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Research Article

Cancer Prevention Research

Autofluorescence Imaging to Monitor the Progression of Oral Potentially Malignant Disorders

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Abstract

Patients with oral potentially malignant disorders (OPMD) must undergo regular clinical surveillance to ensure that any progression to malignancy is detected promptly. Autofluorescence imaging (AFI) is an optical modality that can assist clinicians in detecting early cancers and high-grade dysplasia. Patients with OPMD undergoing surveillance for the development of oral cancer were examined using AFI at successive clinic visits. Autofluorescence images acquired at 133 clinical visits from sites in 15 patients who met inclusion criteria were analyzed quantitatively using an algorithm to calculate the red-to-green pixel intensity (RG ratio). A quantitative AFI threshold for high risk of progression was defined based on the RG ratio and was compared with expert clinical impression and with histopathology when available. Patients were divided into two groups based on their endpoint: surveillance ($n = 6$) or surgery ($n = 9$). In the surveillance group, 0 of 6 (0%) of patients were clinically identified as high risk for progression prior to the study endpoint, whereas 1 of 6 (17%) of patients were deemed at high risk for progression based on AFI during the same time period. In the surgery group, 9 of 9 (100%) of patients were clinically identified as high risk prior to the study endpoint, whereas 8 of 9 (89%) of patients were at high risk for progression based on AFI during the same time period. AFI results tracked over time were comparable with expert clinical impression in these patient groups. AFI has the potential to aid clinicians in noninvasively monitoring oral precancer and evaluating OPMDs that require increased surveillance.

Introduction

The global prevalence of oral potentially malignant disorders (OPMD) is estimated to be 1.5% to 2.5% (1); the management of these patients is a challenge for clinicians. OPMD include a heterogeneous group of morphologically altered oral mucosal lesions, most commonly leukoplakia, with a variable risk of malignant transformation. Follow-up studies have shown that between <1% and 18% of patients with OPMD will develop oral cancer (2). Although certain clinical subtypes of leukoplakia appear to be at a higher risk for malignant transformation than others, currently the presence and degree of epithelial dysplasia are the most important predictors of malignant development (3, 4). However, the assessment of oral epithelial dysplasia grade requires invasive biopsy and is highly subjective, making its predictive value for malignant transformation far from optimal (5). Additionally, not all dysplastic lesions will eventually become malignant, and some may even regress (6). Hence, there exists no consensus on guidelines for active surveillance of patients with these preinvasive lesions (7). There is a need for improved systems to classify malignant potential and a reliable method for follow-up surveillance of patients with OPMD (8).

Management strategies for patients with OPMD are aimed at early detection of malignant transformation or prevention of progression, but there is no consensus on guidelines for active surveillance, so practicing clinicians use different protocols to follow-up their patients (9, 10). The most common approach to management of patients

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with OPMD is surveillance over time with careful conventional oral examination followed by invasive biopsy of suspicious lesions. The subjective nature of clinical examination and potential for sampling error with small biopsy samples may limit the ability of this approach to discern the conversion of OPMD to high-grade dysplasia and cancer (11, 12). Factors such as the severity of dysplasia grade, anatomic site of the lesion, and ongoing exposure to risk factors such as smoking and alcohol may influence the time intervals between biopsies and clinical follow-up. Though some experts recommend surgical excision of OPMD lesions with moderate dysplasia, there is no conclusive evidence that this approach improves survival. Further, complete excision is not feasible without significant morbidity for OPMD with multifocal or diffuse involvement. Thus, a significant proportion of patients with OPMD need more conservative management. At present, limitations in the ability to predict which lesions will undergo malignant transformation inhibits our ability to focus medical resources toward those at highest risk for cancer development.

Optical imaging technologies have the potential to improve the clinical surveillance of patients with widespread OPMDs (13). Widefield autofluorescence imaging (AFI) can be used to rapidly scan large areas of the oral mucosa and identify regions at high risk of neoplasia (14). In the stroma, the progression from precancer to cancer leads to a degradation of the collagen matrix, which results in a loss of blue–green autofluorescence (AF) that is visible as dark patches (15). This decrease in AF intensity can be viewed directly or documented by photography. Although AFI is highly sensitive for detection of dysplasia and cancer, its specificity is limited because benign conditions such as inflammation are also associated with reduced AF (15, 16).

Previous and ongoing follow-up studies have indicated a role for AFI to improve clinical surveillance of patients with OPMDs (12, 17, 18). Commercial AFI instruments such as the VELscope (LED Dental) have become available over the past decade. Poh and colleagues have used the VELscope to identify AF loss in patients presenting with oral squamous cell carcinoma at the time of surgery (19) and have also found that using AFI to guide surgical resection margins can reduce the recurrence rate (17). Rock and colleagues longitudinally followed patients with low-grade dysplasia, and found that subjective fluorescence loss was associated with eventual progression to severe dysplasia or worse (20). However, to the best of our knowledge, all studies that have monitored progression with AF have only used subjective tissue appearance to characterize fluorescence loss.

