Separating the Good from the Bad and Ugly

The Use of Non-Invasive Optical and Molecular Tests in the Management of an Oral Lesion

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Professor, Faculty of Dentistry, UBC
Faculty Disclosure

I have no conflict of interest in relation to the devices or tools used in this presentation.
Vancouver, British Columbia, Canada

University of British Columbia
Patients with
1. Head and Neck Cancers: pre-, during, and post- radiation and/or chemoradiation
2. Oral cancers: pre- and post- surgery
3. Oral precancerous management
4. Hematopoietic malignancies for pre-chemo or pre-BMT assessment
5. Cancers require Biosphosphonates treatment
Oral Mucosal Lesions
Early Detection and Risk Assessment

Finding Solutions through Research !!

Non-invasive Optical and Molecular Tests

1. VELScope®
2. Quantitative Cytology
3. Loss of Heterozygosity
VELScope®

Introducing the VELscope Vx™
Shining Blue Light on Oral Health
Building a Visualization Device

Calum MacAulay  Pierre Lane

Fluorescence Visualization (FV)

**Fluorescence due to FAD/NADH (cellular level)**

**Fluorescence due to collagen cross-links & elastin (in the subtending stroma)**

- Decomposition (or restructuring) of the Collagen Matrix
- ↑ Micro-Vascularization
- → Nuclear Scattering
- ↑ Metabolic Activity (↑ FADH/NAD)
- Epithelial Thickening

**FVR (fluorescence visualization retained)**

**FVL (fluorescence visualization loss)**

Poh et al., 2009 (book chapter)
Identification of clinically not-apparent change

Poh et al., Head and Neck, 2007
FV helps on the decision of where to biopsy

Diffuse non-homogenous leukoplakia at the right ventral tongue
When to biopsy

12 months
The extension of the high-grade oral lesion

Clinical white light image –
Invasive cancer and severe dysplasia (arrow)

Fluorescence visualization (FV)
FV in the operating room

Question 1:

Can we use this tool in the operating room to assist the decision of surgical margin?
FV in the operating room

Severe dysplasia

No dysplasia

Carcinoma

No dysplasia

Clin Can Res, 12(22), 6716-22, 2006
FV can recognize **histologically high-grade and molecularly high-risk areas**.

19/20 lesions has FV positive area outside the tumor area and the FV margin is **not evenly around** the tumor area.

**Clin Can Res, 12(22), 6716-22, 2006**
Question 2: Does the tool really make impact on patient’s outcome, i.e., recurrence rate?
Study Scheme: 2004-2009 British Columbia

Eligible patients (N=246)

- SCC (N=156)
  - T1=104; T2=52
  - FV-guided (N=92; 59%)
  - Control (N=68; 41%)

- HGL (N=90)
  - D3=41; CIS=49
  - FV-guided (N=62; 69%)
  - Control (N=28; 31%)

FV appears to reduce local recurrence

3-year recurrence rate reduces from 41% to **6.5%**.

COOLS trial -
Canadian Optically-guided approach for Oral Lesions Surgical Trial

• Phase III randomized controlled trial
  ▪ 400 patients: Severe dysplasia/CIS; T1 and T2 oral cancer

• 4.8 million, 6 – year project (September 1st, 2010) – milestone driven project

Eligible patient identification at each geographic site

Stratify

Preinvasive high-grade lesions (N=160) →

Randomize

FV*-guided surgery (N= 80 + 120) →

Invasive cancers (N=240) →

White-light-guided surgery (N=80+ 120) →

Endpoint Local recurrence

* FV: fluorescence visualization

Poh et al. BMC Cancer 2011, 11:462
Pan-Canadian Network for Oral Cancer Control
Incidence and Mortality

![Graph showing cumulative incidence of local recurrence and died before local recurrence for HGIL and SCC over months, with numbers at risk and cumulative incidence values.]
Masking by Infection and Inflammation

Oral candidiasis

Lichen planus
Oral candidiasis
Lichen planus
Quantitative Cytology using high-resolution microscopy imaging - cNPS (cytology-Nuclear Phenotype Score)

Development of cNPS to automatically capture alterations in
1. DNA content (ploidy)
2. Nuclear morphology features (~110)
Quantitative Cytology using high-resolution microscopy imaging - cNPS (cytology-Nuclear Phenotype Score)
Quantitative cytology
Two-step screening

Step 1: Clinical white light and/or FV (VELScope®)
Step 2: Quantitative Cytology

<table>
<thead>
<tr>
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<th>FV retained (FVR)</th>
<th>FV loss (FVL)</th>
<th>cNPS-/FVL</th>
<th>cNPS+/FVL</th>
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<td>Trauma</td>
<td>87%</td>
<td>&lt;10%</td>
<td>92%</td>
<td>20%</td>
</tr>
<tr>
<td>Inflammation</td>
<td>100%</td>
<td>23%</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Infection</td>
<td>100%</td>
<td>100%</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

