

GUIDELINE for Streptococcal infections

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Though different streptococci have been isolated occasionally from cats, including *S. agalactiae*, *S. pneumoniae*, *S. suis*, *S. pyogenes*, the most prevalent one is *S. canis* (Greene and Prescott, 2012). *Streptococcus equi* subsp. *zooepidemicus* became an emerging pathogen in dogs, and recently also in cats (Blum et al., 2010; Polak et al., 2014).

Streptococcus canis

This beta-haemolytic Lancefield group G gram-positive bacterium is considered part of the commensal mucosal flora of the oral cavity, upper respiratory tract, genital organs and perianal region in cats. The infection seems to be sporadic in single-cat households, especially in older cats (Greene and Prescott, 2012). Young queens (up to 2 years of age) may carry *S. canis* in the vagina, and the prevalence of infection is generally higher in cats housed in groups. Up to 70-100% of young queens in breeding catteries may carry this bacterium in the vagina, resulting in infection of the kittens, but also in the transfer of passive immunity against *S. canis* via colostrum. The level of maternally derived antibodies, immune response, age, infection pressure, stress and probably also the strain virulence determine whether the bacteria cause disease or not.

Contamination of the umbilical vein may lead to a generalized infection resulting in neonatal septicaemia (Greene and Prescott, 2012). In 3 to 7-month-old kittens, a subclinical infection of the pharynx and tonsils may induce cervical lymphadenitis. In older cats, the infection is usually opportunistic as a result of wounds, surgery, immunosuppression or viral infection (Fig. 1).

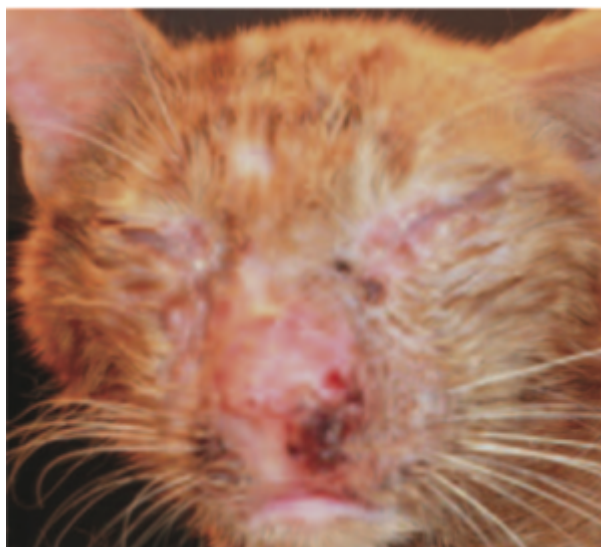


Fig. 1. In adult cats, S. canis infection is usually opportunistic as a result of wounds, surgery, immunosuppression or viral infection. In this shelter cats, viral infection was severely complicated by secondary bacterial infection due to poor hygienic conditions.

Courtesy of Tadeusz Frymus, Faculty of Veterinary Medicine, Warsaw University of Life Sciences, Poland

In up to 10% of cats suffering from chronic upper respiratory tract disease, *S. canis* can be isolated from the nasal cavity (Pesavento and Murphy, 2014; Fig. 2). Conditions associated with this pathogen include abscesses, pneumonia, discospondylitis, osteomyelitis, polyarthritis, urogenital infections, necrotizing fasciitis (toxic shock syndrome), sinusitis and meningitis. Outbreaks in cats with fatal diseases have been reported in shelters and breeding colonies (Pesavento and Murphy, 2014), as all these conditions may result in septicæmia and embolic lesions, especially of the lung and heart (Greene and Prescott, 2012).

