practice and recommend it to others.

The next level of the Colourful Receptionist© model aims to ensure that receptionists are familiar with the meaning of 'client satisfaction' and how it can be achieved.

A reactive mindset to customer care hopes clients will be happy as long as everyone is 'nice and polite'. A proactive mindset, however, is much more focused and is based on the understanding that satisfaction occurs when a client feels that what they have been advised to do 'feels right to them' at a price that they perceive as 'fair'. Of course 'right' and 'fair' are very subjective in that what one client perceives as 'right' may not sound 'right' to another.

The model looks at how receptionists can become more proactive in achieving client satisfaction is discussed using the acronym CLIENT.

C	Contact to consults
L	Listening
I	Inter-personal skills
E	Emotional Intelligence
N	New clients
T	Telephone skills

Convert contact into consults

The ultimate aim of customer service from the businesses' point of view is to convert client contact with the practice into paid-for business in a way that satisfies them so that they will use the practice again and recommend it to others. One of the core objectives of a veterinary receptionist is to engage with and respond to client contacts in a focused, purposeful and effective manner in order to convert 'contacts-to-consults'. 'Consults' means an appointment with the practice be it a 'sick-patient' veterinary consultation, a first vaccination, a booster, a nurse consultation or even a surgical operation such as a neutering… which will need an admission consultation.

Listening

Dealing with clients all day every day is challenging not least because they exhibit a wide spectrum of attitudes, expectations and styles. If we wish to help someone resolve their issue in a way that feels right to them, it is essential that we develop the listening skills which help us understand what their issue is and adopt our recommendations in order to resolve it.

Active listening is a proactive process whereby we consciously set out with the intention of acquiring information about the beliefs and references (ICEBERGS – see diagram) that another party is using to understand or explain an issue of concern. It is especially relevant when different parties hold different point of views.

I	Ideas/hyposeses
C	Concerns: PPPP
E	Expectations
B	Beliefs
E	Evaluations
R	Rights
G	Goals
S	Strategies

Inter-personal skills

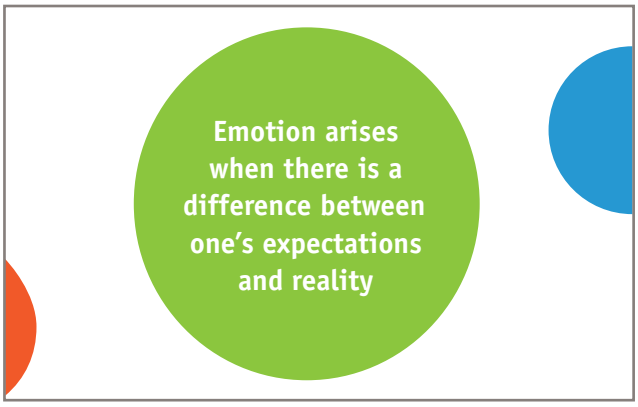
Inter-personal skills comprise both verbal and non-verbal communication as well as understanding crucial inter-personal emotions of trust, respect and assertiveness. Verbal communication is concerned with speaking effectively. The following basic habits are useful:

- Speak clearly: don't mumble, especially on the telephone
- Speak at an appropriate volume: neither too loudly or too softly
- Speak at the right speed: Most people assimilate new information at less than 120 words a minute
- Inflexion: vary the tone of your voice. A monotonous drone sounds disinterested. However an excessively wavy inflexion can be irritating
- Pause: don't feel rushed to respond to questions and conversations immediately, but it is better to pause for a moment in consideration, especially if the question merits it. No one expects, or wants, a gun-slinging attitude in important conversations. A thoughtful person is generally taken more seriously.

Emotional Intelligence

Client satisfaction is an emotion. If we wish to consistently achieve the emotion of client satisfaction, it is essential that we have a 'radar' that informs us if and when we are 'on' or 'off the mark' with each individual client. If we don't recognise when our views are missing the mark with clients, we are heading for a negative emotional outcome; dissatisfaction. Having this information helps us decide whether we need to adapt our approach. To do this we require Emotional Intelligence. The hallmark of any type of intelligence is the ability to adapt. Being tuned into emotion allows us to check that we are 'making sense' and connecting with other people and adapt accordingly.

