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Review Article

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Recent Advances in the Management of Feline Sporotrichosis

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Abstract

Sporotrichosis is a subcutaneous mycosis caused by species of the genus *Sporothrix* that has stood out in recent years, mainly in Brazil. Its importance for public health as an emerging disease is mainly associated with zoonotic transmission by domestic cats, which have a high fungal load on skin lesions, facilitating the dissemination of the agent. Classical transmission is caused by traumatic inoculation of the fungus in a contaminated environment. Propagation between animals occurs due to close contact between cats with other cats or dogs and for humans associated with bites, scratches, and interaction with exudate from infected lesions. The clinical manifestations in felines are variable and can present as localized or fixed, disseminated, cutaneous-lymphatic, and systemic. In human patients, mainly immunosuppressed, extracutaneous forms can occur, evolving even to fatal cases. Early diagnosis is essential to ensure that preventive measures are employed for owners, with the isolation of the etiological agent in culture being considered the gold standard. The lack of strategic plans, adequate control measures, failure to recognize and neglect the disease increases the incidence of cases, especially in more precarious population groups. Thus, due to its importance in public and animal health, the objective of this review was to highlight recent advances concerning the feline disease.

KEYWORDS
Cat, Fungus, Public health, *Sporothrix* spp, Zoonosis.

Introduction

Sporotrichosis is a subacute to chronic mycotic infection of zoonotic character, caused by thermally dimorphic fungi of the genus *Sporothrix* (Queiroz-Telles *et al*., 2019). It is the most prevalent subcutaneous mycosis globally, although it is generally neglected (Queiroz-Telles *et al*., 2017; Rodrigues *et al*., 2020). Currently, it is one of the most important zoonoses due to its impact on public health, with felines being the main sources of infection, transmission and maintenance of the disease (Eyer-Silva *et al*., 2019; Gonsales *et al*., 2019; Etchecopaz *et al*., 2020), highlighting the importance of this species in the epidemiology of the disease (Silva *et al*., 2018).

In Brazil, 91% of the cases described in humans from 2005 to 2008 occurred after contact with cats (Freitas *et al*., 2010). This fact is justified by the proximity of the people to these domestic animals. Cats can present a large number of infectious fungal cells in skin lesions, which enhances the transmission capacity (Rodrigues *et al*., 2020). The lack of appropriate control measures, associated with the expansion of the disease in urban areas, allows severe sporotrichosis cases to occur in more susceptible populations (Silva *et al*., 2012).

Classical transmission is triggered by the sapronotic route, involving the traumatic inoculation of the fungus in a contaminated environment (Orofino-Costa *et al*., 2017; Miranda *et al*., 2018a). Animal transmission can also occur through cat-cat or cat-dog contact and in zoonotic, animal-human form, in addition to possible conidia inhalation (Teixeira *et al*., 2015; Rios *et al*., 2018). Clinical manifestations in felines are wide (Backel and Cain, 2017), with the majority of cases developing skin lesions with nodules that can ulcerate and suppurate (Thomson *et al*., 2019).

Early diagnosis is essential to ensure that adequate conduct is implemented (Gremião *et al*., 2020) as the beginning of immediate treatment to reduce the chance of transmission. The therapeutic approach is challenging due to the zoonotic risk, a prolonged period to obtain clinical cure, and increased refractory cases (Han *et al*., 2017).

In recent years, there has been an increase in the incidence of zoonotic sporotrichosis, which is of concern in several countries, especially in Brazil (Chakrabarti *et al*., 2015; Gremião *et al*., 2017). The absence of appropriate surveillance and strategic plans leads to the unnoticed appearance of fungal infections (Gremião *et al*., 2020). Considering the importance of public and animal health, the objective of this review was to highlight recent advances concerning the disease in cats.

Etiopathogenesis

Sporotrichosis is a fungal infection caused by species of the genus *Sporothrix* belonging to the Ascomycota division, class

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Pyrenomycetes, order Ophiostomatales, and family Ophiostomataceae (Tovikkai *et al*., 2020). These are thermodimorphic fungi that have a mycophilic saprophyte phase at room temperature of 25 to 28°C and a pathogenic phase like yeast between 36 to 37°C, with a morphology similar to a cigar (Carnero *et al*., 2018; Lopes-Bezerra *et al*., 2018). They can be found in vegetation, animal waste, and soils rich in cellulose, with relative humidity above 92% (Téllez *et al*., 2014).