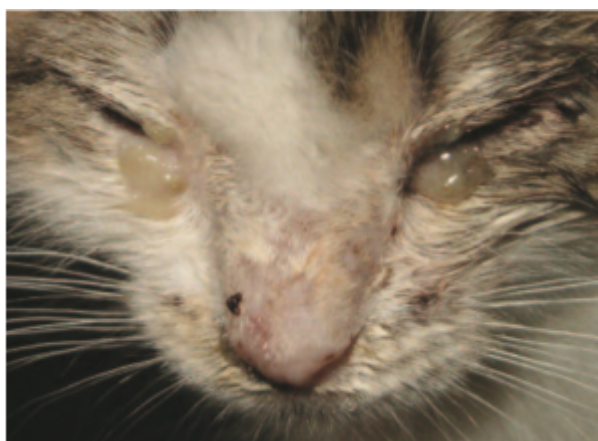


Fig. 2. In some cats suffering from upper respiratory tract disease, *S. canis* can be isolated from the nasal cavity. Courtesy of Tadeusz Frymus, Faculty of Veterinary Medicine, Warsaw University of Life Sciences, Poland

Microscopic examination of exudates or tissue reveals gram-positive cocci (usually in chains), and culture can confirm the diagnosis. *S. canis* is generally sensitive to penicillins, and early antibiotic application is the basis of therapy.

More information can be found in the review by Greene and Prescott (2012).

Streptococcus equi subsp. *zooepidemicus*

Agent and host susceptibility: *Streptococcus equi* subsp. *equi* (commonly referred to as *S. equi*) and *Streptococcus equi* subsp. *zooepidemicus* (*S. zooepidemicus*), beta-haemolytic gram-positive Lancefield group C bacteria, are the most important equine streptococci worldwide (Timoney et al., 1998). *S. equi* is an obligate agent causing strangles, the most frequently diagnosed infectious disease of horses, and one which is both devastating and highly contagious. *S. equi* is host-restricted infecting almost exclusively equines.

S. zooepidemicus is regarded a mucosal commensal, most notably in equines, with a potential to cause serious opportunistic disease secondary to viral infections, heat exposure, transportation or other stressful situations (Hoffman et al., 1993). Believed to be part of the normal microflora of the upper respiratory airways and lower reproductive tract, this bacterium is frequently isolated from suppurative discharge in horses including in cases of complications of viral infections of the upper airways (Hoffman et al., 1993; Timoney et al., 1998). However, in contrast to *S. equi*, *S. zooepidemicus* strains are highly diverse and are not restricted to causing disease in horses. These strains have been found in a wide range of other species including pigs, cattle, sheep, goats, poultry, dogs, cats, guinea pigs, seals, dolphins, monkeys, llama and farmed red deer (de Lisle et al., 1988; Sharp et al., 1995; Soedarmanto et al., 1996; Hewson and Cebra, 2001; Las Heras et al., 2002; Akineden et al., 2007; Timoney, 2008; Pisoni et al., 2009; Blum et al., 2010; Lamm et al., 2010; Priestnall and Erles, 2011; Venn-Watson et al., 2012). Occasionally glomerulonephritis, rheumatic fever, meningitis or purulent arthritis caused by *S. zooepidemicus* have been reported in humans (Kuusi et al., 2006; Abbott et al., 2010; Pelkonen et al., 2013). Many of these zoonotic conditions resulted from contacts with horses or from the consumption of unpasteurized milk of cows or goats.

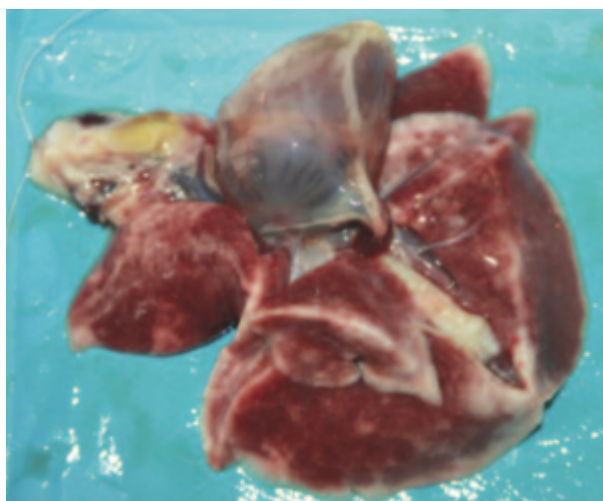
There is increasing evidence that the veterinary role of *S. zooepidemicus* may be underestimated, and concern has been expressed that this bacterium may be “potentially more than just an opportunist” (Björnsdóttir et al., 2012). Several outbreaks in species other than horses have been described. In Asia pandemics occurred in pigs (Feng and Hu, 1977; Soedarmanto et al., 1996). Also, in companion animals, the incidence of infections by this agent has apparently increased. Since 2003, several outbreaks of an acute *S. zooepidemicus* related severe haemorrhagic canine pneumonia have been described in many countries (Chalker et al., 2003; Kim et al., 2007; Gibson and Richardson, 2008; Pesavento et al., 2008; Byun et al., 2009). This disease is highly contagious and often fatal. The most prominent signs were a sudden onset, fever, dyspnoea, and haemorrhagic nasal discharge. Haemorrhagic pneumonia and pleural effusion were recognized *post mortem*. Most outbreaks occurred in shelters, where *S. zooepidemicus* infection caused many deaths. Also kennels and research facilities were involved (Garnett et al., 1982), but individually housed dogs were occasionally also affected (Gibson and Richardson, 2008; Abbott et al., 2010).



