Histopathologic results before and after treatment of actinic cheilitis with 5% topical fluorouracil

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Abstract
Introduction
The aim of this study was to evaluate the clinical and histological effect of fluorouracil 5% cream on actinic cheilitis of lower lip.

Materials and methods
Fluorouracil cream was prescribed for 14 consecutive days with the establishment of clinical and histopathological diagnosis of actinic cheilitis. After a healing period of 45 days from the end of treatment with fluorouracil, a new biopsy was performed to compare the features before and after treatment.

Results
These preliminary results showed that within the estimated period of the study the fluorouracil cream 5% did not show the expected effectiveness, since it did not eliminate the histopathologic changes, nor improved the clinical condition of the lower lip.

Conclusion
In the period and dosage recommended in this study, fluorouracil 5% cream did not produce the awaited effects over lesions of actinic cheilitis since it does not improve the clinical and histopathological pre-existent conditions.

Since it is impossible to predict which lesions may progress to invasive SCC. Therefore, common census suggest the treatment of all lesions mainly those that bring any degree of epithelial dysplasia. Several forms of treatment for AC have been described from surgical to non-surgical; however, an ideal treatment for premalignant lesions should be effective in resolving the lesion and easily administered, with few adverse effects, thus avoiding the morbidity of surgical resection. For this reason, other options have been developed for the treatment of AC such as topical treatments.3,4,8–11,13–17. Due to the high incidence of AC and SCC of the lip in the general Brazilian population, the objective of this study was to evaluate the effect of 5-fluorouracil (5-FLU) over clinical and histopathological features of AC of lower lip.

Material and methods
This study was submitted and approved by the local Ethical Committee (CEP UNOESC-HUST) under number 148/2011. It included patients who had good clinical condition for biopsy and who were clinically diagnosed with AC in the lower lip. Pregnant patients, patients with allergic reactions to the components of 5-FLU and patients with histopathological diagnosis of SCC in any area of the lower lip were excluded.

The clinical diagnosis criteria include three or more of the following conditions:3,4,8–10 (a) clinical atrophy, (b) dryness, (c) scaly lesions, (d) swelling of the lip, (e) erythema, (f) ulceration, (g) blurred demarcation of the vermilion border and skin, (h) white spots or plaques, (i) crusts, (j) symptoms, (l) blotty areas and (m) areas of pallor.
For histopathological aspects, the following conditions were assessed:

(a) architecture: (a.1) irregular epithelial stratification, (a.2) loss of polarity of basal cells, (a.3) droplets of rete ridges, (a.4) increased number of mitotic figures, (a.5) abnormal superficial mitoses, (a.6) premature keratinisation in single cells (dyskeratosis), (a.7) keratin pearls within rete pegs and (b) cytology: (b.1) abnormal variation in nuclear size (anisokaryosis), (b.2) abnormal variation in nuclear shape (nuclear pleomorphism), (b.3) abnormal variation in cell size (anisocytosis), (b.4) abnormal variation in cell shape (cellular pleomorphism), (b.5) increased nuclear-cytoplasmic ratio, (b.6) increased nuclear size, (b.6) atypical mitotic figures, (b.7) increased number and size of nucleoli and (b.8) hyperchromasia.

The study steps followed: (1) clinical diagnosis; (2) first biopsy and histopathological features (H&E staining); (3) treatment of the lesions with topical cream with FLU 5%, commercially available (two times day, 14 days); (4) clinical follow-up and clinical re-evaluation 45 days from the end of treatment and (5) second biopsy (45 days from the end of treatment) followed by histopathological re-evaluation (H&E staining). This second biopsy was performed immediately adjacent to the first.

The histopathological evaluations were conducted by two oral and maxillofacial pathologists and performed independently, blinded to the results before and after treatment. Disagreements were resolved by consensus between the evaluators through re-evaluation of the slides.

Results

Two patients were selected for this study (case 1 and case 2). Case 1 is a female patient who is a 35-year-old farmer with previous history of uterine cancer and case 2 is a male patient who is a 45-year-old farmer, with inconspicuous clinical history.

No significant improvement of clinical lip condition was observed after treatment, and both cases showed recurrence of previous hyperparakeratotic or leukokeratotic aspect of the lip. Figure 1 shows the clinical aspect of the lower lip of case 2 (initial, immediate post-treatment, 45 days later).

Histopathological features of lower lip before and after treatment with 5% topical FLU are summarised in Table 1 and Figure 2. Those preliminary results showed that in the prescribed period, no significant differences were seen in the lower lip in both clinical and histopathological characteristics.

