Oral Manifestations in the American Tegumentary Leishmaniasis

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Abstract

Introduction: American tegumentary leishmaniasis (ATL) can affect the skin or mucosa (mucocutaneous leishmaniasis – MCL) including the oral cavity. MCL oral lesions are often confused with other oral diseases, delaying diagnosis and specific treatment, and increasing the likelihood of sequelae. Thus, increasing the knowledge of the evolution of ATL oral lesions can facilitate its early diagnosis improving the prognosis of healing.

Objectives: Evaluate the frequency of ATL oral lesion and describe its clinical, laboratory and therapeutic peculiarities.

Methods: A descriptive transversal study was carried out, using data from medical records of 206 patients with MCL examined at the outpatient clinics-IPEC-Fiocruz between 1989 and 2013. Proportions were calculated for the categorical variables and the association among them was assessed by the Pearson’s chi-square test. Measures of central tendency and dispersion were used for the continuous variables and their differences were assessed by both parametric (t test) and non parametric (Mann-Whitney) tests. P-values <0.05 were considered as significant.

Results: The most affected site was the nose, followed by the mouth, pharynx and larynx. Seventy eight (37.9%) have oral lesions and the disease presented a lower median of the evolution time than in other mucous sites as well as an increased time to heal. The presence of oral lesion was associated with: the presence of lesions in the other three mucosal sites; a smaller median of the leishmanin skin test values; a longer healing time of the mucosal lesions; a higher recurrence frequency; and a smaller frequency of treatment finishing and healing. When the oral lesion was isolated, it was associated with an age 20 years lower than when the oral lesion was associated with other mucosal sites.

Conclusion: Considering the worst therapy results associated with the presence of oral lesions, we suggest that lesions in this location represent a factor of worse prognosis for MCL.

Introduction

American tegumentary leishmaniasis (ATL) is a parasitic infectious disease transmitted by protozoa of the Leishmania genus, through the bite of a vector (insect) of the Lutzomyia genus [1]. Eighty eight countries in the world are affected by leishmaniasis. In the Americas, ATL is distributed from the South of the United States to the South of Argentina [2,3]. In Brazil (2012) the number of ATL cases was 15.731 and the detection coefficient 11.1 cases/100.000 [4]. The state of Rio de Janeiro is traditionally acknowledged as an ATL endemic area and the infectious agent was identified as L. (Viannia) braziliensis [5].

The most common ATL clinical manifestation is the localized cutaneous form which appears as an ulcerated lesion with granular base and raised borders, which might resolve even without treatment. However, some patients infected by L. (V.) braziliensis (1 to 10%) develop mucocutaneous leishmaniasis (MCL) which usually presents gradual tissue destruction associated with intense inflammatory response [6–8], affecting the upper respiratory and
digestive tracts mucosa. The mucosal lesion usually emerges weeks or years after the initial skin lesion has healed, by probable blood spread from the primary focus [9,10]. However, a mucosal lesion may appear when a skin ulcer is active [11]. The lesions in the mouth are usually associated with nasal involvement, but it is possible to find one or more lesions only in the oral mucosa. These lesions occur more often on the lip and palate, although lesions in the uvula, gums, tonsils and tongue were already identified. They are characterized by ulcerative-vegetative lesion accompanied by coarse granulations. Patients usually complain of pain, dysphagia and odynophagia [12,13]. Besides the difficulty in identifying parasites, MCL is often mistaken with other benign or malignant lesions that affect the mucosal tissue. Thus, it is necessary to use several methods to confirm diagnosis such as: serology, histopathology, *Leishmania* culture and molecular methods [14,15]. Isolated or associated, nasal lesions occur at over than 90% of the cases. Consequently, the MCL diagnosis is based on the investigation of nasal lesions and treatment usually starts without oral lesions investigation and diagnosis. For this reason, literature on this subject is scarce, hampering a better understanding and the diagnosis of this type of mucosal lesion.