We previously reported on the development of AFI instruments and automated image analysis algorithms for detection of oral neoplasia (14, 21, 22). We evaluated the performance of AFI alone and in combination with other imaging modalities on a single time-point basis (12, 18, 21, 22). Here, we report the use of AFI with objective, automated image analysis in long-term longitudinal surveillance of patients with OPMD.

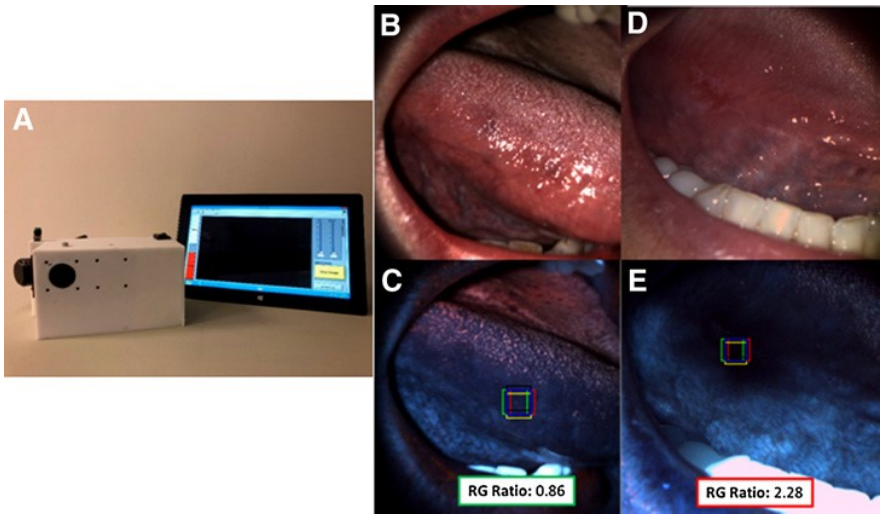

Figure 1.

White light and AFI system with examples of images from two study patients. **A,** Imaging system used to acquire images. **B,** White light image of a right lateral tongue rated clinically normal by the expert clinician. **C,** Corresponding AFI image of right lateral tongue. Blue box represents sites where Red-to-Green pixel intensity (RG Ratio) was originally selected and calculated. RG ratios for sites 10 pixels above (black box), 10 pixels below (yellow box), 10 pixels left (green box), and 10 pixels right (red box) were averaged with the original selected site. There is no loss of AF resulting in a low score of 0.86. **D,** White light image of a right lateral tongue rated clinically abnormal high risk by the expert clinician. **E,** Corresponding AFI image of right lateral tongue. Loss of AF is visually evident at the tracked site and has a corresponding increased RG ratio score of 2.28.

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Methods and Materials

Instrumentation

The widefield AFI system (Fig. 1A) is a compact version of the multispectral digital microscope system, described previously by Roblyer and colleagues (14, 21). Briefly, this system has a 45-mm diameter field of view with a 250-mm working distance and spatial resolution of 0.1 mm. AF illumination is provided by a blue LED, and white light illumination is provided by a white LED. Image acquisition and display are controlled by a tablet computer. With the room lights dimmed to minimize ambient light, the user can switch between white light mode and AF mode. Pressing the acquisition button initiates automatic collection of a series of white light and AFI images, which takes approximately 2 seconds.

AFI procedure and analysis

AF and white light images of the oral mucosa were acquired with the imaging system described in the previous section. A typical complete imaging session in which 2 to 4 oral sites were examined and imaged took approximately 2 minutes. The AF of oral sites was objectively quantified using the average ratio of the red-to-green pixel intensity (RG ratio). At each site, the RG ratio was calculated from a 65 × 65 pixel area (~4 mm × ~4 mm), which roughly corresponds to the size of a punch biopsy. To account for variability in hand selection of sites, the RG ratio was computed for four additional regions offset by 10 pixels (~0.6 mm) above, below, to the left and to the right of the original site. The average of these 5 values was reported as the RG ratio value for the site. Figure 1B–E shows examples of white light and AF images of two study patients. Figure 1B and C represents a white light and AF image for a clinically normal site. The white light image shows no visible lesions, and there is no loss of AF in this image resulting in a low RG ratio of 0.86. Figure 1D and E demonstrate an example of a site with an OPMD identified as abnormal high risk by an expert clinician. In Fig. 1E, a dark patch is visually evident, indicating loss of AF; the site has an increased RG ratio of 2.44.

