SHORT REPORT

Neoadjuvant intralesional methotrexate in cutaneous squamous cell carcinoma: a comparative cohort study

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Abstract

Background Intralesional methotrexate (MTX–il) has been used as neoadjuvant therapy for keratoacanthoma but has only been utilized in a few isolated cases of cutaneous squamous cell carcinoma as neoadjuvant therapy (cSCC).

Objectives The objective of this study was to evaluate the effectiveness in clinical practice of presurgical MTX–il infiltration to reduce the size of the cSCC. Safety and the impact on subsequent reconstructive surgical techniques was also assessment.

Methods Single, retrospective, observational study of two historical cohorts differentiated in time. Subjects included were diagnosed with infiltrating cSCC. Patients included in group-A received neoadjuvant MTX–il and patients included in group-B underwent scheduled surgery without prior infiltration. Univariate and multivariate analyses were performed.

Results Group-A patients (n = 43) showed an average reduction in the tumour area of 0.52 cm^2, while in group-B (n = 43), the area increased by 0.49 cm^2. A multivariate linear regression analysis demonstrated that MTX–il was the only independent variable that significantly reduced the tumour size [mean 42.6% (95% CI: 31.17–54.03)]. Tumours ≥2 cm in size required significantly a lower percentage of complex reconstructions (P = 0.026). Lower lip tumours showed a higher reduction in group treated with MTX–il (P = 0.045). The only complication observed was discomfort during methotrexate infiltration (60.47%).

Conclusions Neoadjuvant MTX–il reduced the presurgical size of cSCC lesions and could simplify their subsequent surgery.

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Conflicts of interest

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Introduction

Cutaneous squamous cell carcinoma (cSCC) is a significant health problem due to its constant increase in incidence and metastatic potential.[1,2]

The elective treatment for infiltrating cSCC is surgery.[3] Since cSCC is generally located in the facial region, surgery can occasionally cause unacceptable morpho-functional results. In these cases, radiotherapy and systemic or intra-arterial chemotherapy[4] are alternative treatments. Therefore, the use of intralesional agents such as 5-FU[5] or Interferon α-2b[6] has been described.

Intralesional methotrexate (MTX–il) has demonstrated utility in the treatment of keratoacanthomas[7,8], a controversial entity regarded as a benign counterpart of cSCC.[9] However, there are only three described cases of its use in infiltrating cSCC as neoadjuvant therapy.[10–12]

MTX–il neoadjuvant to surgery could significantly reduce tumour size in patients with infiltrating cSCC and thus reduce surgery morbidity and provide better functional and aesthetic results.

The objective of this study was to evaluate the effectiveness of MTX–il in reducing the presurgical size of infiltrating cSCC. Its safety and possible impact on subsequent necessary reconstructive surgical techniques will also be evaluated.

Drs Salido-Vallejo and Cuevas-Asencio contributed equally to this work.

Materials and methods

Single, retrospective, observational study of two historical cohorts of patients with clinical and histological diagnosis of infiltrating cSCC from the Dermatology Department of the Reina Sofía University Hospital (Córdoba, Spain). Group-A included all patients between January 2012 and May 2014 who had received infiltrated MTX–il neoadjuvant to surgery. Group-B contained patients with cSCC removed without neoadjuvant therapy between January 2009 and May 2011.

Inclusion criteria for both groups were patients over 18 years of age, histologically confirmed stage I and II infiltrating cSCC per the American Joint Committee of Cancer (AJCC) criteria[13] and face or extremities localization.

Patients with history of hypersensitivity to methotrexate, haematological abnormalities, liver or kidney disease, locally advanced or metastatic cSCC, keratoacanthomas, pregnancy or breastfeeding, immunosuppressed, xeroderma pigmentosum, albinism or epidermolysis bullosa were excluded.

The pre-filled syringes of methotrexate (25 mg/mL) were administered intralesionally into the base of the tumour depending to its size until this acquired a yellowish colour. A single dose was injected 1 week after the baseline visit to avoid delaying the intervention. The surgery was performed within 30 days of diagnosis.

Off-label use of MTX–il was approved by the Pharmacy and Therapeutics Commission. All patients accepted an informed consent form.