In this context, the objective of the present study is to evaluate the frequency of ATL oral lesion occurrence and describe their clinical, laboratory and therapeutic peculiarities.

**Methods**

This retrospective study was conducted by reviewing the medical records of 206 patients with Mucocutaneous Leishmaniasis monitored at the Ambulatory/Laboratory for Leishmaniasis Surveillance of the Evandro Chagas Clinical Research Institute, Rio de Janeiro between 1989 and 2013. The study was approved by the Ethics in Research Committee of IPEC/FIOCRUZ (CAAE: 09994613.6.0000.5262) after signing a Statement of Commitment by the researchers involved who have undertaken to keep confidential the identity of patients, and the confidentiality and privacy data. All procedures performed in patients followed a protocol of care, which in 2002 was submitted and approved by the Ethics in Research Committee of IPEC/FIOCRUZ (CAAE: 0016.0.009-02), entitled "Study for the systematic assessment of the patients with American Tegumentary Leishmaniasis in Leishmaniasis Reference Center - Instituto Evandro Chagas Clinical Research - Fiocruz". Thereafter, all patients sign a Statement of Informed Consent to carry out the clinical protocol and biological sample collection and storage.

ATL diagnosis was established by two or more of the following parameters: consistent epidemiological history, positive reaction in the leishmanin skin test (LST) and the identification of *Leishmania* genus by imprint, culture or histopathologic examination. To assess the presence and location of mucosal alterations of the upper airways tract, all the patients included had been submitted to evaluation of the upper respiratory tracts mucosa by a 30 degrees Karl Storz rigid endoscope, and a 70 degrees Karl Storz rigid videolaryngoscope (Tuttlingen, Germany) and the obtained images were stored through pictures or films.

Socio-demographic (age, gender, occupation and probable infection place), clinical (clinical classification - Table 1 -; symptoms; lesion number, location and characteristics; LST and presence of sequelae), laboratorial (diagnostic methods) and therapeutic (drug used, dose and treatment time) data were collected from the medical records.

Proportions were calculated for the categorical variables. The times of mucosal lesion evolution and treatment did not present normality in the Shapiro-Wilk test, indicating the use of a non-parametric approach. Thus, median (Md) and interquartile range were supplied for those variables (IQR) and for the variable age and LST, mean ± standard deviation were supplied. The association between the categorical variables was assessed by the Pearson’s chi-square test. The difference of the values of the continuous variables was assessed by the t test, in the parametric case and by the Mann-Whitney test in the non-parametric cases. P values smaller than 0.05 indicate statistically significant tests. The Statistical Package for Social Sciences (SPSS) version 16.0 (IBM Company) was used for data analysis.

**Results**

We studied 206 patients with MCL, with a mean age of 51.72 ± 17.61 years, with 72.8% males, 85.4% having acquired the infection in Brazilian Southern region and 24.6% farmworkers.

Table 2 shows the clinical and therapeutic peculiarities of the 206 patients with MCL. Concomitant mucosal was the most frequent form. The most affected site was the nose, followed by the mouth, pharynx and larynx. The oral lesions presented a lower median of the evolution time than in other mucosal sites as well as an increased time to heal. One hundred sixty-four patients (79.6%) completed treatment and 130 (63.1%) were followed up to one year, when 121 (93%) were healed and 9 (7%) presented recurrence.

From the 206 patients with MCL, 78 (37.9%) patients have oral lesions, with a mean age of 50.2 ± 17.19 years and 23.1% were female and 76.9% male. Table 3 shows the clinical, laboratory and therapeutic peculiarities of the 78 patients with oral lesions. Among them, 71 patients (91%) had oral lesion associated with lesions at other mucosal site and 7 (9%) did not present associated lesions. The number of lesions in the oral cavity varied from 1 to 4 (median = 1). The LST and serology were positive in most of the patients (respectively 97.1% and 75.7%). Culture and imprint were more frequently positive than the histopathology (Table 3). The histopathological examinations detected cellular infiltrate in all oral lesions. In addition, 12.3% presented granules, ulcer and/or tissue destruction. Of the total 78 patients, seven (9%) had already received previous treatment: five with meglumine antimoniate, one with amphotericin B and the other with unknown treatment. Of 57 patients followed up to one year, 48 (84.2%) were healed.