Study population

Patients with OPMD in the Head and Neck clinic at the University of Texas MD Anderson Cancer Center (MDACC) were recruited into this study. Patients were eligible to participate if they presented with a premalignant oral lesion, had a history of head and neck cancer or oral premalignant disease but without any clinical evidence of disease, or presented with any condition (such as lichen planus, heavy tobacco use, etc.) that increased their risk for oral cancer development. The study was performed in accordance with IRB-approved protocols at MDACC and Rice University in Houston, TX. Written informed consent was obtained from all subjects.

Longitudinal study protocol

Over a period of 6 years and 3 months, patients were recruited and examined at successive surveillance visits to the Head and Neck clinic and (if applicable) in the operating room immediately prior to surgery for OPMD. At each visit, the clinician (A.M. Gillenwater) first performed a clinical examination of the patient's oral cavity and identified oral sites of interest. Oral sites were selected based on the clinician's judgement and were followed based on the clinician's level of concern; new oral sites were added during patient follow-up if the clinician determined that

Variables to evaluate tissue sites			
Risk level	**Clinical impression (CI)**	**Quantitative imaging (AFI)**	**Pathology (Path)**
4 🔴	Cancer	RG Ratio > 1.53	Cancer
3 🟠	Abnormal high risk	1.34 < RG Ratio ≤ 1.53	Moderate/Severe dysplasia
2 🟡	Abnormal low risk	1.05 < RG Ratio ≤ 1.34	Mild dysplasia
1 🟢	Normal	RG Ratio ≤ 1.05	Normal
Other timeline elements			
⊕	Suspicion flag for CI (First abnormal high risk or cancer)		
📷	Suspicion flag for imaging (Second AFI risk level 3 or first AFI risk level 4)		
⋮	Enrolled on study		
|	Surgery		

Figure 2.

Three variables were used to evaluate tissue sites possible or actual malignant progression. CI was the expert clinician's evaluation (normal, abnormal low risk, abnormal high risk, or cancer) of the site. Quantitative imaging was defined as the RG ratio score of a tracked site in an AFI. Pathology (Path) was the histologic diagnosis (normal, mild dysplasia, moderate/severe dysplasia, or cancer) from a biopsy or surgical specimen of the site. Results for each variable were stratified into four levels. Each group's four levels were assigned a color code (green, yellow, orange, or red) to evaluate suspicion in patient timeline plots. Suspicion flags were designated for CI and AFI to identify when each variable signaled potential for disease progression. Blue dashed lines indicate when a patient was enrolled on study, and red solid lines indicate when a surgery was performed at the tracked site.

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a new site should be tracked. The clinical impression (CI) of each oral site was categorized by the physician as (1) normal, (2) abnormal–low risk for dysplasia, (3) abnormal–high risk for dysplasia, or (4) cancer. Next, white light and AF images that included each oral site were collected using the AFI system described in the previous section. If judged necessary by the clinician as part of standard of care, biopsies were taken of the oral sites or the patient was scheduled for surgery. Tissue specimens were processed and diagnosed in accordance with standard criteria, and pathologic results were classified into the following categories: (1) normal, (2) mild dysplasia, (3) moderate/severe dysplasia, or (4) cancer.

Longitudinal study analysis

A retrospective analysis was performed on a subgroup of the study population to compare the ability of AFI and CI to predict whether a site under surveillance would eventually require surgery due to disease progression. Analysis was restricted to sites with imaging data from multiple visits, and for which a clear diagnostic outcome (i.e., an endpoint) was available. Specific eligibility criteria for this analysis included: (i) site was in a patient that was imaged with AFI for at least six visits; (ii) the site itself was imaged at least four times; (iii) surgery was not performed on the site within 50 days of initial patient enrollment; (iva) at least one pathology result (from a biopsy or surgical specimen) from the oral site, or (ivb) site was under surveillance for at least 4 years with no pathologic evidence of high-grade (moderate/severe) dysplasia or cancer.

Patients meeting the eligibility criteria for the analysis were divided into two groups based on their endpoint: (i) surgery or (ii) continued surveillance. A surgery endpoint was declared when a surgery was performed on the oral site; this was considered a disease-positive outcome for the purposes of analysis. In the absence of surgery, a continued surveillance endpoint was declared at the last time a patient's oral site was observed in the clinic, with no clinical or pathologic evidence of high-grade disease or cancer; this was considered a disease-negative outcome for the purposes of analysis.

