Predicting Clinical Outcome following Oral Potentially Malignant Disorder Treatment - The Role of Adjunctive Diagnostic Techniques

M.L. Goodson, P. Sloan, V. Wadehra, S.J. Johnson, C.M. Robinson, A.M. Mowatt, P.J. Thomson

Abstract

Oral potentially malignant disorders (PMD) precede invasive squamous cell carcinoma (SCC), a lethal and deforming disease of rising incidence. Despite the ability to recognize suspicious mucosal lesions in clinical practice, it is impossible to predict clinical outcome or malignant transformation risk for individual patients. The specific aim of this study was to assess the usefulness of the diagnostic adjunctive techniques, VELscope® autofluorescence imaging and Orcellex® brush biopsy cytology, in addition to conventional clinical examination and histopathology in predicting clinical outcome following PMD treatment. 148 patients presenting with new, single-site PMD lesions were followed for between 20 to 48 months post-diagnosis and overall outcome determined as either disease resolution or persistent/progressive disease. Univariate analysis confirmed that conventional clinico-pathological parameters including patient age, sex, PMD lesion appearance or size, and length of follow up were non-significant predictors of outcome, nor was VELscope® appearance or Orcellex® brush biopsy cytology. Oral site, however, may be a significant factor with floor of mouth and buccal mucosa both prone to persistent/progressive disease (p=0.041). Multivariate analysis showed that interventional laser surgery excision was the most significant predictor of disease-free status (p<0.0001). Although not independent, accurate predictors of clinical outcome both VELscope® imaging and Orcellex® cytology remain useful diagnostic adjuncts during the assessment of PMD patients in specialist services.

Citation:

All Rights Reserved with Photon.
Photon Ignitor: ISJN37924853D658127032014
preceded by potentially malignant disorders (PMD), recognizable mucosal diseases such as localized leukoplakia or erythroplakia lesions or more widespread conditions, which all harbour a significantly increased risk of squamous carcinoma development (van der Waal, 2009). Estimates of the incidence and prevalence of PMD vary widely between geographical locations and populations studied but estimates of prevalence suggest an overall figure of between 2 to 3%, with the vast majority of lesions appearing clinically as leukoplakias, usually presenting on the floor of the mouth, ventro-lateral tongue and buccal mucosa. Whilst previously a disorder predominantly seen in older, male patients, increasing evidence now suggests that a much younger population is at risk (Napier and Speight, 2008).

Despite the ability to identify such disorders in patients, clinicians have been unable to predict lesion behaviour or quantify the risk of malignant transformation. Overall estimates regarding PMD clinical outcome have suggested that 40% change very little or progress only slightly with time, 20% may regress and 20% progress to malignancy, but these are highly anecdotal and retrospective observations and the natural history of PMD remains poorly documented (Speight, 2007; Napier and Speight., 2008). A recent systematic review quoted a 12% cancer rate over a mean transformation time of 4.3 years (Mehanna et al., 2009).

Historically, there has been little consensus on PMD management, although most authorities now recommend incision biopsy for histological assessment followed by whole lesion excision for more definitive histopathological diagnosis and effective treatment (Thomson and Wylie., 2002; van der Waal, 2009).

Recently, a number of potentially useful non-invasive diagnostic adjunctive tools have been developed to attempt to detect early signs of precancerous change within the oral cavity. These include optical imaging techniques such as tissue autofluorescence and brush biopsy procedures but, as most reports in the literature are based primarily upon anecdotal evidence, their true value in clinical practice is yet to be determined (Messadi, 2013).

### 1.2 Tissue autofluorescence

Autofluorescence of normal tissue under high-intensity light excitation is produced by endogenous fluorophores, whereas in dysplastic or neoplastic tissue the increase in cellular DNA, increasing epithelial thickness, altered collagen content and enhanced vascularity combine to alter normal light scattering and absorption leading to colour changes that can be visualized during clinical examination. The VELscope® (Visually Enhanced Lesion Scope, LED Dental Inc., White Rock, British Columbia, Canada) is a portable hand-held device allowing direct visualization of the oral cavity (Figure 1A). Under a 400-460 nm blue excitation light normal mucosa emits a pale green autofluorescence viewed through the handpiece filter, whilst neoplastic or dysplastic tissue appears dark (Figure 1B).