Fig. 3. Purulent nasal discharge and cough may be early signs of *S. equi* subsp. *zooepidemicus*-related diseases in cats. Courtesy of Tadeusz Frymus, Faculty of Veterinary Medicine, Warsaw University of Life Sciences, Poland

Feline *S. zooepidemicus*-related disease

It was thought that this bacterium played no role in diseases of cats until an outbreak was described 2010 in a shelter in Israel (Blum et al., 2010). Early clinical signs included an effusive purulent nasal discharge and cough (Fig. 3), progressing to sinusitis, dyspnoea, pneumonia and death. The vaccination status of this cattery was unknown. Between June 2006 and January 2008, 78 dead cats from a shelter housing approximately 700 animals were submitted for *post-mortem* examination. In 39 of these, the major necropsy findings were severe, acute and diffuse bronchopneumonia (Fig. 4) or bronchioalveolar pneumonia, either suppurative or necrosuppurative. Interstitial multifocal pyogranulomatous pneumonia was present in a few cats, pleuritis in 4 cases, and pyothorax in one animal. Pyogranulomatous meningoencephalitis was found in 4 cats. Necrosuppurative peritonitis was present in one case. The most common histopathological lesions were a diffuse mixed infiltrate of neutrophils, histiocytes and lymphocytes, thickening of the inter-alveolar septa and multifocal bacterial colonies with coccoid forms.



*Fig. 4. In cats succumbing to fatal *S. equi* subsp. *zooepidemicus* infection, the major finding is severe acute and diffuse bronchopneumonia. Courtesy of Karolina Kozłowska, Warsaw, Poland*

S. zooepidemicus was the main pathogen isolated from both the dead cats with signs of respiratory disease as well as from nasal and pharyngeal swabs or bronchoalveolar lavage obtained from sick animals. In the dead cats, *S. zooepidemicus* was isolated from the lungs in all cases, and also from the sinuses in a few. The bacterium was also cultured from the pleura in 2 of 4 cases of pleuritis, from the brain in 3 of 4 cases of meningoencephalitis and from the peritoneum in one case of peritonitis. Usually *S. zooepidemicus* was isolated alone or was dominant in mixed cultures. However, the bacterium was not isolated from any of the 29 dead cats without clinical and pathological signs of respiratory disease, and only from 2 of 10 animals in which respiratory disease was suspected prior to death, but no gross pathological signs were found on necropsy.

S. zooepidemicus could also be isolated from cats showing vague signs of respiratory disease, which possibly shed the organism long before being detected. This could suggest subclinical carriage. In the few cases with lesions suggesting feline infectious peritonitis, the presence of feline coronavirus was ruled out by immunohistochemistry. Tests for feline herpesvirus (FHV) and feline calicivirus (FCV) were not performed but, based on clinical signs, the authors suspected that the cat population in this shelter was infected with both viruses. They assessed the hygienic conditions and ventilation in this cattery as adequate and the facilities as not overcrowded. This could mean that *S. zooepidemicus* may have become persistent in a cattery in spite of sufficient hygienic practices and treatment. The authors speculated that the transfer to this shelter of a group of cats from another cattery (closed due to poor conditions) prior to the disease outbreak might have induced stress that facilitated this epidemic. However, the source of infection remained unknown. The cats had no contact with horses.