For each patient, a timeline was created to represent changes in (i) CI, (ii) histologic diagnosis, and (iii) quantitative imaging. Each variable was stratified into four levels, with a color code associated for each level as shown in Fig. 2. The CI variable was stratified into four levels: (1) normal, (2) abnormal low risk, (3) abnormal high risk, or

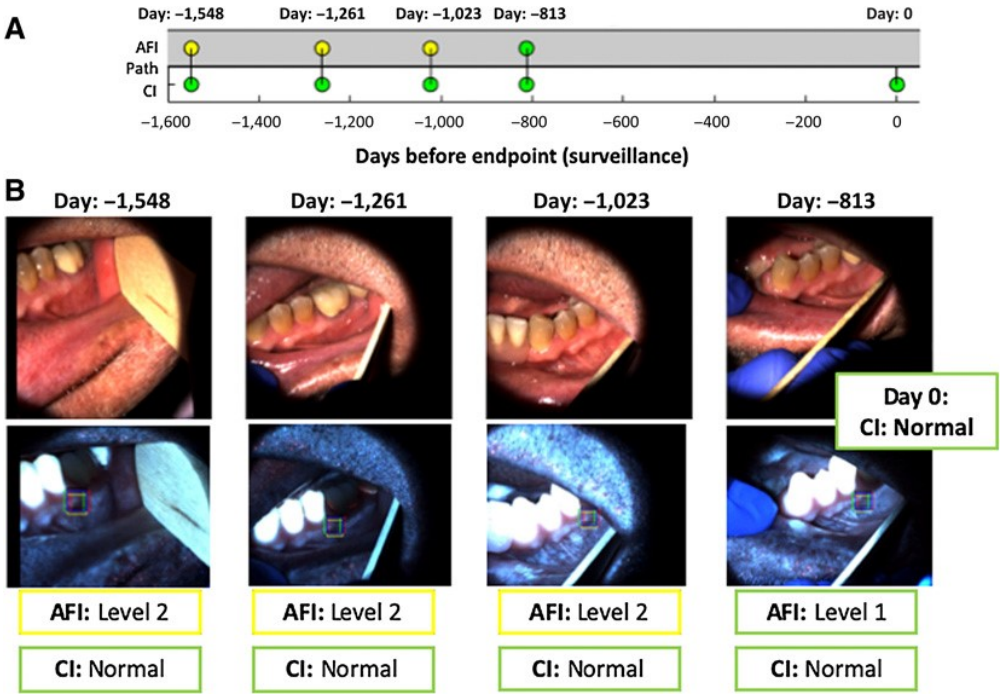

Figure 3.

Timeline and corresponding images of patient who had no biopsies or surgeries during the observation period. **A,** Timeline for the site. Reference Fig. 2 for legend. −1,548 days before the end of this patient's observation period, the patient had an AFI level 2 score and normal CI score. These scores remained the same for days −1,261 and −1,023. On day −813, AFI decreased to level 1. This patient had a normal CI score at the end of the observation period (day 0). This patient had no AFI or CI suspicion flags signaled. **B,** Site's corresponding white light (top row) and AF (bottom row) images for each day in timeline. The square box in each image represents the site evaluation by AFI and CI.

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(4) cancer. Histologic diagnosis was also stratified into four levels: (1) normal, (2) mild dysplasia, (3) moderate/severe dysplasia, or (4) cancer. The quantitative imaging result was stratified into four levels depending on the RG ratio score of the site. The threshold values used to define these levels were found using a previously described data set correlating RG ratio to histologic diagnosis (18, 21) as detailed in the Supplementary Data and displayed in Supplementary Fig. S1. The date of the final observation endpoint (surgery or final surveillance visit) was defined as day 0 in the analysis timeline, with the clinical observation period during which AFI took place indicated by negative day numbers leading up to day 0. Suspicion flags for high risk of disease progression were defined for both the quantitative imaging and CI variables to signal the day of the first indication of a potentially suspicious site. The suspicion flag for AFI was defined as the first occurrence of an AFI level 4 imaging result or the second occurrence of an AFI level 3 result, for a given site. The suspicion flag for CI was defined as the first occurrence of an abnormal high risk or cancer clinical impression from the expert clinician for a given site.

A modified Kaplan–Meier analysis was performed to compare the time interval between when AFI and CI identified that a site was at high risk for progression prior to the study endpoint. In the analysis, the percentage of enrolled patients signaled with a suspicion flag before the final observation endpoint is plotted versus the number of days before the patient's final observation endpoint. Analysis was performed separately for patients with a final observation endpoint of surgery and with a final observation endpoint of continued surveillance.

Results

Analyzed sites

Two hundred sixteen patients were enrolled and imaged at least one clinic visit. Of those, 45 patients were imaged with AFI at six or more visits. Of those 45 patients, 15 oral sites from 15 patients (133 cumulative total clinic visits) met the remaining eligibility criteria and were included in the longitudinal analysis (Supplementary Fig. S2). Nine patients had a surgery endpoint, and 6 patients had a continued surveillance endpoint.