What are emotions?



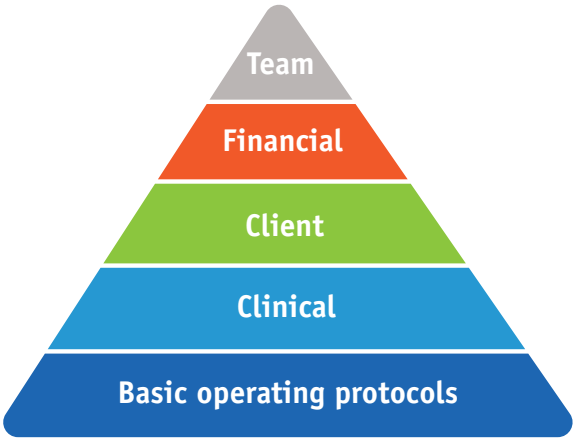
New clients

Often, clients with new pets 'phone around' practices checking prices on key comparison items such as vaccinations and neutering. It is important to be able to recognise these callers and adopt a strategy that will maximise the number of these 'prospect' calls that register with you and book an appointment at your surgery as opposed to simply stating the price and waiting for them to book an appointment. An in-depth discussion of this conversation is beyond the scope of this article but asking the age of the pet, its breed and name is a good way of striking up a conversation and expressing empathy and interest.

Telephone skills

The Basic Operating Skills module covered the importance of mastering the hardware aspects of the telephone system. It is equally as important to master various telephone conversation strategies in order to be both effective and efficient at booking appointments onto the practice diary. Basic skills include answering promptly, projecting a clear and friendly voice and offering or recommending an appointment for every clinical enquiry.

Level 4 – Financial Resolution



Financial resolution means billing and banking the money relating to the sale of services and products. A proactive approach to financial resolution means ensuring receptionists understand the sequence of events which will increase the chances that clients pay their bill.
The Colourful Receptionist© model looks at how receptionists can become more proactive in achieving financial resolution by understanding some of the skills, knowledge and attitudes relevant to this objective. This will be discussed using the acronym PROFIT.

P	Practice finances – what happens to every €100?
R	Requesting payment
O	Offers / promotions / value plans
F	Fair price: is it worth it?
I	Insurance
T	Teamwork; working as a team to get paid

Practice finances – what happens to every €100?

Although veterinary services are often perceived as expensive, many people might may be surprised to hear that veterinary practices are often not that profitable with less than €10 of every €100 taken resulting in profit to the practice owners. The main cost associated with veterinary practices is staff wages, typically accounting for over 40% of all monies received into the business. The next largest bill is the cost of sales, such as the wholesaler, laboratory and crematorium bill, which normally accounts for around 25% of all monies received. Finally, the cost of running the practice premises (heating, light, insurance) account for around 20% of income. That leaves around 10%.

Requesting payment

Sometimes it can feel uncomfortable asking a client for a large amount of money and having to deal with the client's response to their bill. It is useful to have a prepared speech that allows to approach the request for payment in a way that is customer-oriented, caring and yet assertive. A useful rule of thumb is to apply the 'pet, person, payment' formula. The 'pet' piece always comes first. In care industries such as veterinary practice, it is important that we project a greater care about our patients' well-being than money. This can be achieved by a simple enquiry as the client approaches the reception to pay; 'How's he doing?' and 'glad / sorry to hear that'.

The 'people' piece relates to the client. We often see this in action when checking out of hotels; 'was everything OK with your stay?' In veterinary practice we can ask 'Have you any questions before we sort out your next appointment and / or account?' The 'payment' piece always comes last in this sequence of customer and patient care.

