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Topical Oral Cavity Chemoprophylaxis Using Isotretinoin Rinse: A 15-Year Experience

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Objectives/Hypothesis: To determine the utility of isotretinoin oral rinses as a method of chemoprevention for recurrent oral cavity squamous cell carcinoma (SCC), carcinoma in situ, and dysplasia.

Study Design: Retrospective cohort study.

Methods: One hundred forty-three patients were initially enrolled in the study; however, 18 were excluded due to inability to tolerate therapy. The included patients were classified into four groups. Group 1 included patients with multiple early stage oral cavity lesions following initial resection. Group 2 included patients with SCC in situ after excision. Group 3 included patients with multifocal dysplasia following initial CO_2 laser ablation. Group 4 included patients with a history of treated oral cavity SCC with new leukoplakia lesions proven to be dysplastic. Fifty-three patients in the control group did not use post-treatment isotretinoin rinses due to various reasons, whereas 72 patients completed therapy. Minimum use of isotretinoin rinses was 12 months, and minimum follow-up was 24 months. During the follow-up period, all recurrences of carcinoma, in situ disease, and dysplasia were noted and compared with a Fisher test of fit. A Bonferroni correction was applied to increase accuracy and strength of comparison.

Results: Using a Bonferroni correction, the significance threshold was 0.0125. By that cutoff, isotretinoin rinses were found to be associated with lower recurrence in groups 1 and 3 ($P = .00014$, $P = .00002$, respectively) but not in groups 2 and 4 ($P = .4$, $P = .3846$, respectively).

Conclusions: Oral isotretinoin as chemoprophylaxis for patients treated for oral cavity squamous cell carcinoma, in situ disease, or dysplasia may be beneficial in decreasing recurrence rate.

Key Words: Isotretinoin, chemoprophylaxis, oral cavity cancer.

Level of Evidence: 4.

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INTRODUCTION

Over the past century, management of head and neck squamous cell carcinoma (HNSCC) has advanced greatly, from near experimental surgery to evidence-based multimodality therapy. Unfortunately, in that time there have been only minor advancements within the field of chemoprevention and chemoprophylaxis. Slaughter et al. introduced the concept of carcinogen exposure and proximity to invasive cancer as targeting mucosa for eventual malignant progression.[1] This idea of condemned mucosa and field cancerization ignited an early interest in chemoprevention strategies. A lack of meaningful clinical progress and application of chemoprevention can be attributed to the following: 1) clinical trials require patients with a high risk of HNSCC recurrence warranting intervention, 2) chemopreventive agents are often poorly tolerated for prolonged courses, and 3) specific molecular biomarkers have yet to be identified in vitro. Ultimately, an efficacious well-tolerated agent remains yet to be found.[2]

In the last decade there has been a resurgence in chemoprevention due to several landmark studies focusing on the molecular biology of HNSCC. Agents such as estimated glomerular filtration rate (EGFR) inhibitors, β-carotene, and vitamin A analogues have demonstrated promising new frontiers. EGFR is a protein product that is commonly overexpressed in HNSCC cells, prompting immunotherapy trials of erlotinib—an EGFR inhibitor—in patients with high-risk oral premalignant lesions. Although the study did not demonstrate a benefit in cancer-free survival, it did shed light on important points such as dose reduction due to patient intolerance and a common loss of heterozygosity within HNSCC cells.[3] Additional studies centered on the response of premalignant lesions to chemoprevention also demonstrated a lack of long-term cancer-free survival.[4] Although there have yet to be trials with enough power to support the use of chemopreventive agents, they do demonstrate a renewed interest in HNSCC biology as a means for treatment modalities.

Isotretinoin (13-cis-retinoic acid), a vitamin A analogue, was placed into the treatment armamentarium of HNSCC after it was noted that hypovitaminosis could predispose patients to mucosal malignancy. Like

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many chemotherapy agents in the past, isotretinoin was adapted for use in HNSCC from its initial role in the treatment of cystic acne.[5] Multiple preclinical and clinical studies have produced noteworthy results specifically in the HNSCC population. Two of the most convincing studies were conducted by Hong et al. and Kuri et al. Hong et al. hypothesized use of high-dose isotretinoin would help prevent second primaries in patients treated for HNSCC. Isotretinoin was shown to be effective in this role when patients were treated with a course of 50 to 100 mg/m̂2/day for 12 months.[6] In collaboration with MD Anderson Cancer Center, Khuri et al. conducted a randomized control trial with low-dose isotretinoin assessing second primary incidence and survival; patients received 30 mg/d for 3 years. Although this was a large scale randomized control trial, it did not demonstrate statistically significant results.[7] Finally, Perry et al. attempted to recreate the results of the Hong et al. trial with varying doses of isotretinoin. Though multiple options for future research were touted, their study left much to be desired in regard to the future of isotretinoin chemoprevention.[5]