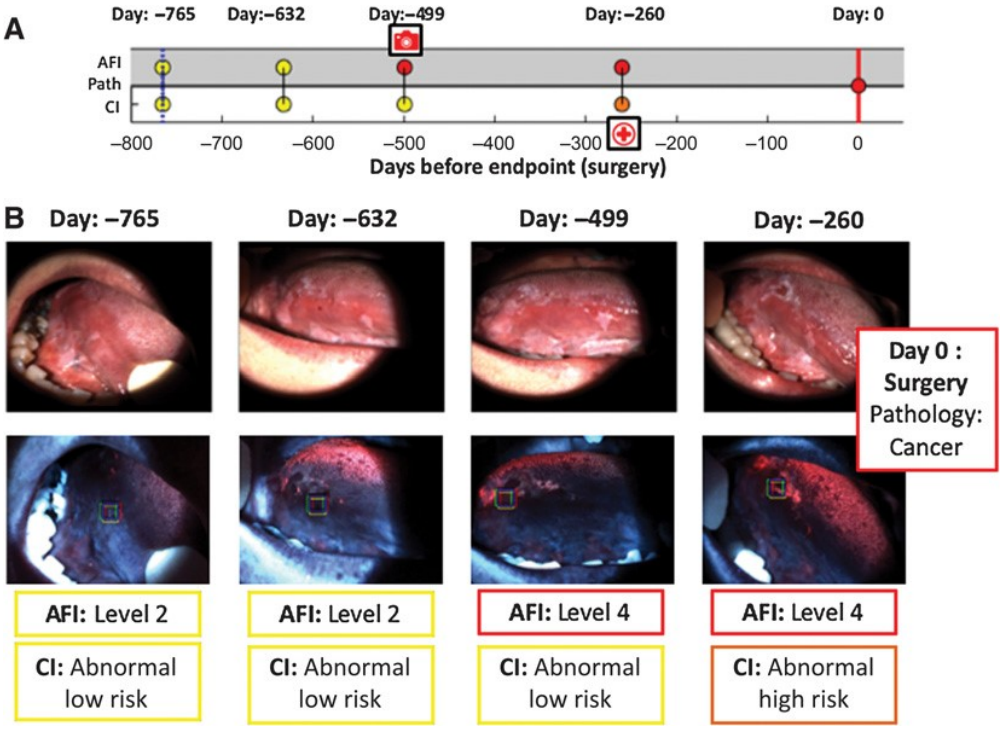

Figure 4.

Timeline and corresponding images of patient who had a surgery during observation period. **A,** Reference Fig. 2 for color classifications used in timeline. 765 days before the end of this patient's observation period, the patient had an AFI level 2 score and abnormal low-risk CI score. At day −632, the patient's AFI score remained at level 2 while CI remained at abnormal low risk. At day −499, the imaging score advanced to level 4 signaling the AFI suspicion flag while CI remained the same. The imaging score remained the same at day −260, whereas CI advanced to abnormal high risk, signaling the CI suspicion flag. There was a surgery 260 days later at this site with a pathological diagnosis of cancer. **B,** Corresponding white light (top row) and AF (bottom row) images for each day in timeline. The square box in each image represents the site evaluation by AFI and CI.

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Representative patient sites

An example of data acquired from a surveillance patient is shown in Fig. 3. The patient's timeline is shown in Fig. 3A. The white light and AF images are shown in Fig. 3B. For this patient, on day −1,548 the CI was normal, and the quantitative imaging result was AFI level 2. At the next two visits (day −1,261 and day −1,023), the CI remained normal and the quantitative imaging result remained AFI level 2. The quantitative imaging result was reduced to AFI level 1 on day −813 while CI remained normal. On day 0, the last observation day for this patient, no imaging was performed but CI remained normal. Thus, for this patient's oral site, neither a CI suspicion flag nor an AFI suspicion flag was triggered over 1,548 days of observation. In the current analysis, both CI and AFI were considered to have correctly predicted the disease-negative continued surveillance endpoint for this site.

An example of data acquired from a patient with a surgery endpoint is shown in Fig. 4. The patient's timeline is shown in Fig. 4A. The white light and AF images are shown in Fig. 4B. For this patient, on day −765, the CI was abnormal low risk and the quantitative imaging result was AFI level 2. On day −632, the CI and AFI remained the same. On day −499, the CI remained abnormal low risk but the quantitative imaging result increased to AFI level 4, triggering an AFI suspicion flag, indicated by the red camera icon on the timeline. On day −260, the imaging result remained at AFI level 4 while CI increased to abnormal high risk, triggering the CI suspicion flag indicated by the red cross icon on the timeline. On day 0, the patient underwent surgery. Histopathology results from the surgical specimen confirmed the presence of cancer at the imaged site. For this patient, an AFI suspicion flag was signaled 499 days prior to surgery, whereas a CI suspicion flag was signaled 260 days prior to surgery. In the current analysis, both CI and AFI were considered to have correctly predicted the disease-positive surgical endpoint for this site, with imaging providing an earlier indication.