The primary effectiveness endpoint was the reduction of tumour area defined as the difference between the initial area (prior to MTX-il) and final area (pre-surgery). Circular or elliptical morphology was assumed for the lesions for calculating tumour areas using the formula $\pi \times r \times s$ [largest (r) and smallest (s) radius]. The evaluations were conducted independently by two investigators. The surgical reconstruction techniques were defined as simple (primary closure) or complex (local flaps or grafts).

The complications presented by patients arising from the administration of MTX–il were evaluated. Laboratory analyses were carried out before and after the infiltrations to evaluate changes in blood counts and in liver and kidney function.

Statistical analysis was performed using G-Stat 2.0 software (Madrid, Spain), with $P < 0.05$ being statistically significant for all results. For comparisons of continuous data, Student's t-test was used. Continuous data were expressed as means $\pm$ standard deviations. For comparing proportions, chi-squared tests or Fisher's tests were applied. A multivariate linear regression analysis was performed to estimate the effect of MTX–il on tumour size reduction, adjusted for other independent variables (gender, age, initial area of tumour, tumour evolution time, time between clinical diagnosis and surgery, tumour thickness and tumour location).

Results

A total of 86 patients were included (43 per group). No differences were found between the baseline characteristics of both groups (Table 1). The mean quantity of infiltrated methotrexate was 0.74 mL (mininum 0.1 mL; maximum 1.3 mL).

In the group treated with MTX–il, the tumour area was reduced by an average of 0.52 cm^2 as compared to the control group, where tumour area increased 0.49 cm^2. This represented a decrease in the final size of the lesion of 1.01 cm^2 in the group treated with MTX–il (Table 2). A total of 76.74% ($P < 0.0001$) of patients in Group-A showed a reduction in tumour area following infiltration of medication (Fig. 1). In Group-B, 90.7% of tumours stayed the same size or showed an increase in size.

It was not possible to obtain a multiple linear regression model based on the variables studied, as, in the end, it was verified that the only independent variable that significantly affected tumour reduction was methotrexate administration. Therefore, on average, in patients receiving MTX–il, tumour size decreased 42.6% [95% CI: 31.17–54.03] compared to the control group.

Table 1 Baseline characteristics of patients in Groups A and B

	GROUP A (MTX-il + surgery) $N = 43$	GROUP B (surgery) $N = 43$	P
Gender n (%)			
Male	26 (60.47)	24 (55.81)	NS
Female	17 (39.53)	19 (44.19)	
Age (years)			
Mean (SD)	82 (8.65)	81 (9.24)	NS
Time between diagnosis and surgery (days)			
Mean (SD)	17.39 (7.42)	23 (17.17)	NS
Tumour evolution time (months)			
Mean (SD)	5.63 (4.84)	6.69 (6.43)	NS
Initial major diameter (cm)			
Mean (SD)	1.91 (0.72)	1.98 (0.91)	NS
Initial minor diameter (cm)			
Mean (SD)	1.62 (0.61)	1.56 (0.7)	NS
Initial area (cm^2)			
Mean (SD)	2.73 (1.89)	2.85 (2.67)	NS
Location n (%):			
High risk	8 (18.60)	6 (13.95)	NS
Auricle	3 (6.98)	4 (9.3)	
Lower lip	5 (11.63)	2 (4.65)	
Low risk	35 (81.40)	37 (86.05)	
Scalp	8 (18.6)	8 (18.6)	
Temple	6 (13.95)	9 (20.93)	
Cheek	12 (27.91)	7 (16.28)	
Nose	3 (6.98)	2 (4.65)	
Upper limb	5 (11.63)	5 (11.63)	
Lower limb	1 (2.33)	6 (13.95)	

NS, not significant; SD, standard deviation.

Table 2 Comparison of groups after surgery

	GROUP A (MTX-il + surgery) *N* = 43	GROUP B (surgery) *N* = 43	Mean difference (95% CI)	*P*
Modification of tumour area (cm^2)				
Mean (SD)	−0.52 (0.85)	0.49 (0.88)	−1.01 (−1.38, −0.64)	<0.0001
Percentage of variation in tumour area (%)				
Mean (SD)	−23.48 (28.01)	19.12 (25.23)	−42.6 (−54.03, −31.17)	<0.0001
Type of closure *n* (%)				
Overall				
Simple	27 (62.79)	21 (48.84)		0.19
Complex	16 (37.21)	22 (51.16)		
Based on initial greater diameter				
<2 cm (stage T1)*				
Simple	15 (68.18)	18 (64.29)		0.773
Complex	7 (31.82)	10 (35.71)		
≥2 cm (stage T2)*				
Simple	12 (57.14)	3 (20)		0.026
Complex	9 (42.56)	12 (80)		

*Based on the AJCC classification.
95% CI: 95% confidence interval; NS: not significant.

