Treatment of refractory feline sporotrichosis with a combination of intralesional amphotericin B and oral itraconazole

IDF Gremião,†‡ TMP Schubach,§ SA Pereira,¶ AM Rodrigues,‖ CO Honse* and MBL Barros‡

Objective To describe the use of intralesional amphotericin B in localised lesions for the treatment of 26 cats from Rio de Janeiro, Brazil, with sporotrichosis refractory to oral itraconazole.

Design Uncontrolled intervention study.

Method The 26 cats in this study were diagnosed with sporotrichosis, confirmed by isolation of Sporothrix schenckii, and presented residual localised skin lesions refractory to treatment with oral itraconazole for a minimum period of 8 weeks. The animals received weekly applications of intralesional amphotericin B in conjunction with oral itraconazole. In cases of owner unavailability, a maximum of 2 weeks between the infiltrations was accepted.

Results Twenty-two (84.6%) of the 26 treated cats achieved clinical remission, 16 (72.7%) of which were cured, and in the remaining six (27.3%) the lesions recurred at the same site. Lack of clinical response was observed in one animal and three owners abandoned treatment.

Conclusion The proposed therapeutic regimen is an adjunctive treatment option for cats with sporotrichosis presenting as residual skin lesions refractory to itraconazole.

Keywords amphotericin B; cats; intralesional therapy; itraconazole; sporotrichosis

Abbreviation IL, intralesional

Sporotrichosis, a mycosis caused by the dimorphic fungus Sporothrix schenckii, affects diverse species of mammals, including humans, with the common house cat the most frequently implicated animal.

Sporotrichosis is frequent in Brazil, the largest number of cases having been reported in Rio de Janeiro, where the first epidemic of sporotrichosis in humans was a result of zoonotic transmission because of the close contact between people and sporotrichosis afflicted cats with skin and mucosal lesions. Between 1998 and 2004, 759 humans, 64 dogs and 1503 cats were diagnosed with sporotrichosis at the Evandro Chagas Clinical Research Institute-Fiocruz. Among these, 85% of the dogs and 83.4% of the patients had contact with cats with sporotrichosis, with 55.8% of the patients reporting cat bites or scratches. A total of 804 people were diagnosed with sporotrichosis between 2005 and 2008, corresponding to an annual increase of 85%.

Treatment of feline sporotrichosis is difficult and presents a challenge to veterinarians since the therapeutic options are limited and the drugs may have adverse effects. Iodides, azolic antifungal agents (ketoconazole and itraconazole), amphotericin B, terbinafine, local heat therapy and surgical removal of the lesions are the current treatment options available for cats with sporotrichosis.

Itraconazole is effective and safe, compared with other oral antifungal agents, and is therefore the drug of choice for the treatment of the disease, especially in situations where cost is not an issue. However, the clinical response is unsatisfactory in some cases, although treatment has proven to be effective in cats and convenient for the owners. In cases of feline sporotrichosis refractory to itraconazole, combined subcutaneous or intralesional (IL) administration of amphotericin B might be an alternative. Amphotericin B has been indicated for the treatment of rapidly progressive or severe systemic mycosis, imidazole-resistant cryptococcosis and disseminated forms of sporotrichosis, despite its use being limited by nephrotoxicity. Reports on the application of amphotericin B for the treatment of feline sporotrichosis are scarce. Intralesional administration of amphotericin B for the treatment of cats with sporotrichosis has been described in three cases, but the results were unsatisfactory. We have reported IL treatment in one case of feline sporotrichosis. In this study, we describe the IL administration of the drug in combination with oral itraconazole in 26 cats with refractory sporotrichosis, which presented as persistent skin lesions.

Materials and methods

An uncontrolled intervention study was conducted. The study consisted of cats with residual localised lesions of sporotrichosis refractory to oral itraconazole from the metropolitan region of Rio de Janeiro, Brazil, and later followed up at the Laboratory of Clinical Research on Dermatozoonosis in Domestic Animals (LAPCLIN-DERMZOO), Evandro Chagas Clinical Research Institute (IPEC)/Oswaldo Cruz Foundation (Fiocruz), between 2007 and 2009.