Twenty-three patients presented oral sequelae (destruction of anatomical structures) after completing treatment, with the uvula as the most frequently affected site in 19 patients, followed by the soft palate in 14 patients.

The clinical, laboratory and therapeutic characteristics of 206 patients with mucocutaneous leishmaniasis was compared as regard the presence or not of oral lesion, and the results are shown in Table 4. The presence of oral lesion was associated with the presence of lesions in the other three upper respiratory mucosal anatomic sites (Nasal p = 0.008; Larynx p = 0.003 and Pharynx p < 0.001), as well as with a greater recurrence frequency (p = 0.003) and smaller occurrence of treatment finishing (p < 0.001) and healing (p < 0.001). We also observed association of the presence of oral lesion and a smaller median value of the LST diameter (p = 0.015), shorter time of mucosal lesion evolution (p = 0.007) and longer time necessary for healing the mucosal lesion in any site (p < 0.001) as well as when only the nasal lesion is considered (p = 0.029).

Some clinical, laboratory and therapeutic characteristics of 78 patients with oral leishmaniasis was compared as regard the presence or not of isolated oral lesions, and the results are shown in
Table 5. We verified an association between isolated oral lesion and a shorter time of mucosal lesion evolution ($p = 0.03$) and smaller LST ($p = 0.025$). Regarding age, a significant difference ($p = 0.002$) was found when the two groups were compared (isolated oral lesion = $31.71 \pm 10.42$ years and oral lesions in the presence of other associated mucosal lesions = $51.93 \pm 16.69$ years).

Table 6 shows some clinical, laboratory and therapeutic characteristics of 206 patients with mucocutaneous leishmaniasis compared as regard the presence or not of isolated nasal lesions.

Table 1. Clinical classification of mucocutaneous leishmaniasis [18].

| Clinical classification                          |  |
|-----------------------------------------------|---|
| Concomitant mucosal form                      | Skin and mucosal lesions occurring at the same time. |
| Mucosal form of undetermined origin           | Mucosal lesions without evidence of previous cutaneous form. |
| Late mucosal form                             | Mucosal lesions with evidence of previous cutaneous form. |
| Contiguous mucosal form                       | Periorificial facial skin lesions expanding into mucosal area. |

Table 2. Clinical and therapeutic characteristics of 206 patients with mucocutaneous leishmaniasis. INI FIOCRUZ, 2014.

| Variables                                      |  |
|------------------------------------------------|---|
| **Clinical classification**                    |  |
| Concomitant mucosal form                       | 78(n) 37.9% |
| Mucosal form of undetermined origin            | 55(n) 26.7% |
| Late mucosal form                              | 52(n) 25.2% |
| Contiguous mucosal form                        | 21(n) 10.2% |
| **Affected sites**                             |  |
| Nasal cavity                                   | 188 91.3% |
| Oral cavity                                    | 78 37.9% |
| Pharynx                                        | 64 31.4% |
| Larynx                                         | 62 30.1% |
| **One site affected**                          |  |
| Nasal cavity                                   | 104(n) 50.5% |
| Oral cavity                                    | 92(n) 44.7% |
| Larynx                                         | 7(n) 3.4% |
| **Two sites affected**                         |  |
| Nasal cavity                                   | 42(n) 20.5% |
| Oral cavity                                    | 36(n) 17.5% |
| Larynx                                         | 24(n) 11.7% |
| **Time of mucosal lesions evolution before diagnosis (months)** |  |
| Nasal cavity                                   | 24 Md 6–60 IQR |
| Oral cavity                                    | 24 Md 6–60 IQR |
| Pharynx                                        | 8 Md 3–29 IQR |
| Larynx                                         | 12 Md 6–26.5 IQR |
| **Treatment**                                  |  |
| Meglumine antimoniate                          | 193 93.7% |
| $5 \text{mgSb}^{5+}/\text{kg/day}$             | 165 86.4% |
| $10–20 \text{mgSb}^{5+}/\text{kg/day}$         | 26 13.6% |
| Amphotericin B                                 | 7 3.4% |
| Other drugs                                    | 6 2.9% |
| **Healing time of the mucous lesions (days)**  |  |
| Nasal cavity                                   | 98 Md 60–178 IQR |
| Oral cavity                                    | 90 Md 58–155.25 IQR |
| Pharynx                                        | 113 Md 59–205 IQR |
| Larynx                                         | 91 Md 59.5–172.25 IQR |
| Larynx                                         | 78.5 Md 55.25–223 IQR |
healing times inversely related to those observed in the oral lesion. We observed association between isolated nasal lesion and a longer time of mucosal lesion evolution (p = 0.024), a shorter time for mucosal lesion healing (p < 0.001) and shorter time for nasal lesion healing (p = 0.007).