Offers / promotions / value plans

It is not uncommon for veterinary practices to run various offers and promotions in an attempt to attract new clients or stimulate further sales. The receptionist team are usually asked and expected to administer, promote and process the offers and the promotional campaign. However, in veterinary practice it is not uncommon that the explanation and the training on details of the offers and the promotions to be less than ideal. Occasionally, veterinary receptionists are cynical and sceptical of offers and promotions for a variety of reasons:

- They perceive the mechanism as too complicated
- They don't believe the offer will work especially if previous attempts have not been successful
- They are concerned about the extra workload on top of an already busy schedule

Basic principles for implementing successful promotions

- Keep it simple.
- Provide the receptionist with an 'offers and promotions brief sheet', which should include some or all of the following:
  - What the promotion is
  - When it begins
  - When it ends
  - Which clients it applies to
  - Which products / services it applies to
  - The purpose of the promotion or why it is perceived necessary
  - The benefit to the pet / client if not obvious
  - A step-by-step mechanism of what to do
  - How clients will find out about the promotion
  - Who staff are expected to do to make clients aware of the promotion
  - Any tips on how to best to approach or not approach promoting the offer
  - Any other relevant details and who to direct queries
- Have all the price codes / pricelist items on the Practice Management System ready to use

Fair price: is it worth it?

Whilst some people find requesting any payment uncomfortable, most people find requesting payment for a service or product which they don't believe to be 'worth it' downright awkward. This sense of hesitancy and uncertainty about value and worth will undermine a client's perception of the value of a practice's services and products. There are several ways to combat this feeling. Understanding where every €100 actually goes is the first step in helping employees realise that most practices cannot be accused of blatant profiteering. Secondly, understanding the technical side using the PREVENT IT module of the Colourful Receptionist© enables staff to be much more informed and aware of the value of technical products and services. Finally, the acronym, ABCDE3 is a useful reminder of what constitutes value in the client's mind: "Always Believe your Charges are Determined by your Expertise, your Efforts and your client's Experience".

Insurance

Insurance claims represent a significant proportion of veterinary practice income. Veterinary receptionists are often asked by clients to explain clients how their insurance policy works and how to make a claim. It is also important to have a policy on how direct insurance claims are administered.

This will include considerations of the following:

- Minimum amount claimed
- Insurance companies that the practice will not deal with in terms of direct claims
- Period before which the amount due reverts back to the client
- Pre-authorisation protocol + / - credit card swipe
- Minimum deposit amounts over and beyond the excess and non-claimable items
- Charge for direct claim

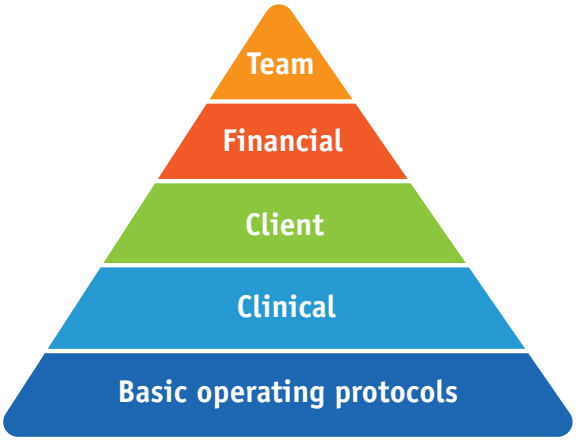
Finally, it is important to point out to clients the difference between Preventative Healthcare payment plans and insurance policies. It isn't uncommon for some clients to assume that they have 'insurance' when they sign up to the practice's PHC plan and have even cancelled their insurance policy and found themselves unable to re-insure their pet for pre-existing conditions.

Teamwork; working as a team to get paid

Whilst veterinary receptionists are the people in the practice who make the actual request for payment, they are not the only person who will influence whether the client actually pays their bill.

The article in EJCAP (2014), Winter 24(4); p46-p54 focused on how the author's Colourful Consultation© model can be used by veterinary surgeons to proactively facilitate the likelihood that clients will arrive at the reception desk, prepared and willing to settle their account.

Level 5 – Team Harmony'n'happiness



The final level takes a proactive approach to ensuring that receptionists are clear about the role they play in achieving and maintaining team harmony and happiness. Team harmony (as well as personal fulfilment) is often rooted in the self-confidence team members have about themselves. The reactive strategy to developing confidence and morale within a team believes (and hopes) that hiring the 'right people' and being 'nice to each other' is sufficient. The risk of this approach is that whilst you may have a group of talented individuals, they are not working as a team.