**Figure 1:** (A) the hand-held VELscope® device in use in the clinic and (B) the dark appearance of biopsy proven severely dysplastic palatal mucosa seen through the device.
1.3 Oral exfoliative cytology
Cytodiagnosis is the process whereby individual cells are loosened from their tissue of origin and transferred to a cytological slide for microscopic evaluation to diagnose disease. Whilst effective sampling of exfoliated oral epithelial cells for cytological analysis has proved problematic in the past, a number of new cell collection devices have been developed recently to facilitate full-thickness sampling of stratified epithelium. The Orcellex® brush (Rovers Medical Devices BV., the Netherlands) is a novel cytobrush with a specially designed head comprising 5 segments of high-density fibres designed for optimal cell collection, storage and subsequent release of cell material from all oral epithelial layers. The brush head is placed firmly against the mucosa and rotated 10 times (Figure 2A), then detached and transferred to BD Sure Path TMethanol-based preservative fluid for transfer to the laboratory.

We have previously shown that the clinical technique is non-invasive, fast and highly acceptable to patients (Goodson et al., 2011). Liquid-based cytology improves cell distribution and produces highly cellular specimens for accurate cytological evaluation (Figure 2B).

Figure 2: (A) the Orcellex® brush biopsy in use to collect cell samples from the oral mucosa and (B) the highly cellular appearance of a Papanicolaou stained thin-layer liquid-based cytology preparation seen under microscopic examination x10.

The accuracy of VELscope® tissue autofluorescence and Orcellex® brush biopsy cytology have not so far been assessed as predictive tools in PMD management. Since they both potentially assess an entire mucosal lesion rather than just examining a small incision biopsy sample, they may have an improved diagnostic and predictive role in determining the nature, clinical behaviour and ultimate outcome of potentially malignant lesions.

2. Objectives of Research
The aim of this paper was to determine the ability of the diagnostic adjunctive techniques, VELscope® tissue autofluorescence and Orcellex® brush biopsy, to act as predictors of clinical outcome following treatment of a cohort of PMD patients, and to compare the efficacy of these techniques with conventional clinico-pathological assessments.

3. Materials and Methods

3.1 Patients
Following ethical committee approval and informed patient consent, 148 consecutive PMD patients attending the Maxillofacial Oncology/Dysplasia clinics at Newcastle upon Tyne in Northern England over a 2-year period (January 2009 to December 2010) were recruited to the study. All were new patients, with no prior history of oral cancer or PMD and all presented with single-site, oral mucosal lesions.

3.2 Clinical assessment
Patients underwent standard clinical examination, supplemented by visual inspection using the hand-held tissue autofluorescence VELscope® instrument. Lesion site, size, appearance and VELscope® grading, using both a binary (‘normal’ green autofluorescence or ‘abnormal’ dark
appearance) and a 4-point increasing severity scale (whereby 0 was ‘normal’, 1 white, 2 grey and 3 black in appearance) were recorded. Patients then underwent a brush biopsy, using the standardized technique described and illustrated above (Figure 2a), followed within 2 weeks by conventional, incision biopsy.

3.3 Laboratory procedures
Cytology specimens were all processed and stained using the BD Prep Stain™ automated process in which, following density gradient centrifugation to enrich the cellular sample and remove obscuring non-diagnostic debris, discretely stained, thin-layer slide preparations were produced. Reporting of specimens was carried out by 2 experienced cytologists (VW and SJJ), applying the Bethesda guidelines for liquid based squamous cellularity (Solomon, 2002). Having confirmed an adequate cellular sample, cytology analysis described cells as normal, hyperkeratosis, or exhibiting cellatypia or mild, moderate or severe dyskaryosis, or suggestive of SCC; the presence of inflammatory cells and candida were noted as appropriate.