The healing time was longer in the presence of laryngeal lesion (median = 121.5 days) than in its absence (median = 92 days) (p = 0.020). Presence or absence of pharyngeal lesions did not show any significant difference in the parameters analyzed.

**Discussion**

When we evaluated 78 patients with oral leishmaniasis we observed that this mucosal site was the second most frequently affected by the disease, and was associated with lesions in other upper respiratory tract mucosal sites, a shorter evolution time and factors of worse prognosis such as: longer healing time, higher recurrence frequency and smaller frequency of treatment finishing and healing. When the oral lesion was isolated, it was associated with an age 20 years lower than when the oral lesion was associated with other mucosal sites.

In accordance with the literature, our patients' group with MCL were majority composed by males aged 50 years or more. However, we identified that the concomitant mucosal form had the highest prevalence, followed by the mucosal form of undetermined origin and the late mucosal form, which is considered by the literature as the most prevalent, was only third in occurrence. This difference might be related to the

| Table 3. Clinical, laboratory and therapeutic characteristics of 78 patients with oral leishmaniasis lesions. INI FIOCRUZ, 2014. |
|-----------------|-----------------|-----------------|
| Variables       | Clinical classification |
|                 | Oral lesions | 78 (n) | 100% |
|                 | Concomitant mucosal form | 34(n) | 43,6% |
|                 | Mucosal form of undetermined origin | 18 (n) | 23,1% |
|                 | Late mucosal form | 18(n) | 23,1% |
|                 | Contiguous mucosal form | 8(n) | 10,2% |
|                 | Oral lesions associated with other mucosal site lesion | 71(n) | 91% |
|                 | Concomitant mucosal form | 30(n) | 42,3% |
|                 | Mucosal form of undetermined origin | 18(n) | 25,4% |
|                 | Late mucosal form | 18(n) | 25,4% |
|                 | Contiguous mucosal form | 5(n) | 7% |
|                 | Isolated Oral lesions | 7(n) | 9% |
|                 | Concomitant mucosal form | 4(n) | 57,1% |
|                 | Contiguous mucosal form | 3(n) | 42,9% |
|                 | Leishmanin skin test (N = 66) | 27,36(X) | ±19,63(S) |
|                 | Serology (N = 66) |
|                 | Positive | 50 (n) | 75,7% |
|                 | Negative | 16 (n) | 24,3% |
|                 | Histopathology (N = 45) |
|                 | Presence of amastigote forms | 24(n) | 53,3% |
|                 | Imprint (N = 13) |
|                 | Positive | 9(n) | 69,2% |
|                 | Negative | 4(n) | 30,8% |
|                 | Culture (N = 34) |
|                 | Positive | 22(n) | 64,7% |
|                 | Negative | 5(n) | 14,7% |
|                 | Contaminated | 7(n) | 20,6% |
|                 | Treatment |
|                 | Meglumine antimoniate | 71(n) | 91% |
|                 | 5 mgSb³⁺/kg/day | 58(n) | 82,9% |
|                 | 10 mgSb³⁺/kg/day | 4(n) | 5,7% |
|                 | 15 mgSb³⁺/kg/day | 5(n) | 7,1% |
|                 | 20 mgSb³⁺/kg/day | 3(n) | 4,3% |
|                 | Amphotericin B | 4(n) | 5,2% |
|                 | Not follow treatment | 3(n) | 3,8% |