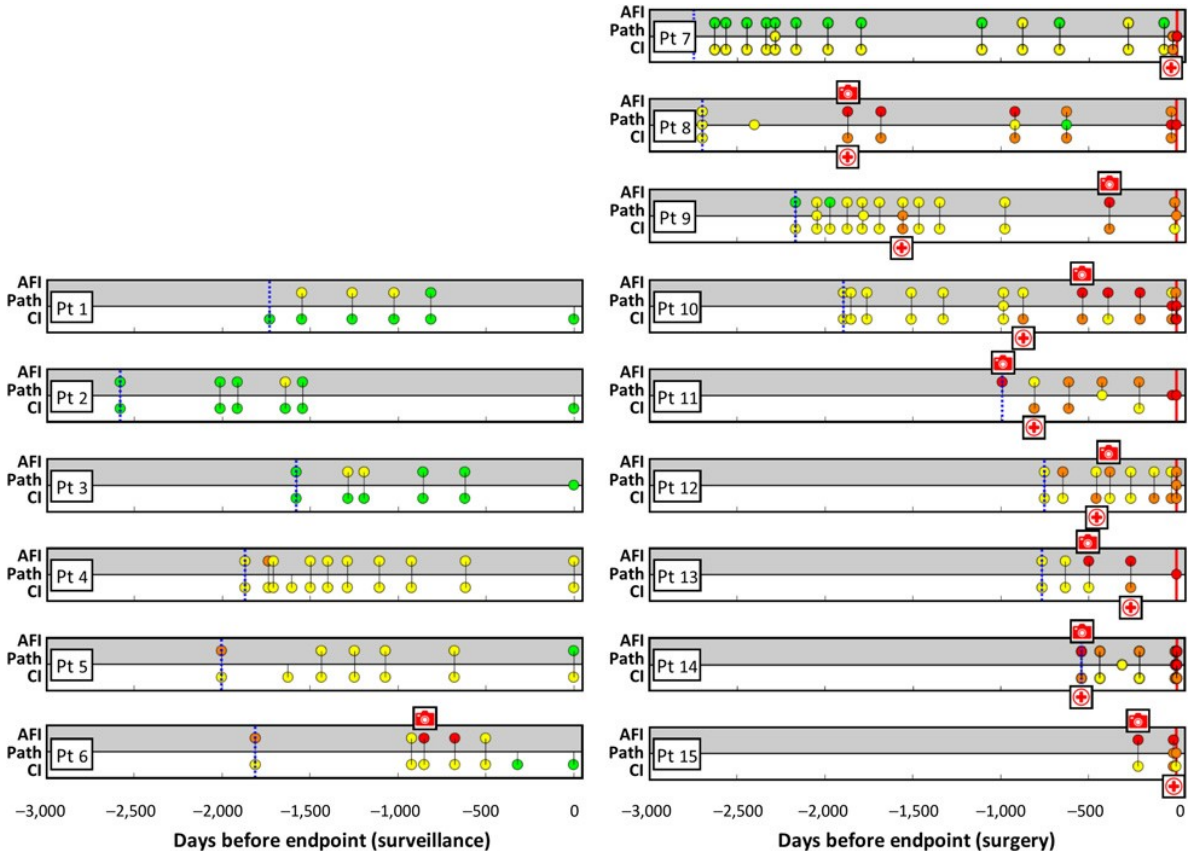

Figure 5.

Timelines for the six continued surveillance patients (left) and for the nine surgery patients (right). Each timeline tracks one patient's site and compares the quantitative imaging (AFI), pathology (Path), and CI variables. Refer to Fig. 2 for timeline element and color information. Note that for the surgery patient group, data are only shown up to the surgery endpoint and follow-up visits are not shown in the timelines.