All incision and laser excision biopsy specimens underwent standardized histopathology examination by 2 experienced oral pathologists (PS and CMR) working to agreed diagnostic criteria and using the World Health Organization (WHO) classification to grade specimens into mild, moderate or severe dysplasia categories or carcinoma-in-situ (Gale et al., 2005); both pathologists independently assessed the biopsy material and discordant grading was resolved by review and consensus.

3.4 Treatment
Interventional management was non-randomized, but based upon a well-established and standardized protocol (Thomson and Wylie, 2002) and all surgical procedures were carried out by the same operators (PJT and MLG). Patients deemed ‘low risk’ for disease progression or malignant transformation, whose lesions exhibited no or only mild dysplasia, who were committed to smoking cessation and risk factor modification and able to attend for regular clinic monitoring, were treated by observation and active surveillance. ‘High risk’ cases, those with severe dysplasia, continuing smokers and alcohol users, young patients or those with no obvious risk factors, were offered laser excision of mucosal lesions within 6 weeks of incision biopsy diagnosis.

3.5 Follow-up and clinical outcome
All patients were followed-up in clinic at 3-monthly intervals for a minimum of 18 months post-diagnosis. Clinical outcome at most recent follow-up appointment was classified using the following definitions (Diajil et al., 2013): Disease Resolution, patient free of PMD disease, Persistent Disease, whereby PMD persisted at the same oral site, Recurrent Disease, when PMD lesion recurred at same site following treatment, Further Disease, distinguishing PMD lesion development at new sites, Malignant Transformation, where invasive carcinoma arose at the same site as a precursor mucosal lesion, and Oral Cancer Development, in which a new site carcinoma developed distinct from precursor lesion sites.

3.6 Justification for research
We are not aware of any other publication that has examined the ability of VELscope® tissue autofluorescence imaging and Orcelex® brush cytology to predict clinical outcome following oral PMD treatment.

3.7 Statistical Analysis
Analyses were performed using SPSS, version 19.0 (Statistical; Package for the Social Sciences, Chicago, IL, USA). Univariate analysis was undertaken using chi-square or t-test to examine the effects of age, sex, lesion appearance, size, site, VELscope® grading, brush biopsy cytology and excision biopsy histopathology on clinical outcome. Backwards multiple logistic regressions were carried out to determine which of the aforementioned covariates significant predictors of clinical outcome were. To optimize sample sizes for statistical calculations, outcome was defined as either: disease resolution or persistent/progressive disease (comprising persistent, recurrent or further dysplastic lesions and malignant transformation or cancer development).

4. Results
4.1 Patient demographics
148 patients participated in the study comprising 86 male (57%) and 66 female patients (43%), with an age range of 28 to 92 years and a mean of 60.2 years. No significant influences of age (p=0.848, independent t-test) or sex (p=0.48, chi-square test) on overall clinical outcome were seen.

4.2 Oral lesions
The vast majority of lesions presented as leukoplakias (137), with 13 appearing as
erthroplakias and 2 erythroleukoplakias. Lesion size varied from 100 to 400 mm² (mean 198 mm²), with 76 arising on the floor of mouth and ventro-lateral tongue, 28 on buccal mucosa, 23 within the soft palate, pillar of fauces and retro molar region, 18 on the alveolar mucosa and 3 on the tongue dorsum. Disease resolution was more commonly observed following treatment of lesions arising on ventro-lateral tongue, palate and labial commissure sites, whilst persistent/progressive disease was particularly seen on the floor of the mouth and buccal mucosa (p=0.041, chi-square test). There were no significant relationships between lesion appearance or size and clinical outcome (p=0.075 and p=0.12 respectively, chi-square test).

