Cutaneous leishmaniasis: new developments in diagnosis and treatment evaluation
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First reported case of disseminated cutaneous leishmaniasis caused by *Leishmania (Leishmania) amazonensis* infection in Suriname

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Submitted
Summary

First case of disseminated cutaneous leishmaniasis caused by *Leishmania (Leishmania) amazonensis* was identified in Suriname using molecular and isoenzymatic assays. The patient was successfully treated with miltefosine, and followed-up with QT-NASBA. This is the first reported case of infection with a different *Leishmania* species from *L. (Viannia) guyanensis* in Suriname.
Introduction

*Leishmania*, a protozoan parasite, causes a wide spectrum of diseases in humans and wild or domestic mammals. Leishmaniasis is one of the most important vector-born tropical diseases targeted by the World Health Organization (WHO) with 12 million people affected every year. In the New World (an area ranging from the South of the United States down to the Rio de Janeiro region in Brazil) the disease is caused by at least ten species of the *Leishmania* and *Viannia* sub-genera. While most species, like for example *Leishmania (Viannia) guyanensis*, cause localized self-limiting ulcer, two percent of patients infected with *L. (V.) braziliensis* develop muco-cutaneous leishmaniasis (MCL), with spread of the parasite to the facial mucosae (nasal, oral, pharyngeal and even laryngeal mucosae). Parasite species of *Leishmania* sub-genus can cause anergic diffuse (ADCL) or disseminated cutaneous leishmaniasis (DCL), with extensive spread of the parasite over the skin. MCL, ADCL and DCL can cause severe patient disfigurement.

Currently, there are only a few documented studies on cutaneous leishmaniasis (CL) in Suriname, a country situated in the Amazon basin of South America. Mean CL incidence from 1979-1985 was estimated to be 4.9 per 1,000 inhabitants for the rainforest interior of the country, and 0.66 per 1,000 as a whole. Infections are most often encountered in humans who work in the forest (gold miners, woodcutters, soldiers), tourists and hunters. So far, only *L. (V.) guyanensis* is described as causative agent of CL in Suriname. Three known vectors of CL are found in the country, *Lutzomyia umbratilis*, *Lu. flaviscutellata* and *Lu. whitmani*, of which the first is known to be the principal vector of *L. (V.) guyanensis*. While *Lu. flaviscutellata* and *Lu. whitmani* are proven vectors of clinical relevant species (*L. (L.) amazonensis* and *L. (V.) braziliensis*), these *Leishmania* species have not been detected up till now in Suriname. However, patients with clinical symptoms typically associated with *L. (L.) amazonensis* and *L. (V.) braziliensis* (like satellite lesions and mucocutaneous involvement) have been encountered, suggesting that other *Leishmania* species do occur.
Case report

A 17-year-old man was presented at the Dermatology Service in Paramaribo with multiple cutaneous ulcerations, nodules and fibrotic plaques disseminated in his face and on his limbs and trunk. Subcutaneous nodules in the course of lymph draining tracts on arms and legs suggested an infectious process with lymphatic dissemination. The patient had acquired the infection at around the age of 5 and came from an inland village Duwatra, located at the Brokopondo lake (central-east Suriname) and had never resided outside the country. The patient received intramuscular pentamidine therapy in 1997, 1998 and 2005, without sustained clinical effect. Because the diagnosis CL was doubted, the patient had also been treated for borderline lepromatous leprosy. In 2006, the diagnosis CL was confirmed with histopathology, culture and PCR. Parasites were cultured and the strain was sent for species identification by multilocus enzyme electrophoresis (MLEE) to the National Reference Centre of *Leishmania* (Montpellier, France). The parasite was also identified by a PCR-restriction fragment length polymorphism method on the ribosomal small subunit and internal transcribed spacers genes (PCR-RFLP SSU-ITS).8

Following promising results obtained with miltefosine treatment in an ADCL patient in Venezuela9, the patient received 150 mg/day oral miltefosine (Impavido®, Zentaris, Germany) for 98 days and was parasitological followed with QT-NASBA to monitor decrease in parasite loads in his lesions. Skin biopsies were collected from one target lesion before treatment, during treatment at day 14, day 28, day 42 (all in duplo) and at day 70 (single biopsy sample). DNA/RNA was extracted and tested in QT-NASBA assay as described before.10

Discussion

The strain causing the infection (MHOM/SR/2006/SP100) was identified as *Leishmania (Leishmania) amazonensis* by PCR-RFLP (Figure 1) and the enzymatic profile was equal to *L. (L.) amazonensis* zymodeme MON-41.
Histopathology showed large macrophages containing abundant *Leishmania* amastigotes and scattered lymphocytes and plasma cells. There was no granuloma formation. A significant clinical improvement was observed during the first two months of therapy with lesions slowly decreasing in size and induration. At day 70, all ulcerative lesions were completely re-epithelialized, without signs of infiltration or lymphangitis. At start of treatment parasite counts of 360,000 and 310,000 parasites per biopsy were detected with QT-NASBA and parasite counts decreased during treatment until 0 parasites/biopsy at day 70 (Figure 2). Histopathology at day 70 revealed no *Leishmania* bodies, but nodular lymphocyte and plasma cell infiltrate and fibrosis. While mild elevation of creatine and urea was observed during the course of treatment no subjective or adverse side effects were reported.