Discussion

There is no consensus regarding the best treatment and its long-term results for AC. Therefore, to validate the best treatment, a comparative re-evaluation of the epithelial characteristics after treatment is paramount since there are reports showing the development of SCC even after laser vapourisation of this condition in the lower lip. For this reason, clinical and histological follow-up is mandatory since there is no other way to verify in which lesions epithelial dysplasia remained and those lesions may progress to invasive SCC. Related to actinic keratosis, some authors indicate that the rate of progression to invasive SCC lies between 0.096% and 0.24% per lesion per year, while for AC the percentage of transformation into ACC may be as high as 16.9% if no treatment is implemented.

Topical 5-FLU cream has been shown to be effective in treatment of certain skin cancers. FLU is a fluorinated pyrimidine that blocks the methylation reaction of deoxyuridylic acid to thymidylic acid and in doing so destabilises DNA. Studies
 tors may influence the results of the therapy with FLU like application form, the amount of drug applied, how much time the drug remains in contact with the lip, the mitotic activity present in the epithelium, the presence of epithelial atrophy and the presence of remnant of lip sunscreen or lipstick. Just these factors and the treatment period may explain the permanence of the epithelial alterations and the relapse of the clinical lesion.

Few studies are available regarding the treatment of AC with FLU22,23. Robinson22 treated AC with four different modalities including topical FLU. FLU was applied topically, with the fingertip, as a 5% solution, three times a day for 14 days to the entire vermilion area of the lower lip. The therapy showed recurrence in 50% of the cases. In that study22, five specimens taken from patients treated with FLU showed epithelial dysplasia with markedly diffuse atypia, indicating the permanence of the epithelial morphological alterations, similar to what was observed in this study. Epstein23 reported the treatment of 12 patients with FLU 5% solution and relapse of the lesions was observed in two patients. The author claimed that topical FLU

Table 1 Histopathological features of lower lip in actinic cheilitis before and after treatment with 5% topical fluorouracil

<table>
<thead>
<tr>
<th>Histopathological features</th>
<th>Case 1</th>
<th>Case 2</th>
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<tbody>
<tr>
<td>Irregular epithelial stratification</td>
<td>X</td>
<td>X</td>
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<tr>
<td>Loss of polarity of basal cells</td>
<td>X</td>
<td>X</td>
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<tr>
<td>Drop-shaped rete ridges</td>
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<td></td>
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<tr>
<td>Increased number of mitotic figures</td>
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<tr>
<td>Abnormal superficial mitoses</td>
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<tr>
<td>Dyskeratosis</td>
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<tr>
<td>Keratin pearls within rete pegs</td>
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<td></td>
</tr>
<tr>
<td>Anisonucleosis</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>Nuclear pleomorphism</td>
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<td>X</td>
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<td>Anisocytosis</td>
<td>X</td>
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<tr>
<td>Cellular pleomorphism</td>
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<tr>
<td>Increased nuclear–cytoplasmic ratio</td>
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<tr>
<td>Atypical mitotic figures</td>
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</tr>
<tr>
<td>Increased number and size of nucleoli</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>Hyperchromasia</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>Solar elastosis</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>Epithelial atrophy</td>
<td>X</td>
<td>X</td>
</tr>
</tbody>
</table>

X, condition present.

The treatment standard through careful explanation to the patients, it is important to add that other factors and the treatment period may explain the permanence of the epithelial alterations and the relapse of the clinical lesion.

Figure 2: Case 2 (A, B, C) showing the histopathological features like hyperkeratosis, hyperchromasia and loss of polarity of basal cells (H&E). Case 2 (D, E, F), histopathological features post-treatment with 5% topical fluorouracil with no significant improvement of the epithelial condition.
is an alternative to surgical excision of the vermilion border of the lip; none after treatment biopsy were taken to confirm the resolution of the cases.

Similar to what was found by others, patients treated with FLU for AC report severe discomfort related to the labial ulcerations including difficulty to eat, drink and speak, making the use of analgesics necessary. Although FLU has a low cost, its adverse effects can be superior to the conventional surgical procedures and do not compensate its use, since the epithelial morphological alterations remained. To finalize, it is important to mention that the study had to be stopped due to ethical reasons since the first two cases showed no improvements of the AC with considerable adverse effects.

Conclusion
In the period and dosage recommended in this study, FLU 5% cream did not produce the awaited effects over lesions of AC since it does not improve the clinical and histopathological pre-existent conditions. Nevertheless, it produced severe adverse effects related to the labial ulcerations producing pain and discomfort.

Abbreviations list
FLU, fluorouracil; AC, actinic cheilitis; WHO, World Health Organisation; INCA, Instituto Nacional de Cáncer José Alencar; SCC, squamous cell carcinoma; 5-FLU, 5-fluorouracil.

References