4.3 VELscope® examination
All 148 patients underwent VELscope® examination, and Table 1 summarizes the results for both the 4-point severity scale and the binary ‘normal/abnormal’ system. No significant relationship was seen between either 4-point or binary VELscope® grading and outcome (p=0.294 and p=0.128 respectively, chi-square tests).

4.4 Brush biopsy and cytology
The results for cytology diagnoses are listed in Table 2; whilst the single most common diagnosis was hyperkeratosis (44), cellular atypia and varying degrees of dyskaryosis were seen in 63 cases. By comparing ‘normal’ cytology results (82) with ‘dyskaryosis’ (66), no significant influence of cytology diagnoses on clinical outcome was noted (p=0.001, chi-square test).

4.5 Incision biopsy histopathology
The results for the 148 incision biopsies are listed in Table 3; hyperkeratosis, and hyperkeratosis plus lichenoid inflammation comprised the most common diagnoses (71) but dysplasia was confirmed in 51 cases. By dividing incision biopsy results into the categories of ‘hyperkeratosis’ (in which 28 out of 35 exhibited persistent/progressive disease) and ‘premalignant’ (in which similar numbers of disease resolution, 58, and persistent/progressive outcomes, 55, were seen) a significant difference in clinical outcome was confirmed (p=0.001, chi-square test).

4.6 Laser excision histopathology
61 patients in this study were judged to be ‘high risk’ and underwent either laser excision (51) or ablation (10). Table 4 confirms that the majority of excised lesions exhibited dysplasia or carcinoma-in-situ (34). Disease resolution was more common in laser treated patients (53 out of 61), compared with non-laser patients (12 out of 87); this was statistically significant (p<0.001, chi-square test).

<table>
<thead>
<tr>
<th>4 Point System</th>
<th>No. of Lesions</th>
<th>Binary System</th>
<th>No. of Lesions</th>
</tr>
</thead>
<tbody>
<tr>
<td>0 (Normal Mucosa)</td>
<td>25</td>
<td>0 (Normal / Green)</td>
<td>81</td>
</tr>
<tr>
<td>1 (White)</td>
<td>56</td>
<td>1 (Abnormal / Dark)</td>
<td>67</td>
</tr>
<tr>
<td>2 (Grey)</td>
<td>49</td>
<td></td>
<td></td>
</tr>
<tr>
<td>3 (Black)</td>
<td>18</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Table 2: Orcellex® Brush Cytology Diagnoses for 148 Mucosal Lesions

<table>
<thead>
<tr>
<th>Cytology Diagnosis</th>
<th>No. of Lesions</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal</td>
<td>27</td>
</tr>
<tr>
<td>Hyperkeratosis</td>
<td>44</td>
</tr>
<tr>
<td>Candidal Infection</td>
<td>12</td>
</tr>
<tr>
<td>Cell Atypia</td>
<td>16</td>
</tr>
<tr>
<td>Mild Dyskaryosis</td>
<td>17</td>
</tr>
<tr>
<td>Moderate Dyskaryosis</td>
<td>11</td>
</tr>
<tr>
<td>Severe Dyskaryosis</td>
<td>19</td>
</tr>
<tr>
<td>Squamous Cell Carcinoma</td>
<td>2</td>
</tr>
</tbody>
</table>

Table 3: Incision Biopsy Histopathology Diagnoses for 148 Mucosal Lesions

<table>
<thead>
<tr>
<th>Histopathology Diagnosis</th>
<th>No. of Lesions</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hyperkeratosis</td>
<td>35</td>
</tr>
<tr>
<td>Hyperkeratosis + Lichenoid Inflammation</td>
<td>36</td>
</tr>
<tr>
<td>Candidal Infection</td>
<td>6</td>
</tr>
<tr>
<td>Mild Dysplasia</td>
<td>24</td>
</tr>
<tr>
<td>Moderate Dysplasia</td>
<td>15</td>
</tr>
<tr>
<td>Severe Dysplasia</td>
<td>12</td>
</tr>
<tr>
<td>Proliferative Verrucous Leukoplakia</td>
<td>19</td>
</tr>
<tr>
<td>Squamous Cell Carcinoma</td>
<td>1</td>
</tr>
</tbody>
</table>