The indications for IL treatment were based on a diagnosis of sporotrichosis confirmed by isolation of Sporothrix schenckii in culture and the presence of skin lesions refractory to treatment with oral itraconazole. The criteria for refractory sporotrichosis were the lack of clinical response for at least 8 weeks under oral itraconazole administration. Susceptibility tests for antifungal drugs were not used. The exclusion criteria were: age under 6 months and over 12 years, mucosal or multiple (more than five) cutaneous lesions and the presence of nasal secretion and/or sneezing.
For IL administration of amphotericin B, 5 mL 2% lidocaine hydrochloride and 5 mL distilled water were added to a flask containing 50 mg of the drug to obtain a final concentration of 5 mg/mL amphotericin B and 1% lidocaine. The animals were sedated with 10% ketamine hydrochloride (10–15 mg/kg) and 1% acepromazine (0.1 mg/kg) intramuscularly. Amphotericin B was directly infiltrated into the lesions with a 0.38 \times 13 \text{ mm} (27.5 G1/2) needle attached to a 1-mL disposable syringe until lesion swelling was achieved. The needle was moved in different directions to guarantee infiltration of the whole lesion (Figure 1). Intralesional amphotericin B was applied either once a week or every other week until complete healing of the lesion. Blood samples were collected for routine serum chemistry and haematological examination before and during the study. Oral antifungal therapy (100 mg itraconazole/day) was maintained throughout the study and for an additional month after lesion healing. Clinical cure was evaluated one month after the end of IL amphotericin B treatment. The cats were followed up clinically for 6 months after discharge. All procedures of this study were approved by the Ethics Committee on the Use of Animals of the Oswaldo Cruz Foundation (CEUA-Fiocruz).

Results

Of the 26 cats in the study, 17 (65.4%) were males. Age ranged from 10 months to 11 years (median 3.5 years) and weight from 2.5 to 5.5 kg (median 4.1 kg). The period of itraconazole therapy before starting IL amphotericin B ranged from 12 to 120 weeks (median 32 weeks). The duration of clinical refractoriness to oral itraconazole therapy prior to intervention with IL amphotericin B varied from 8 to 55 weeks (median 13 weeks).

All animals were in good general health. Most lesions were located on the head, especially the nasal bridge (n = 10, 38.5%) (Figure 2) and nasal plane (n = 4, 15.4%). Simultaneous involvement of the nasal bridge and plane was observed in three cases (11.5%). Other sites affected were the right pinna in two cats and one each of left pinna, both right and left pinnas, left forelimb, right forelimb, left dorsolumbar region, face, and left hindlimb. Cutaneous nodules were observed in 17 (65.4%) animals and ulcers in 9 (34.6%).

Twenty (76.9%) cats had one lesion, three (11.5%) had two lesions, two (7.7%) had three lesions and only one animal (3.8%) had four lesions. A total of 36 lesions were detected in the 26 cats and all lesions were locally infiltrated.

Twenty-two (84.6%) of the 26 treated cats achieved clinical remission, 16 (72.7%) of which were cured (Figures 3, 4) and in 6 (27.3%) presented recurring lesions at the same site, the recurrence appearing 2 (n = 2), 3 (n = 2) and 5 (n = 2) months after clinical cure. Three (11.5%) animals’ owners abandoned treatment and there was a lack of clinical response in one (3.8%). No deaths occurred during the study period. Clinical inactivity and recurrence were observed in animals with nasal lesions, except for one cat in which the lesion was located on the forelimb.

The number of IL applications of amphotericin B ranged from 1 to 5 (median 2 applications), with a drug volume of 0.5 to 1.5 mL (2.5–7.5 mg) per application (median 0.7 mL/3.5 mg). The total volume of IL amphotericin B administered to the animals ranged from 0.5 to 3.8 mL (2.5–19 mg) (median 1.6 mL/8 mg). The maximum dose per visit exceeded the dose calculated for parenteral use in 24 (92.3%) cases.

Of the 16 cats that achieved clinical cure, a single application was effective in four; seven cats required two applications, four required three applications and one required five applications.

The adverse clinical effects observed during treatment were the formation of a sterile abscess in four (15.4%) and oedema in three (11.5%) animals, regardless of the volume of amphotericin B administered. Spontaneous abscess drainage was observed in two cases and needle puncture of the abscess was necessary in the remaining animal, which occurred 1 week after the amphotericin B infiltration. Seven (26.9%) animals presented with discomfort during infiltration of the drug into the nasal region, as indicated by head movement. Prolonged
post-anaesthetic recovery, lethargy and hyporexia were reported by the owner of one animal on the day after the amphotericin B applications.

The haematological parameters remained within normal limits. However, a mild increase in alanine aminotransferase levels appeared in five animals before and during the study, with no clinical signs suggestive of hepatotoxicity. The results obtained are summarised in Table 1.

Discussion

The present study describes IL administration of amphotericin B treatment of cats with sporotrichosis showing an unsatisfactory clinical response to itraconazole alone, although itraconazole has proved to be effective in cats and convenient for the owners. In this study, most animals had lesions in the nasal region. According to Malik et al., the nasal region of cats does not have a rich supply of blood nor does it possess easily accessible mobile skin, which would facilitate reconstructive surgical procedures. Thus, the cure of infections in this specific area may be problematic. Crothers et al. described the case of a cat that received itraconazole (10 mg/kg orally, daily for 4 years) until death and the lesions on the nose were still present at death. The cat died of an unrelated cause.