The *Sporothrix schenckii* complex comprises at least six species (Backel and Cain, 2017). The main ones that affect humans and animals are *Sporothrix schenckii* sensu stricto, *S. brasiliensis*, *S. globosa*, and *S. luriei* that compose the clinical clade (Zhou *et al*., 2014; New *et al*., 2019), being *S. brasiliensis* the most important in feline sporotrichosis, considered the most virulent of the complex and associated with outbreaks in Brazil (McCarty and Pappas, 2014; Rodrigues *et al*., 2016a). In contrast, the species of the *Sporothrix pallida* complex (*S. mexicana*, *S. chilensis*, *S. palmicuminata*, *S. humicola*, *S. pallida*, and *S. stylites*) integrate the environmental clade and are slightly pathogenic (Rodrigues *et al*., 2018; Makri *et al*., 2020).

Studies have shown clinical and environmental isolation of *S. globosa* in Asia, Europe, and the Americas, *S. schenckii* in North and South America, and *S. mexicana* in environmental isolations in Mexico (McCarty and Pappas, 2014). Although it has a worldwide occurrence, the etiology of the agents is not uniformly distributed in the different locations (Gutierrez-Galhardo *et al*., 2015).

The fungus is inoculated from contact wounds or traumatic injuries (Lloret *et al*., 2013), thus establishing a pre-patent period that varies from three to 84 days, mean of 21 days (Larsson, 2011). The infection can remain at the point of inoculation or extend systemically through lymphatic drainage or blood vessels, especially in immunosuppressed patients (Schubach *et al*., 2012; Lloret *et al*., 2013).

Several virulence factors are produced during the pathogen-host interaction. However, they are not well understood (Almeida-Paes *et al*., 2015; Della-Terra *et al*., 2017). Virulence profiles change according to the host's pathogen and immunity (Orofino-Costa *et al*., 2017). One of the factors is the production of melanin (Romero-Martinez *et al*., 2000; Orofino-Costa *et al*., 2017). This prevents phagocytosis by macrophages, enabling the onset of infection and resistance to antifungals (Almeida-Paes *et al*., 2016; Mario *et al*., 2016).

Moreover, the fungus can recognize fibronectin, laminin, and type II collagen, which are three important extracellular matrix glycoproteins that allow adhesion and interaction with the tissue (Lima *et al*., 2004; Teixeira *et al*., 2009). Thermotolerance is essential for infection in the host (Almeida-Paes *et al*., 2015). Only skin lesions are observed in isolates capable of growing at 35°C, but not at 37°C, with no lymphatic or visceral involvement (Barros *et al*., 2011).

Epidemiology

Sporotrichosis is a zoonotic disease with a cosmopolitan distribution, predominantly in subtropical to tropical regions (Rodrigues *et al*., 2016a; Han and Kano, 2021). Considered endemic in Brazil, Mexico, Uruguay, Peru, Japan, India, China, and South Africa (Backel and Cain, 2017). It is considered the most common subcutaneous mycosis in Latin America (Mora-Montes *et al*., 2015).

In Brazil, it is considered an emerging zoonosis (Gremião *et al*., 2017), and most cases occur in the south and southeast of the country, with the state of Rio de Janeiro classified as an epidemic area for sporotrichosis (Oliveira *et al*., 2011; Gremião *et al*., 2015; Sanchotene *et al*., 2015; Poester *et al*., 2018). In this state, the disease was reported in 1503 cats and 64 dogs by the Clinical Research Institute Evandro Chagas (Fiocruz, Rio de Janeiro) from 1998 to 2004 (Schubach *et al*., 2005). When considering the records up to 2015, 4.703 cases of sporotrichosis in cats were described (Gremião *et al*., 2017) and, until the year 2017, 4.916 cases (Rodrigues *et al*., 2020). However, the disease's real prevalence is difficult to estimate due to the negligence of the cases (Rodrigues *et al*., 2018).