Table 4: Excision Biopsy Histopathology Diagnoses for 51 Laser Excised Mucosal Lesions

<table>
<thead>
<tr>
<th>Histopathology Diagnosis</th>
<th>No. of Lesions</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hyperkeratosis</td>
<td>2</td>
</tr>
<tr>
<td>Hyperkeratosis + Lichenoid Inflammation</td>
<td>2</td>
</tr>
<tr>
<td>Candidal Infection</td>
<td>3</td>
</tr>
<tr>
<td>Mild Dysplasia</td>
<td>16</td>
</tr>
<tr>
<td>Moderate Dysplasia</td>
<td>7</td>
</tr>
<tr>
<td>Severe Dysplasia</td>
<td>10</td>
</tr>
<tr>
<td>Proliferative Verrucous Leukoplakia</td>
<td>10</td>
</tr>
<tr>
<td>Carcinoma-in-Situ</td>
<td>1</td>
</tr>
</tbody>
</table>
4.7 Clinical outcome
The follow-up time in this study ranged from 20 to 48 months, with a mean of 26.7 months; there was no significant difference in follow-up time between disease resolution and persistent/progressive disease groups (p=0.485, independent samples t test).

Table 5 contrasts outcome data at most recent clinic appointment for both the observation and laser intervention treatment groups. Whilst the majority of laser excision cases showed disease resolution (87%), most of the observation cohort exhibited persistent disease (75%) but also a much higher level of further (new site) dysplasia compared to the laser intervention group (10% and 2%, respectively).

Table 5: Clinical Outcomes for 61 Laser Treated Patients (Excision or Ablation) and 87 Observation (Non-Laser) Patients

<table>
<thead>
<tr>
<th>Clinical Outcome</th>
<th>Laser Patients (%)</th>
<th>Observed Patients (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Disease Resolution</td>
<td>53 (87%)</td>
<td>12 (14%)</td>
</tr>
<tr>
<td>Persistent Disease</td>
<td>0 (0%)</td>
<td>65 (75%)</td>
</tr>
<tr>
<td>Recurrent Disease (Same Site)</td>
<td>5 (8%)</td>
<td>0 (0%)</td>
</tr>
<tr>
<td>Further Disease (New Site)</td>
<td>1 (2%)</td>
<td>9 (10%)</td>
</tr>
<tr>
<td>Squamous Carcinoma Cell</td>
<td>2 (3%)</td>
<td>1 (1%)</td>
</tr>
</tbody>
</table>

Multivariate analysis showed that observational treatment and thus absence of interventional laser surgery was the most significant predictor of a persistent or progressive disease outcome (p<0.0001, chi-square test of correlation between covariates).

5. Discussion

5.1 Prediction of clinical outcome
The ability to predict clinical outcome for oral potentially malignant disorders remains elusive in clinical practice, probably due to a lack of understanding of the natural history of the disease, confusion over terminology, limited agreement on therapeutic interventions and uncertainty regarding patient follow-up (Dajil et al., 2013). In the absence of meaningful randomized, prospective clinical trials, however, results from patient cohort studies such as this one provide the most consistently reliable data for determining outcome statistics following oral precancer treatment.

It is interesting that a significant predictor of persistent or progressive PMD disease in this study was the anatomical site of origin of the mucosal lesion. We have previously demonstrated inherently high levels of cell proliferative activity at both floor of mouth and buccal mucosa sites, and this may explain the resulting enhanced risk of on-going dysplastic disease (Thomson et al., 1999).