Amphotericin B has been indicated for the treatment of rapidly progressive or severe systemic mycosis, imidazole-resistant cryptococcosis and disseminated forms of sporotrichosis. Systemic administration of amphotericin B has been performed by intravenous or subcutaneous injection two or three times a week in a large volume of 2.5% dextrose and 0.45% sodium chloride for several weeks. However, the duration of intravenous amphotericin B therapy was limited since cats tend to develop cephalic and jugular vein thrombosis as well as nephrotoxicity. Three cats with sporotrichosis have been previously treated with intravenous amphotericin B without satisfactory results. For these reasons, IL rather than systemic administration of the drug was chosen in the present study.

Local infiltration of amphotericin B has been described for the treatment of human patients with localised fungal infections such as chromoblastomycosis, alternariosis and sporotrichosis as well as for the treatment of a horse with conidiobolomycosis. This route of administration is associated with high tissue concentrations, tissue conservation and few adverse systemic consequences. In the study of Gremião et al., three applications of IL amphotericin (5 mg/application) resulted in the successful treatment of a cat with a skin lesion on the nasal bridge that had resisted 9 months of itraconazole treatment, demonstrating that this therapeutic approach can be efficacious in certain cases of feline sporotrichosis. According to Malik, intralesional amphotericin B can be safely used as a sole or adjunctive therapy and thus is a useful therapy for the treatment of fungal infections of the skin and subcutis in cats, especially in veterinary medicine where the cost of medications or complex surgery can be a critical aspect of treatment.

We chose to administer IL amphotericin B in combination with oral antifungal treatment since studies have shown that the combined administration is more effective than application of the drug alone for the treatment of fungal infections, with a reduction not only in the time of treatment but also in the cumulative amount of amphotericin B necessary for cure. In the present study, preservative-free 1% hydrochloride was used as previously described. However, discomfort during infiltration of the drug was perceived in some animals. Other workers diluted amphotericin B in 2% or 0.5% procaine, or used 1% or 2% lidocaine. According to Whiting, amphotericin B is incompatible with local anesthetics and both procaine and lidocaine can cause precipitation of the drug. However, no precipitation of amphotericin B was observed in our study.

The ideal concentration of amphotericin B for local application is still unknown. Vieira et al. reported good results with a concentration of
Table 1. Summary of the results of treatment of 26 cats with sporotrichosis by intralesional administration of amphotericin B

<table>
<thead>
<tr>
<th>Case no.</th>
<th>Sex</th>
<th>Age (years)</th>
<th>Weight (kg)</th>
<th>Lesion location</th>
<th>Clinical aspect</th>
<th>No. of lesions</th>
<th>No. of AMB applications</th>
<th>Total amount of AMB (mg)</th>
<th>Time of follow up* (days)</th>
<th>Adverse effects</th>
<th>Outcome</th>
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<td>↑ ALT</td>
<td>LCR</td>
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<td>NP</td>
<td>Nodule</td>
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<td>2</td>
<td>1.2–6.0</td>
<td>7</td>
<td>Cure</td>
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<td>Abscess</td>
<td>Recurrence</td>
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AMB, amphotericin B; NB, nasal bridge; NP, nasal plane; RFL, right forelimb; LFL, left forelimb; LHL, left hindlimb; RP, right pinna; LP, left pinna; ALT, alanine aminotransferase; LCR, lack of clinical response.

*Time of follow-up was calculated after cessation of therapy until remission of the lesion.
IL amphotericin B of 1 mg/mL, whereas Clark\textsuperscript{25} successfully used 25 mg/mL. However, in the latter case the infiltrated areas became purulent after therapy. In the present study, we used a concentration of 5 mg/mL, as reported in other studies.\textsuperscript{22,23,26,30}

The infiltrations were applied either weekly or every other week according to the availability of the owners, and the administration frequency in regard to the response had no influence whatsoever. The applications were well tolerated by most animals, despite some cases of discomfort during infiltration of the drug in the animals, regardless of the application interval. Only one cat had a prolonged anesthetic recovery, presumably due to acepromazine. Some investigators performed applications twice weekly, weekly, or every other week.\textsuperscript{22,23,25,26}

In one study,\textsuperscript{25} the frequency of these injections was determined by the discomfort caused by the injection and the healing delay of the chromoblastomycotic lesion after each injection.