The primary aim of this study was to demonstrate the experience of a single group practice with reconstituted oral isotretinoin rinses for the prevention of oral cavity squamous cell carcinoma (OCSCC), squamous cell carcinoma (SCC) in situ, and dysplasia. This represents the largest study of its kind in the English literature, as not only do other studies examining topical therapy lack significant patient numbers, but other related studies have focused on systemic oral therapy. As irritation is the only side effect, and most insurance companies are willing to cover this drug, this low-risk therapy could prove to be a useful adjunct in the treatment of this patient population.

MATERIALS AND METHODS

Prior to beginning this study, approval was received from the institutional review board (IRB) of JPS Hospital, Fort Worth, Texas.

This was a retrospective study evaluating the utility of using oral chemoprophylaxis to prevent recurrence in patients surgically treated for OCSCC, carcinoma in situ, or dysplasia. Patients treated during the 15-year time period between 1999 and 2014 were included in the study. The minimum follow-up period was 24 months. All patients were treated by the senior author (Y.D.) at a cancer treatment center in Fort Worth, Texas.

Patients treated during this time were categorized into one of four groups based on primary lesions and treatment plan. See below for full description of groups.

Group 1

Group 1 was comprised of patients with multiple early stage (stage 1 or 2) oral cavity cancers with clear margins and no high-risk features following resection. In this group, as in others, multiple lesions was defined as any number of lesions greater than a singular focus. These patients were all treated surgically, and no patients required radiation or chemotherapy as adverse features were not present in the primary specimen or neck dissection specimen if performed.

Group 2

Group 2 was comprised of patients who initially had a focus of early stage (stage 1 or 2) invasive carcinoma and subsequently developed carcinoma in situ following resection of the primary lesion. These patients had no radiation or chemotherapy, as they were early stage and did not have any adverse features of the primary lesion or neck dissection specimen if performed. Patients who developed in situ disease went on to receive an excisional biopsy if <1.5 cm or an incisional biopsy followed by CO_2 laser ablation if >1.5 cm.

Group 3

Group 3 was comprised of patients with multiple foci of mild or moderate dysplasia initially treated with CO_2 laser ablation who then had a recurrence of dysplasia. The recurrent dysplasia was again ablated with a CO_2 laser if >1.5cm or underwent an excisional biopsy if <1.5 cm. Dysplasia was diagnosed based on clinical suspicion followed by histologic confirmation.

Group 4

Group 4 was comprised of patients with any-stage OCSCC initially treated with surgery but then developed leukoplakia, which was found to be dysplasia on biopsy. Patients with advanced disease, stage 3 or 4, were also treated with radiation with or without chemotherapy depending on the findings of surgery. Leukoplakia was biopsied as with other groups in an excisional or incisional fashion depending on size. The entire cohort of patients was recommended to begin oral chemoprophylaxis with reconstituted 0.2% isotretinoin rinses twice daily, for 1 minute duration each, following completion of ablative therapy. Patients who used the medication for a minimum of 12 months were thought to be appropriate for analysis. Patients not beginning therapy due to insurance coverage or lack of desire were used as a control group. Patients beginning therapy who were noncompliant due to odynophagia or intolerable taste or those who aborted therapy prior to the 6-month minimum time period were not included in the study. Patients continuing therapy and the control group patients were followed closely for a minimum of 24 months by the senior author, a surgical oncologist, as well as a medical oncologist.

During this follow-up time period, any recurrence of invasive SCC, carcinoma in situ, or dysplasia was recorded. Data between patients using isotretinoin rinses and those not using the rinses were tracked to analyze difference in recurrence rate. Moreover, recurrences in each group were compiled together and analyzed between both treatment arms to determine if the values were significantly different through the use of a Fisher test of fit and a significance threshold of .05 (α). Given that multiple comparisons were made using the same dataset, a Bonferroni correction was applied to increase accuracy and make the significance criteria more stringent.

RESULTS

A total of 143 patients fitting the inclusion criteria were referred for post-treatment oral isotretinoin use. Fifty-three patients were unable to begin treatment due to a variety of reasons centered mainly on socioeconomic reasons, and thus this treatment arm was assumed to be a control for the study. Of the 53 patients who did not pursue isotretinoin treatment, 38 were male and 15 female, with an average age of 48.4 years.