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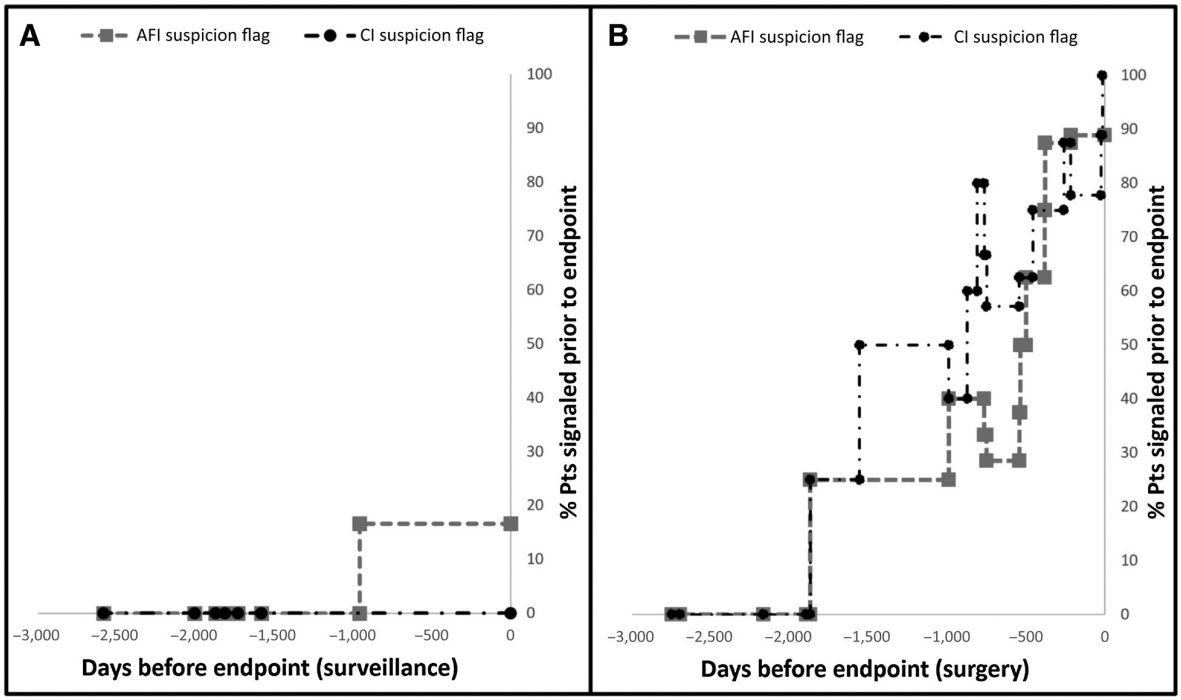

Figure 6.

Modified Kaplan–Meier analysis of AFI and CI results, showing the percentage of patients over time with an AFI suspicion flag or a CI suspicion flag (as defined in Fig. 2) raised prior to the study endpoint. **A,** Surveillance group ($n = 6$). In this group 0/6 (0%) of patients had a CI suspicion flag raised, whereas 1 of 6 (17%) of patients had an AFI suspicion flag raised prior to the endpoint. **B,** Surgery group ($n = 9$). In this group, 9 of 9 (100%) of patients had a CI suspicion flag raised, whereas 8 of 9 (89%) of patients had an AFI suspicion flag raised prior to the endpoint. Note that the running percentage value may either increase (when a new suspicion flag is raised) or decrease (when a new patient enrollment occurs without a suspicion flag).

Performance of imaging

Figure 5 displays the timelines for all patients included in the analysis. Patients #1 to #6 had a continued surveillance endpoint and did not have surgery at the oral site. None of the 6 patients with a surveillance endpoint had CI suspicion flags signaled; only one patient, patient #6, had an AFI suspicion flag signaled. Patients #7 to #15 had a surgery endpoint with surgery occurring on the date defined as day 0. Eight of 9 patients with a surgery endpoint had AFI and CI suspicion flags signaled. One patient, patient #7, had a CI suspicion flag signaled 24 days before surgery but no AFI suspicion flags. Imaging was not performed on this day due to unavailability of instrumentation.

Figure 6 shows the results of the modified Kaplan–Meier analysis, calculated based on the timelines shown in Fig. 5. For the continued surveillance patient group, no CI flags were signaled (0 flags signaled/6 patients enrolled) at any point of the observation period; therefore, the curve remained at zero; there was one AFI flag signaled for the continued surveillance patient group at day −958 causing the curve to raise to 17% of patients signaled (1 patient signaled/6 patients enrolled). For the group with a surgery endpoint, 25% of patients enrolled on day −1,870 had signaled (patient 8 had both the AFI and CI flag signaled at the same clinic visit). Half of the patients with a surgical endpoint signaled by CI at day −1,557 (2 patients signaled/4 patients enrolled at day −1,557), whereas half of the patients signaled by AFI (4 patients signaled/8 patients enrolled at day −535).

Discussion

Patients with OPMD pose several clinical challenges. There is currently no precise method to accurately predict an individual's risk for oral cancer. Better tools are needed to monitor high-risk and post-surgery patients for development or recurrence of malignant tumors. Improved methods are required to evaluate early-stage clinically evident lesions, especially those with mild dysplasia for which histopathology is a weak prognosticator of subsequent malignant transformation (23).