Injection of amphotericin B may cause a slight elevation of temperature and anorexia,\textsuperscript{23,25} and Costello et al.\textsuperscript{23} observed a systemic reaction characterised by chills and mild lymph node enlargement when an overdose of 40 mg amphotericin B was inadvertently injected. In the present study, blood and biochemical analysis disclosed no adverse effects in the cats, even when the maximum systemic dose was exceeded, in agreement with others\textsuperscript{29} who have observed no systemic consequences in a human patient with cutaneous alternariosis after IL administration of the drug. The asymptomatic increase in alanine aminotransferase in five animals might be attributed to the use of itraconazole.\textsuperscript{35,36}

In the present study, oedema and the formation of a sterile abscess at the site of IL infiltration occurred in some cases, irrespective of the volume of amphotericin B administered. Oedema has been described by Whiting as one of the disadvantages of the local administration of amphotericin B, since concentrated solutions of this drug act as tissue irritants. Adverse effects related to the local administration of large volumes of amphotericin B, such as local irritation or a sterile abscess, may occur.\textsuperscript{22} Nevertheless, these authors found no adverse effects in a cat with cryptococcosis successfully treated by IL injection of amphotericin B into the nasal region in combination with subcutaneous amphotericin B and oral antifungal agents. One study reported a severe local tissue reaction accompanied by pain, erythema and oedema after injection of an overdose of the drug,\textsuperscript{22} and in another, the areas became purulent shortly after the onset of IL therapy of a granulomatous lesion involving the entire helix and lobe of the left ear.\textsuperscript{25}

The present study confirmed the effectiveness of IL amphotericin B as previously reported.\textsuperscript{22,23,25,26,29} Treatment abandonment was due to non-compliance by owners. Clinical inactivity and recurrence were observed in animals with lesions in the nasal region, and lesions located in the nasal region are more difficult to treat.\textsuperscript{16} In addition, we believe that in cases of recurrence, tissue levels of the drug may have only been fungistatic,\textsuperscript{35} or treatment duration was insufficient, with viable fungal organisms persisting at the sites affected as in cases of feline cryptococcosis.\textsuperscript{36}

In conclusion, IL amphotericin B is a promising alternative for the treatment of feline sporotrichosis refractory to azolic antifungal agents and further clinical trials should be carried out to determine the ideal concentration/formulation of this drug for this application. Other studies such as cryotherapy should be encouraged as a complementary therapy in sporotrichosis with residual lesions refractory to systemic antifungal agents.

References

BOOK REVIEW


This is one of the newer handbook-style texts which attempt to meet the needs of busy practitioners who desire a readily available resource. As such, one has to forgive a book like this for deficiencies in depth of knowledge, but the importance of the accessibility of information then becomes paramount.

Each subject heading is listed in alphabetical order in the contents and by subject in the contents by system and index. Despite this, the text failed to achieve the imperative aim of maintaining a logical approach to categorising topics, given the field of emergency medicine. When I attempted to search for rodenticide toxicity, I failed to find it in the contents and contents by systems and, most disappointingly, it was only referenced twice in the index (as part of the differential in electric cord injury and as a risk factor in epistaxis). More glaringly, the index failed to cross-reference the major section on rodenticide toxicity, which is under the topic heading of Anticoagulant rodenticide toxicity. This could be forgiven, considering the potential disparity of nomenclature of presenting emergencies, but it highlights the need for accurate and comprehensive cross-referencing.

Potential discrepancies are further highlighted when considering the presentation of a patient with pulmonary oedema, where the veterinarian is faced with the perplexing inconsistency of having cardiogenic pulmonary oedema listed as a major chapter under ‘pulmonary oedema, cardiogenic’, whilst non-cardiogenic pulmonary oedema is listed under ‘non-cardiogenic pulmonary oedema’. This is exacerbated by the fact that non-cardiogenic pulmonary oedema is not listed in the contents by system under ‘respiratory’, nor indeed under any system, unlike its cardiogenic counterpart.

Disappointingly, even the promise of alphabetical listing is sometimes not met, with the chapter topic ‘pulmonary thromboembolism’ immediately preceding ‘pulmonary contusions’. In addition, many of the topics are titled in a way (i.e. non-cardiogenic pulmonary oedema) that being able to find them is dependent on a large component of the investigative process already being done, creating a veterinary catch 22.

The struggle to find the appropriate topic overshadows what is otherwise a text with excellent content. The details of each section are by and large up to date and are set out in such a way, with bullet points and clear headings, that makes the information easily digestible. However, my sense of humour was piqued to read that the rate of cardiac compressions is optimised when performed to the beat of the song Staying Alive, and could not help but wonder what more recent graduates do when faced with an arresting patient (presumably dying from ‘Saturday Night Fever’). Equally, one suspects a fleeting nod to James Bond when it is suggested that cats with FIP have a clot forming effusion which froths when ‘shaken, not stirred’.

Overall, this text forms a useful addition to the library of any veterinary practitioner, especially those with an interest in emergency medicine. Whilst the problems with indexation and referencing are significant in a text whose usefulness is contingent on speed and ease of access, this problem may be minimised if the text entices the reader to engender a familiarity that helps them successfully navigate the book. Whether the adrenaline-driven environment of the ER is the setting to test such patience will ultimately determine the popularity of this text in practice.

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