5.2 VELscope® imaging
Consensus opinion in the current literature suggests that VELscope® examination is highly subjective, unable to definitively diagnose epithelial dysplasia in the oral cavity and cannot readily discriminate between ‘high’ and ‘low-risk’ lesions (Awan et al., 2011; Farah et al., 2012; Messadi, 2013). We have previously reported on improving the reliability and objectivity of VELscope® using a 4-point severity grading score and confirmed its use in distinguishing a range of varying oral lesions including hyperkeratoses (white appearance), lichenoid inflammatory lesions (variably dark), and mild, moderate or severe dysplasia’s and carcinoma-in-situ (increasingly grey/black) and SCC (black); Goodson et al., 2011; Goodson et al., 2013.

To our knowledge, this is the first study to directly assess the predictive value of tissue autofluorescence in PMD management. Neither the 4-point nor the binary grading systems were ultimately predictive of lesion behaviour, but there is no doubt that VELscope® imaging is a helpful adjunct during visual examination of the oral cavity and is particularly beneficial in the follow-up of multiple lesion PMD patients in specialist clinics (Hamadah et al., 2010).

5.3 Brush biopsy cytology
This is also the first study to directly assess the predictive value of Orcellex® brush biopsy cytology on clinical outcome following oral potentially malignant disorder treatment. The number of patients with disease resolution was similar in both ‘normal’ (31) and ‘dyskaryosis’ cytology categories (34), whilst persistent/progressive disease was more commonly seen in patients with ‘normal’ results (51 compared to 32 ‘dyskaryosis’), presumably because dyskaryotic lesions were usually excised.

We previously reported upon both patient acceptability and diagnostic efficacy of Orcellex® brush cytology in PMD patient assessment (Goodson et al., 2011b), but it is disappointing that there was no clear predictive role for the technique in this study. There is no doubt, however, that brush biopsy
is well tolerated and popular with patients and certainly has a role as an additional monitoring tool during longer term patient follow-up regimes (Goodson et al., 2011b).

5.4 Histopathology diagnoses
Histopathological examination remains the ‘gold standard’ for both provisional incision biopsy and definitive excision biopsy diagnoses in PMD management and significantly influence clinical outcome. As we have previously noted, however, disease severity often requires up-grading following excision specimen examination (Goodson et al., 2011a), and in this study, 12 out of 51 laser excision specimens were re-classified as more severely dysplastic compared with their initial, incision biopsies.

5.5 Clinical outcome
The clinical outcomes reported for the patients in this study were comparable to that previously presented in a number of Newcastle cohort studies, as shown in Table 6. Although benefiting from a large sample size (148 patients), this study analysis was complicated by the division of patients into observation and laser intervention sub-groups, although this is clearly a ‘real-life’ clinical scenario and diagnostic adjunctive techniques were applied to all presenting patients regardless of treatment decisions.

Table 6: Comparative Clinical Outcome Data (Newcastle Patient Cohort Studies 2002 – 2013)

<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>57 Patients</td>
<td>199 Patients</td>
<td>40 Patients</td>
<td>78 Patients</td>
<td>100 Patients</td>
<td></td>
</tr>
<tr>
<td>Disease Resolution %</td>
<td>76</td>
<td>67</td>
<td>60</td>
<td>64</td>
<td>62</td>
</tr>
<tr>
<td>Persistent Disease %</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Recurrent Disease (Same Site) %</td>
<td>6</td>
<td>15</td>
<td>22</td>
<td>18</td>
<td>18</td>
</tr>
<tr>
<td>Further Disease (New Site) %</td>
<td>12</td>
<td>14</td>
<td>18</td>
<td>14</td>
<td>14</td>
</tr>
<tr>
<td>Squamous Cell Carcinoma %</td>
<td>6</td>
<td>4</td>
<td>0</td>
<td>4</td>
<td>7</td>
</tr>
</tbody>
</table>

5.6 Disease resolution
In this investigation, 61 patients underwent either CO₂ laser excision or ablation and, whilst resolution of PMD disease was seen in 87% of laser patients, disease-free status was seen in only 14% of observed cases. The use of laser surgery was found to be a significant predictor of disease resolution in the study and this is probably unsurprising owing to the demonstrable efficacy of interventional laser surgery in PMD treatment. It is notable, however, that recurrent or further dysplasia (10% of cases) and malignant transformation (3%) remain a risk and that long-term patient follow-up and surveillance is mandatory for all PMD patients (Thomson and Wylie, 2002; Stocker et al., 2005; Hamadah and Thomson, 2009; Diajil et al., 2013).