In 2010, a fatal *S. zooepidemicus* infection in two mature domestic cats housed in separate shelters was also described in Canada (Britton and Davies, 2010). Both animals had been resident for several months in the shelter prior to a sudden onset of a peracute disease with nonspecific clinical signs, and blindness in one of them, followed by death within 24 h. *Post-mortem* examination revealed rhinitis and meningitis and *S. zooepidemicus* was cultured from the nasal cavity and brain. Both cats had tested negative for feline leukaemia virus (FeLV) antigen and were seronegative for feline immunodeficiency virus (FIV) antibodies. Polymerase chain reaction (PCR) of lung, nasal mucosa and brain, performed *post mortem*, revealed that both cats were also negative for FCV and feline coronavirus (FCoV), and one was positive for FHV. Interestingly, other cats in these shelters remained normal. Neither of the succumbed cats, nor their shelter attendants, had had contact with horses.

The pathogenic role of *S. zooepidemicus* in cat colonies was revealed following a recent investigation of cat hoarding (Polak et al., 2014). In this study, about 2000 cats were removed from four sanctuaries following reports consistent with animal hoarding. During intake examination, 27% of the animals (366/1368) showed respiratory disease. A subset of 81 cats with respiratory signs was tested for infectious agents by PCR, and 55% were positive for *S. zooepidemicus*.

A case of acute *S. zooepidemicus* meningoencephalitis was also described in an exclusively indoor cat in the USA in 2011 (Martin-Vaquero et al., 2011). It was likely secondary to otitis media/interna, as recognized by computed tomography. The patient presented neurological signs of a central vestibular lesion and left Horner's syndrome. From the cerebrospinal fluid, with marked neutrophilic

pleocytosis, *S. zooepidemicus* was isolated in pure culture, but PCR results for *Toxoplasma gondii*, FCoV and FeLV were negative, as well as *Cryptococcus* sp. antigen enzyme immunoassay. A bulla osteotomy and debridement were performed, and according to resistance profile results, the cat was treated with trimethoprim-sulfamethoxazole for 8 weeks. The patient recovered fully.

In addition to the infections of domestic cats presented above, a fatal suppurative meningoventriculitis with intralesional *S. zooepidemicus* was also described in an elderly, captive snow leopard in Japan (Yamaguchi et al., 2012). This animal had had no contact with horses, but defrosted horse meat was fed routinely and was presumed to be the source of infection.

Epidemiology in small animals

It is generally considered that, in contrast to *S. canis*, *S. zooepidemicus* is not a part of the normal flora of dogs and cats (Smith, 1961; Bailie et al., 1978; Biberstein et al., 1980; Devriese et al., 1992). Nevertheless, both canine and feline subclinical infections have been observed (Chalker et al., 2003; Abbott et al., 2010; Acke et al., 2010; Blum et al., 2010). *S. zooepidemicus*-related diseases secondary to viral infections have been described in dogs, especially in cases with distemper, canine influenza virus (CIV) infections and other conditions (Yoon et al., 2005). The bacterium may also act as a primary cause of canine pneumonia, sometimes with a peracute course, although experimental infections have not been performed (Gower and Payne, 2012).

Horses are common carriers of this bacterium, and so contact with horses is a potential source of infection (Acke et al., 2010). Dogs experimentally infected with CIV and then kept together with healthy horses acquired *S. zooepidemicus* pulmonary infection (Yamanaka et al., 2012). Indirect transmission should also be taken into consideration, as equine streptococci may survive outdoors for up to several days, and indoors probably longer (Weese et al., 2009). It has been speculated that contact with staff members could explain outbreaks in canine research facilities and urban kennels, where direct contact with horses is excluded (Priestnall and Erles, 2011). Certainly *S. zooepidemicus* is able to spread between dogs through direct contact, and outbreaks in shelters usually affect large numbers of animals within a short time.

The same probably applies to cats. It has been postulated that close confinement of animals, such as in shelters, research laboratories and other facilities, appears to be the major risk factor for the development of *S. zooepidemicus*-associated disease in dogs and cats (Chalker et al., 2003; Britton and Davies, 2010). Co-infection with other respiratory pathogens, as well as age and health of the animal on entry to the facility has been shown to be unrelated to later colonization of the respiratory tract by *S. zooepidemicus* in dogs (Garnett et al., 1982; Chalker et al., 2003). Though it has been shown that strains with the same clonal complex are pathogenic for both dogs and cats (Britton et al., 2018), the role of dogs as a source for feline infections is not known; however, in one shelter, canine haemorrhagic pneumonia caused by this bacterium did not spread to cats located in an adjacent building of the same facility (Byun et al., 2009).