*L. (L.) amazonensis* is known to cause different clinical forms of cutaneous leishmaniasis (CL) of which disseminated (DCL) and anergic diffuse cutaneous leishmaniasis (ADCL) are the most serious manifestations. Both forms are characterized by the presence of heavily parasitized macrophages and a negative delayed hypersensitivity skin-test reaction prior to treatment. DCL is an intermediate form between localized CL and ADCL. Prior to treatment DCL patients show the presence of lymphocytes and plasma cells in histopathology, and during treatment the macrophage reaction will be replaced by a
nodular lymphocyte and plasma cell infiltrate. The histopathological picture in skin sections of our patient corresponded with this description. Furthermore, the patient showed lymphatic dissemination of infection and presence of erythematous infiltrated plaques, both characteristics of DCL.³

![Log QT-NASBA results during miltefosine treatment](image)

**Figure 2.** Log QT-NASBA results of duplicate skin biopsies collected from the same lesion during treatment. End = end treatment.

In contrast to ADCL, DCL seems to have a good prognosis of cure, even for more difficult patients who were unsuccessful in earlier treatments, with total restoration of the cell-mediated immune response.³ Pentamidine, the only available treatment in Suriname for CL⁵, had been administered to the patient without sustainable clinical effect. While initially promising results had been described with miltefosine in an ADCL patient⁹, in a following clinical study 15 out of 16 ADCL patients presented new lesions after suspension of treatment.¹¹ In this study, the DCL patient showed a similar spectacular clinical improvement during the course of miltefosine treatment with complete remission at day 70. At this time point, no *Leishmania* parasites were detected with histopathology and QT-
NASBA. The patient was followed 58 days (Figure 3) and seven months after end of therapy, and so far the patient did not develop new lesions and histopathology remained negative. However, clinical follow-up of these patients remains of great importance.

Figure 3. Infiltrated lesions on the hands before (a) and 58 days after treatment (b)

In general, \textit{L. (L.) amazonensis} infection is rare in humans.\textsuperscript{12} Bordering countries like French Guiana on the eastern side of Suriname, report very few patients (around 1.9\%) infected with this species.\textsuperscript{12} It seems that the sand fly vector of \textit{L. (L.) amazonensis}, \textit{Lu. flaviscutellata}, is not attracted to man and not active during the day.\textsuperscript{13} From an earlier study it was known that this vector is present in Suriname.\textsuperscript{4} Whether the parasite \textit{L. (L.) amazonensis} is circulating in Suriname was not known up until now.

Many gold diggers from the northern part of Brazil work and travel in Suriname and are familiar with CL. In the State of Pará in Brazil, a region bordering Suriname in the south, the infection rate with \textit{L. (L.) amazonensis} is higher (34.8\%)\textsuperscript{14} and it is conceivable that infected gold diggers from that area have introduced \textit{L. (L.) amazonensis} into Suriname. Our patient used to live in a village where many Brazilian gold diggers worked around the time he got infected. Migration of labourers is associated with an increased risk for CL infection.\textsuperscript{15} However, the zymodeme MON-41 seems to be wide-spread in Central and northern part of South America, since the same zymodeme has also been reported in
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Venezuela, Brazil, Panama, French Guiana and Colombia (Pratlong F. & Dedet J.P., Montpellier International Cryobank of Leishmania, 2007). For this reason, speculations on the origin of the infection described here can only be done cautiously. Further research is necessary to identify the number of infected patients with the same or other clinical relevant species in Suriname, and the possible need for implementation of other medicines.

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Chapter 6

References

3 Silveira FT, Lainson R, Corbett CE. Further observations on clinical, histopathological, and immunological features of borderline disseminated cutaneous leishmaniasis caused by Leishmania (Leishmania) amazonensis. Mem Inst Oswaldo Cruz 2005; 100: 525-34.
14 Silveira FT, Lainson R, Shaw JJ, De Souza A, Ishikawa E, Braga R. Cutaneous leishmaniasis due to Leishmania (Leishmania) amazonensis in
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