Transmission occurs mainly through inoculation by cutaneous traumas and contaminated debris from plant material in the environment (Lopes-Bezerra *et al*., 2018). On the other hand, animal transmission is triggered by close contact between cat-cat or cat-dog, and in zoonotic form, animal-human, being associated with bites, scratches, and interaction with exudate from infected lesions (Miranda *et al*., 2013; Montenegro *et al*., 2014; Rodrigues *et al*., 2016a).

Areas with high feline population density tend to be important in animal transmission. The disease occurs mainly in urban areas without basic sanitation and in places with precarious conditions and difficulty accessing health services (Sanchotene *et al*., 2015; Gremião *et al*., 2020). The disease predominates in young adult cat males, unneutered, not domiciled, or with access to the street, without racial predisposition (Bazzi *et al*., 2016; Backel and Cain, 2017; Han and Kano, 2021).

The spread of the disease in this species is related to animal behavior due to the territorial dispute and the intimate contact that cause the traumatic inoculation of the fungus (Montenegro *et al*., 2014). When digging the soil or in contact with tree bark, the claws allow the acquisition of propagules (Rodrigues *et al*., 2014). It is important to note that they have a peculiar habit of cleaning licking, which can contaminate the oral mucosa and other parts of the body (Rodrigues *et al*., 2018).

This species has a high transmission capacity due to a large number of yeasts in skin lesions, claws, nasal and oral cavities (Madrid *et al*., 2012; Duangkaew *et al*., 2019). Schubach *et al*. (2002) found in their study the isolation of *S. schenckii* in 100% of the skin lesions, 66.2% of the nasal cavities, 41.8% of the oral cavities, and 39.5% of the nails of cats affected by sporotrichosis. In addition, the fungus does not die with the host, and it infects the soil, continuing transmission (Rodrigues *et al*., 2013). The feces of contaminated animals are identified as possible sources of the microorganism in nature (Montenegro *et al*., 2014; Rodrigues *et al*., 2015), and infection can also occur through inhaling spores (Backel and Cain, 2017).

This mycosis has been described in different animal species such as dogs, armadillos, horses (Téllez *et al*., 2014), rats, cattle, goats, pigs, mules, dolphins, camels, donkeys, chimpanzees, and birds (Madrid *et al*., 2012; Rodrigues *et al*., 2013), as well as invertebrates, like ants and spiders (Moaven *et al*., 1999; Miller and Keeling, 2002).

Clinical signs

Clinical manifestations range from self-limiting, with single lesions, to fatal systemic involvement (Macêdo-Sales *et al*., 2018). The determining factors for the appearance of clinical signs are the host's immune response, the size of the inoculum, pathogen virulence, and the initial trauma's depth (Barros *et al*., 2011; Rodrigues *et al*., 2016b). In dogs and cats, the presentations of the disease are classified into distinct forms: localized or fixed, disseminated, cutaneous-lymphatic and systemic (Mascarenhas *et al*., 2018).

The cutaneous form is characterized by ulcers and multiple nodules with crusts, alopecia, abscesses, or cellulitis in the extremities of the limbs, tail, head, and mainly in the region of the nasal plane (Lloret *et al*., 2013; Rodrigues *et al*., 2018; Han and Kano, 2021). In addition, lesions in the dorsal, ventral, and testicular regions can also be observed (Almeida *et al*., 2018).

Generalized injuries result from self-inoculation due to feline hygiene habits, an initial skin lesion, or hematogenous spread after primary respiratory infection (Lloret *et al*., 2013; Rodrigues *et al*., 2018). In the cutaneous-lymphatic form, there is an involvement of the skin, subcutaneous and lymphatic tissue. In this form, the animals present nodular or ulcerated lesions at the inoculation site of the agent, which follows a lymphatic trajectory, developing lymphangitis and regional lymphadenitis (Schubach *et al*., 2004; Crothers *et al*., 2009; Téllez *et al*., 2014).