Pathogenesis in small animals

The pathogenesis of *S. zooepidemicus* infection in small animals is poorly understood. The existence in dogs of both subclinical and clinical infections of different severity suggests that some isolates might be more pathogenic. In many dogs, the rapid onset of disease and progression of the clinical signs are similar to human toxic shock syndrome caused by *Streptococcus pyogenes* (Priestnall et al., 2010). Toxic shock is characterized by a hyper-reactive inflammatory response resulting in increased vascular permeability, vasodilatation, increased coagulation and migration of inflammatory cells to the site of infection (Lappin and Ferguson, 2009). Pyrogenic exotoxins produced by some streptococci, including *S. pyogenes*, act as superantigens by binding simultaneously to major histocompatibility complex (MHC) class II receptors on macrophages and T-cell receptors, bypassing conventional antigen presentation, and leading to the activation of a large proportion of T lymphocytes (Fraser and Proft, 2008). The resulting overproduction of proinflammatory cytokines has been linked to increased virulence and has also been suggested to contribute to the pathogenesis of some streptococcal diseases. Marked elevation of the mRNA of some proinflammatory cytokines was also observed in dogs with *S. zooepidemicus*-induced pneumonia, and three superantigen genes were prevalent among canine isolates of the bacterium (Priestnall et al., 2010). So far, no clinical signs similar to the toxic shock syndrome have been described in cats. Various typing methods have been used to determine the virulence factors and genetic relationships among different *S. zooepidemicus* isolates; M-like protein, IgG-binding proteins and fibronectin binding protein appear to be the main virulent factors of this bacterium (Jonsson et al., 1995; Timoney et al., 1995; Hong, 2005).

To date, the factors underlying the differences in pathogenicity of some isolates/genotypes in cats and dogs remain unknown.

Diagnosis

A tentative diagnosis of a streptococcal infection can be made basing on the history, clinical signs, lesions and the presence of gram-positive coccus chains in the lesions. In cats with respiratory disease, *S. zooepidemicus* can be isolated from nasal and pharyngeal swabs, as well as from bronchoalveolar lavage, and from lung samples or other lesions in fatal cases (Blum et al., 2010). Selective

media for gram-positive organisms, such as Columbia agar with 5% sheep or horse blood containing colistin and nalidixic acid should be used. If Lancefield group C streptococci grow, the presence of *S. zooepidemicus* can be confirmed by biochemical methods such as the API20 Strep kit (BioMérieux). In contrast to *S. equi*, *S. zooepidemicus* has the ability to ferment ribose, sorbitol and lactose, properties that are commonly used to discriminate the subspecies (Bannister et al., 1985). Real-time PCR was found to be more sensitive than conventional culture (Baverud et al., 2007).

Treatment

There is only one report of an effective treatment of an acute *S. zooepidemicus* meningoencephalitis in a cat (Martin-Vaquero et al., 2011). Trimethoprim-sulfamethoxazole administered over several weeks was the main antibiotic. In suspected cases, treatment with broad-spectrum antibiotics should be initiated as soon as possible, and then adapted, if required, according to the results of culture and sensitivity tests. *S. zooepidemicus* isolates from dogs were found to be susceptible to penicillin, ampicillin, amoxicillin and enrofloxacin (Byun et al., 2009). Some isolates were found to be resistant to tetracycline and doxycycline (Garnett et al., 1982; Pesavento et al., 2008).

Prevention

There are little data about the management of *S. zooepidemicus* infections in catteries. However, sick cats should be isolated, and staff should wear protective clothing when caring for them. Hands, premises and all contaminated equipment should be thoroughly cleaned and disinfected. Quaternary ammonium disinfectants, phenol-based solutions or oxidising agents are generally recommended.

Though attempts have been made, there are no *S. zooepidemicus* vaccines available for any species.

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