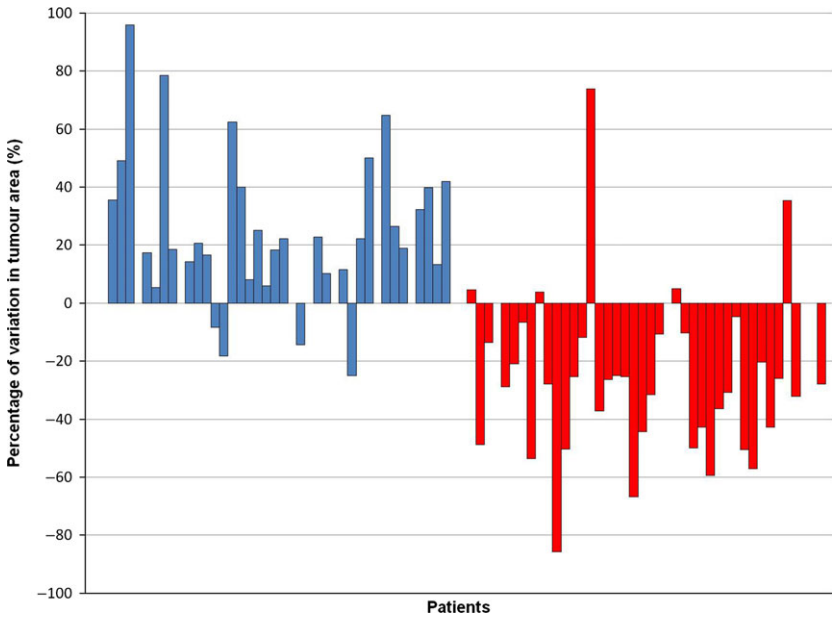

Figure 1 Variation in tumour area in both groups. A decrease in tumour area was observed in Group-A treated with neoadjuvant MTX–il (red) at the time of surgery as compared to its initial area. Group B (blue) showed tumour growth in practically all patients at the time of surgery.

Group-A patients required complex surgical reconstruction in 37.2% of cases, while this was required for 50% of patients in Group-B (Table 2). We found significantly significant differences with regard to the percentage of complex reconstructions in the subgroup of patients with a lesion diameter of ≥2 cm, with it being lower in the group treated with MTX–il ($P = 0.026$).

After a mean follow-up period of 16.03 months, no local relapses or distant metastases were observed in the group treated with MTX–il.

In a *post hoc* analysis of Group-A, a greater reduction in tumours located in the lower lip was observed [48.70%; Interquartile range (IQR):37.17–50.51] compared to the rest of locations [25.21%; IQR:0.00–36.39] ($P = 0.045$). Also in tumours smaller than 2 cm (29.82%) vs. ≥2 cm (13.68%), although was not statistically significant ($P = 0.052$).

No local or systemic complications were related except discomfort during methotrexate infiltration in 60.47% of patients.

Discussion

Methotrexate is a dihydrofolate reductase inhibitor that blocks cell division and results in an antiproliferative effect. Furthermore, MTX increases WWOX gene expression, which promotes caspase activation and apoptosis of cSCC cells.[14] Locally, infiltration achieves higher MTX intracellular concentration in cSCC cells, which are $>10^4$ times more sensitive to the cytotoxic effect of MTX.[15] The success shown by MTX–il in the treatment of keratoacanthomas both as monotherapy[7] and neoadjuvant to surgery[8] has led to various authors using it in some cSCC cases.[10–12,16]

In our study, we observed a significant reduction in tumour area for the cSCCs treated with MTX–il (Fig. 2) being the only independent variable that affected final tumour area in our multivariate analysis. The Group-B cSCCs increased in size an average of 19.12%, which implies that treating cSCCs pending surgery with neoadjuvant MTX–il can provide an average of a 42.6% decrease in tumour area. This final difference in tumour area can be seen in the greater number of complex surgical reconstructions (flaps or grafts) necessary for the control group as compared to the MTX–il group, although this was not statistically significant in our analysis. Only five tumours increased in size, with two cases of increased growth for no apparent cause.