On the other hand, extracutaneous can occur as unusual cutaneous-lymphatic sequelae, with the liver and lung being the preferred sites of dissemination (Crothers *et al*., 2009; Mascarenhas *et al*., 2018). The spleen, kidneys, lymph nodes, and testicles may also be involved (Schubach *et al*., 2003; Madrid *et al*., 2012). Systemic clinical signs are nonspecific and include lethargy, anorexia, and fever (Lloret *et al*., 2013). Injuries to the upper respiratory tract and nasal cavity are usually seen in the most severe cases (Duangkaew *et al*., 2019).

It is assumed that coinfection with feline immunodeficiency virus (FIV) and/or feline leukemia virus (FeLV) has no significant association with manifestations in cats with sporotrichosis (Schubach *et al*., 2004; Pereira *et al*., 2010). However, Miranda *et al*. (2018b) found that the poor general condition of the animals and the presence of feline retroviruses, in general, can influence the clinical manifestation of animals with sporotrichosis. Despite this, the disease can occur independently of retroviruses (Lloret *et al*., 2013; Gremião *et al*., 2017).

In humans, cutaneous-lymphatic sporotrichosis is the most common, followed by the cutaneous form, with possible systemic manifestation associated with the individual's immune status (Rodrigues *et al*., 2013; Oliveira *et al*., 2014; Paixão *et al*., 2015; Rodrigues *et al*., 2018). Chronic use of illicit drugs, immunosuppressive medications, secondary to acquired immunodeficiency syndrome (AIDS), alcoholism, and diabetes increase the risk of spreading the disease with the involvement of internal organs and neurotropism exhibited by *S. brasiliensis* (Orofino-Costa *et al*., 2017; Queiroz-Telles *et al*., 2019). In addition, there is a description of the infection in transplant patients (Da Silva *et al*., 2017; Amirali *et al*., 2020).

Diagnosis

The early diagnosis of feline sporotrichosis is essential to ensure immediate treatment and adequate prevention to avoid zoonotic transmission, especially for immunosuppressed people (Pereira *et al*., 2011; Gremião *et al*., 2020). The initial tools are based on the animal's history, anamnesis, and clinical examination. For confirmation, it is necessary to use complementary exams, such as cytopathology, histopathology, mycological culture, serological tests, and molecular diagnosis (Lloret *et al*., 2013; Rodrigues *et al*., 2015; 2018).

Cytopathology

Cytopathology is performed by collecting specimens obtained from lesions' smears with secretions, ulcerated, or fine-needle aspirates from nodules or abscesses (Lloret *et al*., 2013; Rodrigues *et al*., 2018). Analysis of this material reveals several round, oval, or cigar-shaped yeast-like forms present within neutrophils and macrophage, or in the extracellular medium (Jessica *et al*., 2015). Compared to other diagnostic methods, cytopathology is a simple, non-invasive, highly sensitive, and less expensive technique, which is an alternative to monitor the fungal load of lesions during treatment in animals (Miranda *et al*., 2018a).

Histopathology

Histopathology is a valuable method in lesions with intact nodules. The material is obtained through incisional or excisional biopsy and necropsy (Larsson, 2011; Bazzi *et al*., 2016). In some cases, a mixed, granulomatous, and pyogenic inflammatory process that encompasses the dermis, subcutaneous tissue, and deeper tissues can be observed (Lloret *et al*., 2013; Rodrigues *et al*., 2018). The analysis is performed by staining with hematoxylin and eosin (HE) (Silva *et al*., 2018). However, for the evaluation of yeasts, especially when there is less amount in the lesion, special stains such as periodic acid Schiff (PAS) and Grocott Methenamine Silver (GMS) allow better visualization of the agent (Silva *et al*., 2013; Bazzi *et al*., 2016). Immunohistochemistry is another alternative for histological recognition; however, its sensitivity is still being evaluated in felines (Gremião *et al*., 2015).