N- total number, n-valid numbers, X - mean, S - standard desviation.
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systematic examination conducted on our service, where all the patients suspected of ATL were examined by a multidisciplinary team which included dermatologists, infectious disease specialists, otorhinolaryngologists and dentists. Regardless of the complaints, the patients were submitted to upper respiratory tracts endoscopy allowing an early diagnosis of the mucosal lesions. The conduct of mucosa systematic examination recommended in 1922 by Klotz and Lindenberg, is rarely followed, remaining restricted to punctual initiatives, such as the case of Boaventura et al. (2006) [16] where the concomitant mucosal lesion was observed in 2.7% cutaneous leishmaniasis (CL) cases [16].

Taking into consideration that the oral cavity have a shorter evolution time than the other mucosal sites, the systematic examination might have favored the identification of the mouth as the second most frequent site. Most studies do not even mention the oral cavity as a location, merely describing isolated regions of the mouth such as palate and lips or associating it with the pharynx [10–19]. In addition, the association with conditions that favor local inflammation and infection such as poor oral hygiene [21] might mask the ATL lesions, thus explaining the small frequency of oral involvement reported in literature.

Isolated nasal cavity lesions showed the greatest time of evolution, suggesting that the nose can singly evolve for a long time without requiring medical attention. We suggest that the location of the lesions in the oral cavity may lead to greater discomfort of the patient who would seek for medical care earlier. Isolated nasal cavity lesions showed the greatest time of evolution, suggesting that the nose can singly evolve for a long time without requiring medical attention. We suggest that the location of the lesions in the oral cavity may lead to greater discomfort of the patient who would seek for medical care earlier. It is possible that the association of lesions at nasal cavity with other upper respiratory tract sites is not necessarily the natural history of the disease, but a more severe outcome in certain patients [10,22,23].

### Table 4. Comparison of the presence or not of oral lesion with clinical, laboratory and therapeutic characteristics of 206 patientes with mucocutaneous leishmaniasis. INI FIOCRUZ, 2014.

| Presence of Oral Lesion | Yes (N = 78) | No (N = 128) | p |
|-------------------------|-------------|-------------|---|
| Variables               | n           | n           |   |
| Male                    | 60 76,9%    | 90 70,3%    | 0,301 |
| Presence of Cutaneous Lesion | 42 53,8% | 56 43,8% | 0,159 |
| Presence of Cutaneous Scar | 23 29,5% | 46 35,9% | 0,341 |
| Presence of Nasal Lesion | 66 84,6% | 122 95,3% | 0,008 |
| Presence of Laryngeal Lesion | 33 42,3% | 29 22,7% | 0,003 |
| Presence of Pharyngeal Lesion | 46 59,0% | 18 14,3% | <0,001 |
| Recurrence              | 21 30,9%    | 14 12,6%    | 0,003 |
| Treatment Finishing     | 53 70,7%    | 111 91,0%   | <0,001 |
| Healing after one year  | 48 84,2%    | 82 98,8%    | 0,001 |
| Mucosal Lesion Evolution (months) | 73 12 Md | 101 24 Md | 0,007 |
| LST (major diameter)    | 66 21 Md    | 109 30 Md   | 0,015 |
| HT of Mucosal Lesions (days) | 61 125 Md | 106 80,50 Md | <0,001 |
| HT of Nasal Lesions (days) | 54 116,5 Md | 100 80,50 Md | 0,029 |
| HT of Laryngeal Lesions (days) | 25 77 Md | 23 80 Md | 0,773 |
| HT of Pharyngeal Lesion (days) | 38 91 Md | 10 96 Md | 0,812 |
| Treatment Duration (days) | 75 51 Md | 123 41 Md | 0,217 |