The *post hoc* analysis conducted on the group treated with MTX–il allowed us to characterize the subgroup of patients most likely to benefit from this technique. There was an increased shrinkage in the tumours located on the lip and a statistically tendency in tumours with a diameter <2 cm (T1 according AJCC).[13] This, in addition to the decreased percentage of complex reconstructions required for the subgroup of patients with an initial diameter ≥2 cm in the MTX–il group, suggests that although the tumour reduction could be greater in small-sized tumours, neoadjuvant MTX–il has a greater impact in tumours ≥2 cm.

While MTX–il reduced the size of most of the cSCCs in our study, the degree of response is lower than that obtained in keratoacanthomas.[7,8] Nevertheless, these data support the etiopathogenic connection between both tumours.

Our safety data matched those observed in the literature, with MTX–il showing an excellent safety profile. Contrary to the published articles, more than half of our patients showed mild discomfort during infiltration, however, administration of local anaesthesia or any other intervention were not required. The prior determination of kidney function in patients avoided the

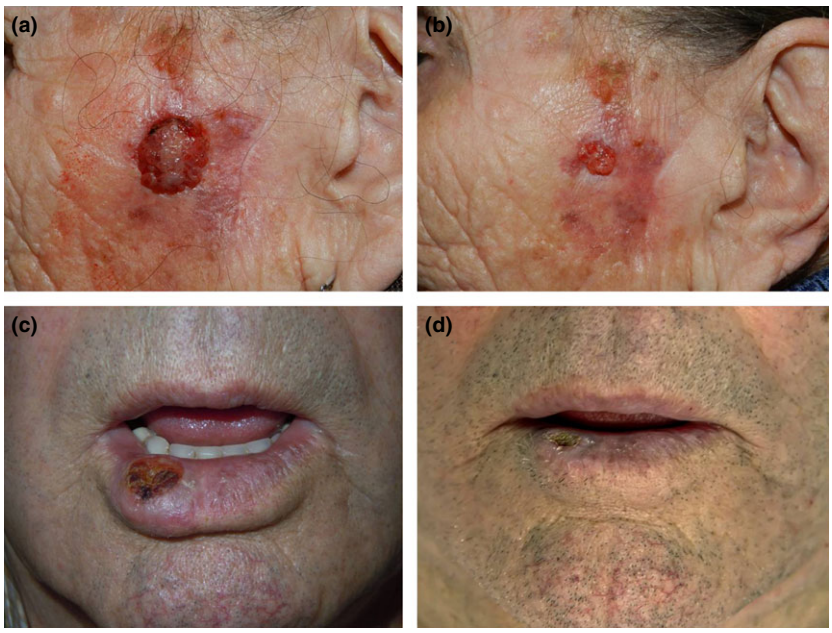

Figure 2 Cutaneous squamous cell carcinoma. Reduction in tumour area in patients with cutaneous squamous cell carcinomas in cheek and lower lip after (a, c) and before (b, d) the intralesional administration of MTX.

possible development of pancytopenia due to the MTX–il, as the cases observed in the literature occurred in patients under dialysis.[7,16]

There are several limitations to our study. This is a single-centre retrospective study with inherent disadvantages. We were not able to find statistically significant differences in some analysis, probably due to our small sample size. In addition, the results of *post hoc* analysis must be interpreted bearing in mind that the study was not developed for this purpose.

In our study, we observed a reduction in presurgical size of cSCC lesions that received neoadjuvant treatment with MTX–il, especially in lesions located on the lower lip. The administration of MTX-il is a simple, safe and inexpensive technique that does not delay the intervention itself. The data suggest a decrease in complex surgical reconstructions that is most evident in tumours ≥2 cm in diameter.

However, its use as monotherapy is not justified due to the risk of development of metastasis, except for palliative purposes in those cases when other currently available treatments have failed or are contraindicated.

The trends shown in our study suggest the possible utility of MTX-il as neoadjuvant therapy to surgery for infiltrating cSCC. Randomized controlled trials are needed to evaluate the efficacy and safety of MTX-il in order to establish an optimal treatment regimen, as well as to define the characteristics of the patients who could benefit the most from its administration.

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