M	team Meetings
O	Order and organisation
R	Recognition and rewards
A	Appraisals
L	Leadership, vision, KPIs
E	Engagement

Team meetings

Team meetings are important ways of creating a shared understanding about what we consider to be the right standards, why they matter, as well as how to carry out various tasks in order to meet our targets and achieve our goals. Team meetings are much better at understanding

and setting the emotional tone of how we work compared to written communications. Whilst a full review of team meetings are beyond the scope of this article, it is important to hold them regularly, to ensure that they are chaired well and that at least basic minutes and action points are taken and distributed to all relevant parties afterwards, even if they were or were not in attendance.

Order and organisation

Stress equals uncertainty x urgency. Uncertainty caused by chaos and disorganisation are major causes of stress if we can't find what we are looking for, especially if we are already under time pressure (i.e. urgency). Whilst most practices have a filing system it only works as well as it is maintained. Understanding and actually using the practice filing system for both hard copy and digital documents makes retrieving important files predictable and efficient and saves individuals time and stress. This also applies to having a clear system for storing and accessing contact details. Labelling drawers and files using a standard font and format looks professional as well as being effective.

Recognition and rewards

Noticing and acknowledging good work and high standards is a significant component of building and maintaining team morale. Whilst the aim isn't to create a shallow or sycophantic culture, taking the time to recognise persistence, dedication, patience, attention to detail as well as care and kindnesses to colleagues makes a significant difference. A simple 'thanks for your help today' goes a long way. Rewards can be both formal and informal. Sometimes simply passing on a bottle or chocolates offered by a client to a colleague who has made a particular effort recently can be much appreciated. Of course rewards can also be formal in terms of remuneration but it tends to be the more day-to-day gestures of recognition and reward that make the most difference to team morale.

Appraisals

Appraising, evaluating and encouraging colleagues to explore and develop their potential is a key ingredient of morale. Whilst there are many means and models of appraising staff, the author uses The Colourful Appraisal© template helps identify specific areas of strength, weakness and opportunities for training in relation to the skills, knowledge and attitudes required to proactively pursue the four desired outcomes of the Colourful Culture©

The colourful appraisal

	Clinical resolution	Client satisfaction	Financial resolution	Harmony and happiness
Skills				
Knowlegde				
Attitude				

Leadership

Leadership is a valuable commodity in any organisation, and is a crucial component of organisational success. Many morale problems can be tracked back to less than ideal leadership knowledge, skill or attitudes. Leadership and culture are two sides of the same coin: what leaders believe and demonstrate is often reflected in the skills, knowledge and attitudes of the team. If practices are truly sincere in proactively pursuing the four outcomes referred to above, leaders must believe wholeheartedly in the importance of these outcomes and lead by example about how they wish them to be achieved within their team/surgery.

Engagement

Engagement means 'to be connected to'. When we refer to engagement about our work we are referring to the vigour, the dedication and the absorption we associate with our work. In other words, do we find it stimulating and interesting and something to which they really want to devote time and effort (the vigour component); do we see it as a meaningful thing to do (dedication); and does it stimulate our concentration and prevent us from becoming bored (absorption). The ultimate sensation of engagement is experienced as a sense of 'flow' which occurs when we are doing something that stretches and challenges our knowledge, skills and abilities such as resolving a conundrum or dealing with a complicated problem. This is why leadership and appraisals are so important to sustainable engagement and ultimately to team morale; we need to combine a vision about what it is we are trying to achieve (4 outcomes) and why as well as a clear concept of the skills, knowledge and attitudes we need to achieve them as well as a mechanism for tracking and developing them.

In conclusion, this and a previous article in EJCAP (2014), Winter 24(4); p46-p54 has focused on how the author's Colourful Consultation© and Colourful Receptionist© model are used to proactively pursue the four outcomes of sustainable clinical practice.