Mycological culture

Mycological culture is considered the gold standard for the diagnosis of sporotrichosis. This can be performed from exudates, although should be performed mainly from skin biopsy sample (Lloret *et al*., 2013; Backel and Cain, 2017). However, it can take about 30 days for a conclusive result, thus delaying the start of antifungal therapy (Jessica *et al*., 2015; Sanchotene *et al*., 2015; Gonsales *et al*., 2019),

In the generalized form of sporotrichosis, materials such as blood, urine, secretions, in addition to the cerebrospinal fluid, can be used in making the diagnosis (Rodrigues *et al*., 2018). The cultivation of these samples can be performed in media such as Sabouraud Agar with chloramphenicol or cycloheximide (Oliveira *et al*., 2014; Rodrigues *et al*., 2018), with filamentous colonies varying in white coloration the cream, which over the days become blackened (Madrid *et al*., 2012; Oliveira *et al*., 2014). A microculture is carried out on a blade from this colony, making it possible to observe micromorphological aspects, with conidia in shape similar to "daisies" (Larsson, 2011).

Others tests

Serological tests have been studied to diagnose and monitor patients (Rodrigues *et al*., 2018). A study evaluated the ELISA test to identify *S. schenckii* antibodies in felines, demonstrating sensitivity and specificity above 90%, which can be a valuable screening method (Fernandes *et al*., 2011).

Other techniques such as PCR have been successful in their use, making it possible to identify *S. schenckii* in biopsy samples (Kano *et al*., 2005). In addition, a recent study found that real-time multiplex PCR showed high sensitivity and speed, being a promising tool for the diagnosis of feline sporotrichosis (Zhang *et al*., 2019).

Treatment

The treatment of feline sporotrichosis is challenging due to the limitation of antifungals with high costs and adverse effects (Gremião *et al*., 2015). Itraconazole, ketoconazole, potassium

iodide, terbinafine, amphotericin B, local thermotherapy, cryosurgery, and surgical resection are currently available treatment options (Han and Kano, 2021).

The frequently used therapy consists of azoles, with itraconazole being safer than ketoconazole, which is commonly associated with adverse reactions, such as liver dysfunction, vomiting, and changes in cortisol metabolism (Han and Kano, 2021).

The drug of choice is itraconazole at a dose of 8.3 - 27.7 mg/kg every 24 hours (Gremião *et al*., 2015). Continuation of treatment must be maintained for at least one month to avoid recurrences (Duangkaew *et al*., 2019; Makri *et al*., 2020; Gremião *et al*., 2021). Nevertheless, as it is a hepatotoxic medicine, there is a significant risk of causing liver failure in the long term. Thus, it is essential to monitor the patient's serum biochemistry during treatment (Thomson *et al*., 2019).

Animals with respiratory disorders, cutaneous, and/or nasal mucosa alterations are predisposed to therapeutic failure (De Souza *et al*., 2018). In these cases, it is necessary to maintain continuous treatment for at least 60 days after clinical cure (Gremião *et al*., 2020). Potassium iodide may be an option in cases refractory to itraconazole (De Souza *et al*., 2016), although it may have side effects in cats such as iodism (Makri *et al*., 2020). The mechanism of action of this medication is still not very well elucidated (Reis *et al*., 2016|). However, its action has been described by inhibiting the formation of mature biofilms from *Sporothrix* spp. in filamentous and yeast form (Brilhante *et al*., 2019).

A study of 48 cats that received potassium iodide orally as treatment at doses ranging from 2.5 to 20 mg/kg every 24 hours demonstrated a clinical cure in 47.9% of cases (Reis *et al*., 2012). In addition, the combination of this drug with itraconazole is a good alternative for cases refractory to this medication, especially when there are respiratory signs (Rocha *et al*., 2018).

The effectiveness of using fluconazole in the treatment of feline sporotrichosis remains unclear (Gremião *et al*., 2020). Despite this, Crothers *et al*. (2009) reported the clinical cure of a cat with cutaneous lesions and respiratory signs with the administration of the antifungal at a dose of 10 mg/kg orally, once a day, for 80 weeks. However, some studies have demonstrated low in vitro efficiency of medication against *S. brasiliensis* (Stopiglia *et al*., 2014; Brilhante *et al*., 2016).