N- total number, n-valid numbers, Md - median, **bold- significant p value.**

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### Table 5. Evolution of the mucosal lesions, leishmanin skin test and healing times of 78 patients with oral leishmaniasis compared as regard the presence or not of isolated oral lesions by the Mann-Whitney test with p value. INI-FIOCRUZ, 2014.

| Presence of Isolated Oral Lesion | Yes (N = 7) | No (N = 71) | p |
|----------------------------------|-------------|-------------|---|
| Variables                        | n | Median | n | Median |   |
| Mucosal Lesion Evolution (months) | 7 | 4 | 66 | 20 | 0,03 |
| LST (major diameter)             | 6 | 14 | 60 | 22,5 | 0,025 |
| HT of mucosal lesions (days)      | 6 | 116,5 | 55 | 135 | 0,934 |
| HT of Oral Lesions (days)        | 6 | 116,5 | 57 | 109 | 0,456 |
| Treatment Duration (days)        | 7 | 66 | 68 | 50,5 | 0,623 |

N- total number, n-valid numbers, LST - leishmanin skin test, HT- healing time, **bold- significant p value.**

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Additionally, the age of the patients with isolated oral lesions was 20 years lower than the observed in patients with lesions in other sites, which may be due to the fact that isolated oral lesions were found in patients who sought medical attention for investigating active cutaneous lesions and not with mucosal lesions at a later stage evolution. It is possible that mucosal lesions, at least in some cases, have insidious and early onset, just passing unnoticed and being diagnosed as CL, only being later mistakenly diagnosed as mucosal late form.

Regarding the diagnosis of the oral lesion, we could not find in literature studies describing results obtained exclusively in this site. However, when we compare it with the general diagnosis of MCL diagnosis, we observe similarity of isolation rates in culture, although a higher detection of amastigotes in the histopathologic examination and imprint. [24,25] ELISA showed reactivity higher than 90% as mentioned in literature. [26] It was already reported that lesions with shorter evolution time have more parasites [27] suggesting that the early diagnosis carried out at our service might have favored the number of positive exams; in addition it is possible that the oral lesion presents a more significant parasitism than the nasal lesion that favor a influx of parasites through spread by the blood system and the higher number of potentially host cells of the parasite.

The median of the LST values in patients with oral lesion was 9 mm smaller than of those without oral lesion. Additionally, despite the shorter evolution time of oral mucosal lesions until their diagnosis, patients that had lesions in this location, presented longer times for general mucosal and nasal mucosal healing, 45 and 36 days respectively than the patients without oral lesion. In addition, the presence of oral lesion was associated with a higher number of recurrences and a smaller frequency of treatment finishing and healing up to one year after treatment. The occurrence of therapy failure in CL was associated with a short time of lesion evolution (less than 2 months), before the beginning of treatment, and a smaller LST intensity, which could reflect a weak cell immune response. [28] The presence of oral lesion in ATL was also associated with a reduction of food intake, with consequent malnutrition and longer time for lesion healing. [29]

And, finally, the use of small doses of meglumine antimoniate, different from what is indicated in literature [18] proved to be efficient in MCL patient treatment including the oral location.

As conclusion, our results suggest that oral involvement in ATL is associated with worse therapeutic results and may be considered as a factor of worse prognosis in its mucosal form.

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Author Contributions

Conceived and designed the experiments: DCSC MRP JSM ACCM AFS MFM LPQ EMC CMVR. Performed the experiments: DCSC MRP JSM ACCM AFS MFM LPQ EMC CMVR. Analyzed the data: DCSC MRP JSM ACCM AFS MFM LPQ EMC CMVR. Contributed reagents/materials/analysis tools: DCSC MRP JSM ACCM AFS MFM LPQ EMC CMVR. Wrote the paper: DCSC MRP JSM ACCM AFS MFM LPQ EMC CMVR.

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