N=68 N=28 N=12
Suspicious oral lesion

Biopsy

Persistent
3-4 weeks after eliminating the possible etiologies
Suspicious oral lesion

Biopsy

Persistent
3-4 weeks after eliminating the possible etiologies

BC Oral Biopsy Service
Infobahn Oral Biopsy Service (https://iobs@dentistry.ubc.ca)

- Get pathology report online
- Accurate description and diagnosis
- Manage patients and biopsy history
Welcome to Your OBS Dashboard

- Manage My Patients:
  - Create or Manage Patients
  - Manage Biopsy History
  - View Pathology Reports

- Make a Referral:
  - Refer patients with:
    - Oral cancer
    - Oral dysplasia

- Manage My Profile:
  - Account details
  - Clinics
My Patient » Mucosal Biopsy - Add General Information

Patient iOBS Biopsy Identifier: 2016031501

Patient: Simpson, Homer, 1234567890, 1955-05-12, Male, Caucasian

General Information

Tobacco Usage (please check all that apply)
- Do not know
- Never
- Ever - Smokeless
- Ever - Smoking tobacco

Alcohol Information
- Do not know
- No
- Yes, if Men ≥ 3 drinks per day; Women ≥ 2 drinks per day

Biopsy Information
- Date of Current Biopsy: 2016-03-15
- Previous Oral Biopsy: Yes ☐ No ☑
ORAL MUCOSAL BIOPSY
<table>
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<tr>
<th>Biopsy Code</th>
<th>A</th>
<th>Biopsy Location</th>
<th>L. Middle dorsal tongue</th>
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<tr>
<td>Symptoms</td>
<td></td>
<td>Discomfort</td>
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</tr>
<tr>
<td>Duration</td>
<td></td>
<td>For Unknown</td>
<td>Months</td>
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<tr>
<td>Lesion Size (the largest dimension)</td>
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<td>1 - 2 cm</td>
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<tr>
<td>Colour</td>
<td></td>
<td>Mainly white</td>
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<tr>
<td>Clinical Appearance</td>
<td></td>
<td>Polyp / Lump / Bump</td>
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<tr>
<td></td>
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<td>Rough-surface / Verrucous</td>
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</table>
BC ORAL BIOPSY SERVICE
Oral Mucosal Biopsy

Biopsy Date: 09/10/2014

PATIENT  aaa, bbb  PHN: N/A; DOB: 11/10/2010; Female; Asian

HABIT  Tobacco: Smoking, Current, 20 cigarette(s)/day, 1 year(s); Alcohol: No; Marijuana: Do not wish to answer

PREVIOUS ORAL BIOPSY  No

CURRENT BIOPSY  Photo Attached: Yes (e-mail)

Bx A: Clinical Information: Incisional; 20mm x 12mm; Right Lateral Tongue; Asymptomatic; White, 60 month(s)
D/D: lichen planus, r/o dysplasia

Bx B: Clinical Information: Incisional; 8mm x 5mm; Left Buccal Mucosa; Asymptomatic; Ulcerated, 4 week(s)
D/D: lichen planus

Please mark site of lesion(s) on the printed form

CLINICIAN  Dr. F. Catherine Poh

Pathology Report Sent To: (Office 1) 675 West 10th Avenue, Vancouver, BC, V5Z1Z3

Please submit PRINTED FORM WITH SPECIMEN to the BC Oral Biopsy Service Vancouver General Hospital, Room 1400 JPPN1, 910 West 10th Avenue, Vancouver BC, V5Z 1M9.

Submit  Edit
SEARCH THROUGH YOUR PATIENTS

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<tr>
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<th>First Name</th>
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My Patient » Patient-Biopsy List

**Patient:** Simpson, Homer

1234567890, 1955-05-12, Male, Caucasian

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</table>

Showing 1 to 2 of 2 entries
Gold standard for risk assessment

Presence and degree of dysplasia
What is dysplasia?

I. At cellular level:
Criteria often used for dysplasia (WHO):

a. Irregular stratification or loss of polarity of the cells in the epithelium.
b. Increased mitoses
c. Nuclear hyperchromatism.
d. Increased nuclear/cytoplasmic ratio.
e. Polymorphism of cells.
f. Abnormal keratinization...