5.7 Persistent disease
Persistence of same site mucosal disease was seen in 65 out of 87 observed lesions (75%), but not in any cases following laser excision. There are few data in the literature with which to compare these figures, although we have previously shown that 77% of observed PMD lesions are likely to persist 3 years post-diagnosis without surgical intervention (Thomson et al., 2009).

5.8 Recurrent disease
Reappearance of same-site dysplastic lesions was seen in 8% of laser treated patients, which is well within the range of 6 to 22% previously reported for our treated patient cohorts (Table 6); it is unsurprising that ‘recurrence’ was not reported in the observed patient cohort.

5.9 Further disease
Appearance of further (new-site) dysplastic lesions occurred in 10 patients, but was more frequently seen in observed (10%) compared to laser surgery patients (2%). This is an interesting observation and, whilst possibly anecdotal, certainly adds weight to the role of intervention as opposed to observation in PMD management, although it is difficult to convincingly explain why localized laser treatment might reduce the influence of field change can cerization in this way.

5.10 Malignant transformation
Three patients developed same-site SCC during the study period: 2 at 36 months post-laser excision of severely dysplastic lesions and 1 at 39 months at the site of an observed mildly dysplastic lesion. This is similar to our
previous report that SCC development is most likely to occur during the first 3 years following PMD treatment (Diajil et al., 2013). Although involving only 1 out of 87 observed patients, it is clinically significant that malignant transformation was seen in a non-treated PMD lesion exhibiting only mild dysplasia.

Research Highlights
• Interventional laser surgery is an effective treatment modality in PMD management
• Reliable prediction of clinical outcome for individual PMDs remains elusive
• Tissue autofluorescence imaging and brush biopsy cytology are useful adjunctive techniques in specialist PMD clinics, but are not in themselves accurate predictive tools
• Further research is warranted to determine the precise role of VELscope® imaging and Orcellex® brush cytology in PMD diagnosis and management.

Limitations of Research
As a one-centre, UK-based, non-randomized patient cohort study, there are limitations to the global significance and applicability of these study results.

Recommendations
Interventional laser surgery is recommended as the most effective treatment modality for oral PMD lesions. Whilst VELscope® tissue autofluorescence imaging and Orcellex® brush cytology do not predict individual lesion behaviour, they are recommended as useful adjuncts to the assessment and clinical follow-up of oral lesions in specialist practice. Further assessment of these techniques is warranted in larger, multi-centre clinical trials.

Conclusions
Whilst neither tissue autofluorescence imaging nor brush biopsy cytology proved to be independent, accurate predictors of clinical outcome in this study, they remain useful diagnostic adjuncts during the assessment, interventional treatment and post-treatment surveillance of PMD patients in specialist practice. Interventional laser surgery to excise PMD lesions remains the preferred treatment modality.

Authors’ Contributions, Competing Interests and Funding Aspects
ML Goodson led the data collection and analysis, assisted by AM Mowatt, whilst PJ Thomson and ML Goodson coordinated the clinical assessment and management of all patients in this study. V Wadhera and SJ Johnson provided specialist cytology diagnostic services and P Sloan and CM Robinson oral pathology expertise. All authors contributed to the intellectual content and writing of the manuscript. No competing or conflicts of interest are declared, nor was any funding received for this study.

Acknowledgements
The authors wish to acknowledge the help of colleagues in the Departments of Cellular Pathology and Medical Physics at the Royal Victoria Infirmary in Newcastle upon Tyne without whom, this study would not have been possible.

References


leukoplakia management. Oral Oncology. 47 S128-129.