There are limited data to assess the ability of optical imaging to aid in noninvasively monitoring OPMD patients for disease progression. This analysis demonstrates the ability of a simple, noninvasive, rapid imaging technique to help clinicians objectively identify which OPMD lesions require closest surveillance and are likely

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to eventually require surgery. In the surgery endpoint group, 50% of the patients had an AFI suspicion flag signaled around 500 days prior to their eventual surgery; over 90% had an AFI suspicion flag signaled at 400 days before surgery. A clinician could potentially use this information to help determine which sites to closely monitor for disease progression. Commercial AFI instruments such as the VELscope (LED Dental) are based on subjective interpretations and require an expert clinician to evaluate the severity level of the imaged lesion. The capability to objectively differentiate low-risk and high-risk OPMD could have important implications for clinical patient management in high resource settings as well as in areas with lower skilled examiners.

The complexity of evaluating patients with OPMD can be appreciated through the analysis of this patient cohort. In many cases, it is difficult to assemble and analyze images of a lesion over multiple visits, because the image may be captured at a slightly different field of view, angle, or perspective. Also, as these patients were under long-term surveillance at a tertiary cancer hospital, many of them had complex patient histories with multiple lesions being tracked over time. For example, some patients who did not have a surgery at an oral site that was being imaged did have surgeries at other areas of the oral cavity. Due to these complexities, a limitation of this study is that a relatively small number of patients met our inclusion criteria, though we had a large number under surveillance.

Numerous studies with differing patient populations have assessed AFI for detection of oral cancer and precancer, and it has been shown to have high sensitivity in identifying abnormal tissue (14, 21). In this study, we also found AFI to be very sensitive. Particularly, we chose the second AFI level 3 score in a timeline as the suspicion flag because the first AFI level 3 score resulted in too many false-positive results. Additionally, AFI raised a suspicion flag for one of the surveillance patients when CI did not. Regardless, AFI can be a useful aid for assisting clinicians in identifying any potential high-risk lesions.

It is evident there is still room for improvement in using optical imaging and clinical examination to monitor a patient over time. For example, patient #7 did not have an AFI or CI suspicion flag over a long period of observation. A CI suspicion flag was signaled on day −24, and a biopsy was taken at this clinic visit revealing high-grade dysplasia; following surgery, histopathology results from the surgical specimen indicated the presence of cancer. Unfortunately, imaging was not performed on the day of the clinic visit or surgery, so a comparison with quantitative imaging cannot be made. Regardless, this patient had been seen 50 days earlier on day −74 and had a risk level 1 imaging score and abnormal low-risk CI score. This patient highlights the complexity of managing oral premalignant lesions even under expert clinical evaluation and monitoring with AFI.

Because of the retrospective nature of this study and analysis, the AFI imaging results were not used to make clinical decisions such as performing a biopsy or proceeding to surgery. It is intriguing to consider whether monitoring these quantitative and objective data over time may assist the clinician in decision-making during clinical surveillance for patients with OPMD. The ability to objectively and noninvasively monitor for progression of OPMD would augment clinical decision-making at the patient level. The capability to detect trends in the probability of neoplasia at OPMD and the need for increased surveillance due to AFI suspicion would be of great value. Although these preliminary observations are encouraging, long-term follow-up of a large cohort of patients is needed to assess the clinical usefulness of these optical methods in monitoring OPMD patients.

Disclosure of Potential Conflicts of Interest

R.A. Schwarz has ownership interest (including patents) for IP licensed from the University of Texas at Austin by Remicalm LLC. A.M. Gillenwater has ownership interest (including patents) for IP licensed from the University of Texas at Austin by Remicalm LLC. R. Richards-Kortum has ownership interest (including patents) for IP licensed from the University of Texas at Austin by Remicalm LLC. No potential conflicts of interest were disclosed by the other authors..

Authors' Contributions

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Development of methodology: K.D. Cherry, R.A. Schwarz, A.M. Gillenwater, R. Richards-Kortum

Acquisition of data (provided animals, acquired and managed patients, provided facilities, etc.): K.D. Cherry, R.A. Schwarz, E.C. Yang, I.S. Vohra, H. Badaoui, M.D. Williams, N. Vigneswaran, A.M. Gillenwater, R. Richards-Kortum

Analysis and interpretation of data (e.g., statistical analysis, biostatistics, computational analysis): K.D. Cherry, R.A. Schwarz, E.C. Yang, M.D. Williams, A.M. Gillenwater, R. Richards-Kortum

Writing, review, and/or revision of the manuscript: K.D. Cherry, R.A. Schwarz, E.C. Yang, M.D. Williams, N. Vigneswaran, A.M. Gillenwater, R. Richards-Kortum

Administrative, technical, or material support (i.e., reporting or organizing data, constructing databases): K.D. Cherry, R.A. Schwarz, H. Badaoui

Study supervision: K.D. Cherry, A.M. Gillenwater, R. Richards-Kortum

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