Posaconazole showed high in vitro efficacy against *S. brasiliensis* (Borba-Santos *et al*., 2015), and an in vivo study showed efficacy in a murine model of disseminated sporotrichosis (Fernández-Silva *et al*., 2012). There is a report with cytoreduction, itraconazole, and subsequently, posaconazole in a feline with *S. pallida* infection, with success in the clinical resolution (Thomson *et al*., 2019). High costs can hinder treatment, and further studies are needed to confirm the efficacy of the drug in feline sporotrichosis (Borba-Santos *et al*., 2015).

Terbinafine is also a fungicide option that inhibits ergosterol synthesis in the cell wall (Han and Kano, 2021). The efficacy in animals is unknown, but it was satisfactory in treating two dogs with sporotrichosis caused by *S. brasiliensis* (Viana *et al*., 2018) and in humans (Francesconi *et al*., 2011).

There are no reports of clinical cure by administering amphotericin B intravenously in cats, and its use is restricted due to adverse reactions (Gremião *et al*., 2015). Intralesional application is a possibility in treating residual sporotrichosis lesions refractory to itraconazole; however, sedation of the animal is necessary (De Souza *et al*., 2016). A study using intralesional amphotericin B and oral itraconazole in 26 cats with sporotrichosis demonstrated clinical cure in 72.7% of cases, and in 27.3%, the lesions recurred in the same location (Gremião *et al*., 2011).

Local hyperthermia can be an alternative in the fixed cutaneous form of the disease since the growth of the fungus is inhibited due to high temperatures (Honse *et al*., 2010). The application of cryosurgery associated with the use of itraconazole proved to be efficient in achieving clinical cure in 11 out of a total of 13 cats with sporotrichosis described in one study (De Souza *et al*., 2016). In addition, there are reports of surgical resection as a possible therapeutic approach (Gremião *et al*., 2006; Hirano *et al*., 2006).

Due to the zoonotic potential of the disease, treatment by the owners is difficult, preventing the control of the disease (Nakasu *et al*., 2021).

Prognosis

The prognosis depends on the animal's general conditions, presence of clinical signs involving the respiratory system, extent, and location of the lesions (Gremião *et al*., 2015). It is considered reserved to poor due to the zoonotic potential of the etiologic agent, high costs with long-term treatment, possible side effects of antifungals, and resistant strains (Han and Kano, 2021). It becomes unfavorable when the owner cannot perform the appropriate treatment associated with the patient's systemic and respiratory impairment (Almeida *et al*., 2018).

Prevention

Educational programs for health professionals and the community to improve feline management, informing about essential aspects of the disease's spread and promoting the reproductive control of these animals through castration, are essential to avoid transmission between species and for humans (Barros *et al*., 2011; Rodrigues *et al*., 2016a; 2018). Moreover, the availability of free veterinary services with appropriate treatments and planning for controlling stray animals are important strategies (Lecca *et al*., 2020).

The awareness of tutors in restricting the animal's access to a closed environment, without access to the street, especially in endemic areas, and the need to cremate the cats that die, preventing the spread and permanence of the fungus in the environment, are precautions recommended to reduce the incidence of the disease (Lloret *et al*., 2013; Rodrigues *et al*., 2018).

Veterinarians are included in the risk group for sporotrichosis, so it is extremely important to manipulate infected patients or with lesions suggestive of the disease using protective equipment, sanitary clothes, gloves, maintaining good hygiene, and disinfecting the environment of work (Lloret *et al*., 2013).

In Brazil, one method proposed for control is the vaccination of felines; however, the knowledge of antifungal immunity and the interactions between pathogens and host is still scarce (Schwartz *et al*., 2016; Téllez-Martínez *et al*., 2019). A cell wall protein-based vaccine candidate conferred protective immunity on a mouse model, encouraging this strategy; however, more studies are needed for new discoveries (Portuondo *et al*., 2016).

Conclusion

Sporotrichosis is a zoonosis of extreme importance and requires special attention from health services. Despite the increase in the number of cases, it is still a neglected disease, and it is necessary to emphasize the role of the feline as a potential transmitter of infection to humans. An early diagnosis for immediate initiation of treatment, to reduce the chance of transmission, effective prevention/control actions, plans, and educational measures to raise awareness of the population are essential to prevent the spread of the disease and, consequently, the appearance of new outbreaks.

Conflict of interest

The authors declare that they have no conflict of interest.

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