In group 1 patients with multiple early stage OCSCC lesions recurring after prior surgical treatment,

TABLE I.
Findings From Patients in Group 1 for Fisher Test

	Group 1	
	No isotretinoin	Isotretinoin
Recurrence	8	2
No recurrence	0	12
Statistical significance	$P = .000141$ (significant)	

TABLE III.
Findings From Patients in Group 3 for Fisher Test

	Group 3	
	No isotretinoin	Isotretinoin
Recurrence	22	6
No recurrence	9	36
Statistical significance	$P = .000002$ (significant)	

there were eight patients who had not begun oral isotretinoin therapy. Six patients developed new SCC of whom one died of pulmonary embolism in the postoperative period and two died of distant metastasis at 8 and 36 months postoperatively. One patient developed two separate locoregional recurrences that were surgically salvaged, and one patient developed SCC in situ (Table I).

In group 2 patients with SCC in situ following excision, there were four total patients who did not begin oral isotretinoin use.One patient went on to develop recurrent dysplasia needing CO_2 laser ablation at 3 years following initial treatment (Table II).

In group 3 patients with multiple sites of dysplasia recurring after CO_2 laser ablation, there were a total of 31 patients who did not take oral isotretinoin. Sixteen had repeat CO_2 laser ablation at 17 months following initial treatment. Two developed SCC in situ and four developed SCC, two of whom died of locoregional disease and one of distant metastasis at 4 years post-treatment (Table III).

In group 4 patients with leukoplakia found to be dysplasia following resection of any oral cavity SCC, 10 patients were in the control. One developed SCC but died of locoregional recurrence at 9 months following surgical treatment. Three patients developed dysplasia requiring repeat CO_2 laser ablation at an average of 14 months after initial resection (Table IV).

Ninety patients were placed on oral isotretinoin, with 72 going on to complete the minimum therapy. Eighteen patients, 12 male and 6 female, with an average age of 51 years, were excluded from the study as they either could not tolerate the medication or were noncompliant with use.

Seventy-two patients continued isotretinoin use for the minimum 12 months period. In this cohort, there were 48 male and 24 female patients, with an average age of 51 years. In group 1, 14 total patients completed therapy with isotretinoin. One patient developed SCC in situ 9 months after stopping treatment, and another patient developed two new oral cavity early-stage SCC lesions at 8 months and 3 years following discontinued use of isotretinoin.

In group 2, six total patients met the minimum use for isotretinoin rinses, and there were no patients with recurrence of SCC, SCC in situ, or dysplasia.

Forty-two patients in group 3 used rinses post-treatment, with one patient developing SCC in situ during the ninth month of treatment. Four patients had dysplasia during the treatment course that required repeat CO_2 laser ablation. One patient developed SCC 2.5 years after treatment, but died from an unrelated event 6 months following the second surgery.

Finally, in group 4, 10 patients completed used of oral isotretinoin. One patient developed dysplasia at 5 months following treatment and required CO_2 laser ablation. One patient developed SCC at the seventh month of treatment and died from distant metastases.

To analyze if oral isotretinoin use impacted post-treatment recurrence of SCC, SCC in situ, or dysplasia, patients in each treatment arm were classified as having had a recurrence of any pathology or having had no recurrence. These values were compared between patients treated with oral isotretinoin and those who were not treated with the agent. A comparison using a Fisher test of exact fit was conducted among each of the four groups to determine statistically significant differences, with an α value set at .05. Given that four tests were performed with the dataset, the Bonferroni correction was calculated to be 0.0125.

Using the significance threshold of 0.0125, isotretinoin use was associated with lower recurrence in groups 1 and 3 ($P = .000141$ and $P = .000002$, respectively) but not in groups 2 and 4 ($P = .4$ and $P = .3846$, respectively).

After individual group analyses were performed, all patients were analyzed together by combining the subjects from all four groups. Yielding a P value of $< .01$, the

TABLE II.
Findings From Patients in Group 2 for Fisher Test

	Group 2	
	No isotretinoin	Isotretinoin
Recurrence	1	0
No recurrence	3	6
Statistical significance	$P = .5$ (not significant)	

TABLE IV.
Findings From Patients in Group 4 for Fisher Test

	Group 4	
	No isotretinoin	Isotretinoin
Recurrence	4	2
No recurrence	6	8
Statistical significance	$P = .3846$ (not significant)	

difference between recurrence among patients using isotretinoin rinses compared to the control group were statistically significant.