II. At architectural level:
Mild dysplasia

Dysplastic cells involving basal and parabasal cells
Moderate dysplasia
Dysplastic cells involving the lower half of the epithelial cells
Severe dysplasia
Dysplastic cells involving the lower 2/3 of the epithelial cells
Carcinoma in situ (CIS)

Dysplastic cells involving all the epithelial layers from bottom to top
Invasive squamous cell carcinoma
Basement membrane is disrupted by the dysplastic cells
Suspicious oral lesion

Biopsy

Persistent
3-4 weeks after eliminating the possible etiologies

BC Oral Biopsy Service

No Dysplasia

Community clinics

Low-grade Lesion
(verrucous hyperplasia, mild, moderate dysplasia)

Risk assessment clinics
@ Experienced community practitioners
Combined Head & Neck and Oral Medicine Clinics

High-grade Lesion
(severe dysplasia, carcinoma in situ, squamous cell carcinoma)

Treatment triage clinics
@ Oral Oncology, BC Cancer Agency

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Community clinics
Histological progression model of oral lesions

- Hyperplasia
- Mild Dysplasia
- Moderate Dysplasia
- Severe Dysplasia/CIS
- Invasive SCC

Low grade dysplasia
High risk? Low risk?
Loss of Heterozygosity (LOH) to Predict Malignant Risk for Oral Premalignant Lesions

• Published retrospective (2000) and prospective (2012) studies using LOH at 3p14 and/or 9p21 to predict risk of progression (25-35% in 5 years)
Loss of heterozygosity at 9p21

Collaborate with Cancer Genomic Lab, BCCA
Funded by Genome BC

High-risk profile of tongue and MR3 (LOH at 9p21 and/or 3p14, and 17p13) was significantly associated with progression (HR, 6.7; 95% CI, 2.6-17.6) with a specificity of 98.4% at identifying progressors.
Time-to-Progression Event Chart

Liu et al., 126 (1), 54–62(2018)
ddPCR platform:

- Using internal control – no need for control samples
- Can detect homozygous deletion
- Can be used to test brushing samples – non-invasive approach
ddPCR platform- non-invasive approach

Gains: 3q, 5p, 7p, 8q, 11q, 20p
Loss: 3p, 4q, 8p, 9p, 18q

Gains: 3q, 5p, 7p, 8q, 11q, 20q
Loss: 4q
iTOP Clinic
Combined Otolaryngology Head & Neck Surgery & Oral Medicine Clinics

4.8 million
944.735 km²
• *in vivo* Optical Devices
• Quantitative Cytology using Nuclear Phenotype Score
• Molecular tests
A good picture is better than a thousand words!

High resolution digital clinical images
High resolution digital clinical images
Toluidine blue (TB) is a metachromatic, acidophilic stain (Tolonium Chloride) and has higher affinity to nucleic acid.

Possible mechanisms:
- ↑ DNA &/or RNA and/or
- Defective intercellular barrier
  → the dye to reach deeper cell layers with higher DNA and RNA content
Toluidine Blue

Where is the lesion and where to biopsy?
55 y/o M, smoker, HCV + (#4278); Smoking tobacco since age 13, 40 pack year smoking history.

Carcinoma *in situ*
How to manage these at-risk oral lesions
Manage these at-risk oral lesions:

1. Cryotherapy
2. Topical Photodynamic Therapy
Topical Photodynamic Therapy using 20% 5-ALA (5-Aminolevulinic Acid)

58F NS
Family cancer history

18 months
3rd Topical photo dynamic therapy using 20% 5-ALA
Topical Photodynamic Therapy using 20% 5-ALA (Aminolevulinic Acid)

31 F Nonsmoker, moderate dysplasia follow up for 7 years

PDT x 2

2009

2016 0715

2016 0930
Effective Management of Oral Precancers

Clinical Trials of Effective Topical Treatments

Topical Photodynamic Therapy

32 F Nonsmoker

2017 March 24
A PATIENT'S JOURNEY

Oral lesion

Precancer

Cancer

Screened by GPs or dentists, using conventional white-light (VELScope exam)

QC brushing automatic cytology to sieve off trauma, inflammation, infection

Specialists for assessment & biopsy

Cryotherapy
Topical photodynamic therapy

REGISTRY (iOBS) of all mucosal biopsies
Patient information, pathology, molecular information (LOH, etc)
Acknowledgement

Collaborators
COOLS study group
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Dr. Calum MacAulay
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Dr. Stuart Peacock
Dr. Eitan Prisman
(Dr. Michele Williams)
Dr. Jonn Wu
Dr. Stephen Yip
Dr. Lewei Zhang
(in alphabetical order)

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Tony Han
Jessica Chen
Curtis Hughesman
Maria Lopes
David Lu
Kelly Liu
Katya Parfenova

All patients & families

UBC DENTISTRY
BC Cancer Foundation
BC Cancer Agency
Genome British Columbia
CIHR IRSC