DISCUSSION

Although the use of isotretinoin for oral chemoprophylaxis has been disputed, there have been studies in the past decade suggesting a possible role of various agents as tumor biology has been further elucidated. This study represents the largest single-center experience with topical oral isotretinoin use for the prevention of OCSCC, carcinoma in situ, and dysplasia. Most studies previously have discussed systemic isotretinoin use but have not focused on topical delivery. Although numerous factors exist that could influence outcome, and by no means are the data reported in this article meant to provide generalized recommendations, the experience posed in this study simply suggests a role for this agent and encourages further investigation.

The findings of this study suggest that the use of oral isotretinoin rinses following primary treatment may be useful in preventing recurrence of early-stage SCC, dysplasia, and carcinoma in situ. Regardless of initial pathology, following treatment with surgery or laser ablation and post-treatment use of oral isotretinoin rinses, patients with multiple oral cavity early-stage cancerous foci and multifocal dysplasia appeared to have decreased recurrence following treatment ($P = .000141$ and $P = .000002$, respectively), whereas patients with SCC in situ after excision and patients with dysplasia following treatment of oral cavity SCC (any stage) did not have the same response. It is uncertain why patients from groups 1 and 3 had a significant benefit, whereas those from groups 2 and 4 did not. Because groups 1 and 3 had multiple abnormal lesions, indicating diffuse field cancerization, these findings suggest a potential benefit of topical isotretinoin in patients with multiple lesions when compared to patients with a single isolated lesion who may have less severe of a field cancerization effect.

Isotretinoin and other vitamin A derivatives have been shown to have a chemotherapeutic effect, as evidenced by the use of high-dose vitamin A compounds in induction chemotherapy for malignancy. Retinoids have been used in the treatment of lymphoma due to their effect on apoptosis and cell proliferation by modulating gene expression.[8] It is possible that oral rinses of this compound allow direct contact with oral cavity mucosa and have a toxic effect on malignant cells or dysplastic cells with malignant potential. In this study, patients began using the medication immediately following completion of therapy, suggesting an early exposure to raw surfaces inside the oral cavity, which may have increased absorption. As field cancerization is an often-discussed concept in head and neck malignancy, field treatment with therapeutic agents, similar to topical cutaneous treatments, may provide benefit.

The use of isotretinoin has been debated, and routine use has not been established given the dearth of significant findings. The majority of studies have focused on systemic retinoid therapy to assess for recurrence of oral cavity lesions. A study done by Perry et al. examining 151 patients treated for primary malignancies of the head and neck were stratified into three treatment categories, one taking a high-dose of oral isotretinoin, one group taking a moderate dose, and one a placebo group. At the end of 3 years of treatment, the investigators concluded that no significance difference was found in prevention of recurrence ($P = .7$), second primary malignancies ($P = .9$), or disease-free survival ($P = .8$).[5] However, it is important to note that these patients were taking isotretinoin orally, which arguably may have increased systemic delivery, but possibly may not have increased direct exposure to affected mucosa, which makes direct comparison with our study difficult.

On the contrary though, a previous study by Shah et al. examined 16 patients with oral leukoplakia treated with varying doses of topical retinoids as 13-cis-retinoic acid. They reported 11 patients with positive response to treatment, with only two showing recurrence, which was not significant for cytologic regression. One patient who had an initially questionable response during retinoid treatment went on to be free of recurrence following cessation of treatment. Their study is unique in that they experimented specifically with topical retinoids. Although their study utilized a lozenge delivery system, which could have allowed for a longer and more objective way of measuring topical delivery, their findings were encouraging and suggestive of a possible role for retinoid treatment similar to the results posited in the current study.[9]

It is evident that the literature is sparse on the subject of oral cavity chemoprevention, and even more so on topical chemoprevention for OCSCC and precursor lesions. Although the data are conflicting, and studies with significant power are limited, it is apparent that there may be a role for a preventive chemotherapeutic vehicle. Isotretinoin rinses, as reported in our study, could represent a well-tolerated and low-risk modality of chemoprevention. The current study is the only study to date reporting a robust experience with topical isotretinoin rinses. Although our study has promising results, they represent the practice of a single institution, and as such cannot be used as recommendations to other practitioners. It would be intriguing to see if a similar study, perhaps on a multi-institutional scale, would be able to reciprocate the results and make a stronger conclusion. In the future, patients could be stratified based on alcohol and tobacco use as well to determine if this was an independent factor influencing the therapeutic potential of isotretinoin. As human papillomavirus (HPV) has become of greater importance in certain head and neck cancers, future trials correlating HPV status to recurrence results could also prove to be useful.

CONCLUSION

Based on the 15-year experience of a single institution, reconstituted topical isotretinoin rinses may be beneficial in the chemoprevention of recurrent OCSCC and similar high-risk premalignant mucosal lesions in select patients by targeting field cancerization; however, further studies are needed to make generalized recommendations.

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