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**Human papillomavirus (HPV) associated oropharyngeal
cancer: Case prevalence, diagnosis, and the potential for
screening in New Zealand.**

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of the requirements for the degree of

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Abstract

The incidence of oropharyngeal cancer (OPC) is increasing in developed countries, with many cases caused by Human Papilloma Virus (HPV). Since the mid-1990s, the incidence of OPC in New Zealand has almost quadrupled, but the role of HPV is uncertain. Thus, the objectives of this research were to establish the burden of HPV positive OPC in New Zealand, investigate diagnostic methods, early detection through PCR and cytology, and risk factors in a pre-vaccine population.

The results presented in this thesis demonstrate an increased prevalence of HPV positive OPC from 61.9% in 1996-98 to 87.5% in 2010-12 in the study population. HPV 16 was responsible for 98.5% of HPV positive OPC and results from the multivariable model showed an HPV positive patient was more likely to be aged under 60 years old and diagnosed in 2010-12. Descriptive Analysis of questionnaire data from OPC patients found that having ever given oral sex was the most significant risk factor for having an HPV (p16) positive tumour.

Comparison of p16, CK19, and HPV 16 DNA, RNA detection and viral load revealed ten cases in which the HPV status was incorrectly classified based on p16 alone, showing the need for clearer guidelines around the reporting of p16 results. These results are of particular importance as de-escalated therapies for HPV positive cases are under investigation. Moreover, viral nucleic acid and cytological abnormalities were detectable in brushings taken from conscious OPC patients. These previously undescribed cellular changes are comparable to cervical precancerous lesions and

showed a continuum of dysplasia in p16 positive cases only. This warrants further investigation.

Overall, this research has shown HPV positive OPC is a significant burden on the New Zealand health system and its incidence is increasing, thus supporting the recent inclusion of males into the nationally funded immunization schedule for Gardasil®9. However, OPC cases will continue to increase until the current vaccinated cohort reaches middle age. It is crucial that until this time, we focus on the early detection, improved diagnostics and reduced morbidities from treatment in the current pre-vaccine population.

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List of publications and presentations

Publications

Lucas-Roxburgh, R., Benschop, J., Lockett, B., van den Heever, U., Williams, R., & Howe, L. (2017). The prevalence of human papillomavirus in oropharyngeal cancer in a New Zealand population. *PLoS ONE*, 12(10), 1-13. doi: 10.1371/journal.pone.0186424

Conference Presentations

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Lucas-Roxburgh, R., Benschop, J., Lockett, B. & Howe, L. Descriptive analysis of a pre-vaccine population of oropharyngeal cancer patients in New Zealand (poster presentation). HPV 2018, 32nd International Papillomavirus Conference. October 2-6, 2018, Sydney, Australia.

Lucas-Roxburgh R., Benschop J., Lockett B., van den Heever U., Williams R. & Howe, L. Prevalence of HPV positive oropharyngeal cancer in New Zealand. 10th New Zealand Immunization conference and pre-conference workshop. September 8-9, 2017 Wellington, New Zealand.

Lucas-Roxburgh, R., Benschop, J., Lockett, B., Williams, R. & Howe, L. Detection of human papillomavirus (HPV) 16 E7 protein in oropharyngeal cancer biopsies with

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Lucas-Roxburgh, R. How old is too old: p16 testing on Oropharyngeal biopsies. Roche Histopathology User Group. March 22-28, 2015, Taupo, New Zealand.

Lucas-Roxburgh, R. (2015) Connect four: Using HRM to detect HPV genotypes in Oropharyngeal Cancer. Roche Molecular Diagnostics User Group. March 22-28, 2015, Taupo, New Zealand.

Invited speaker presentations

Lucas-Roxburgh, R. HPV and associated cancers / The burden of oropharyngeal cancer in New Zealand. Whanganui Inter-Professional Education (WIPE): HPV and oropharyngeal cancers. April 3, 2018, Whanganui, New Zealand.

Lucas-Roxburgh, R. HPV in oropharyngeal cancer. The immunisation advisory centre professional development day. May 9, 2018, Wellington, New Zealand.

Lucas-Roxburgh R. Prevalence of HPV positive oropharyngeal cancer in New Zealand. National Cervical Screening Program Clinical Update. November 29, 2017, Palmerston North, New Zealand.

Lucas-Roxburgh, R. HPV-associated Oropharyngeal Cancer in New Zealand. Royal College of Pathologists of Australasia (RCPA) Annual Scientific Meeting. September 23-25, 2016, Napier, New Zealand.

Lucas-Roxburgh, R. HPV Associated Oropharyngeal Squamous Cell Carcinoma. New Zealand Institute of Medical Laboratory Science (NZIMLS) South Pacific Congress. August 17-21, 2015, Auckland, New Zealand.

Awards

2017 Massey University Three minute thesis (3MT) – People’s choice award.

2015 Roche User Group, Histopathology – Best Scientific Content: How old is too old: p16 testing on Oropharyngeal biopsies.

Preface

An article (of the non-academic, non-peer reviewed, and glossy magazine variety) sparked my interest in HPV related head and neck cancers. The article reported on the increase in the number of females with HPV related head and neck cancers in the United States and warned of the dangers of oral sex. Armed with only a cytologist's knowledge of cervical HPV, questions were automatically generated. Is there some sort of transformation zone? And could we screen for this disease? During the PhD, these and many other questions were raised and answered, and a whole new set of questions were born.

Thesis structure and format

This thesis aims to assess the burden of HPV positive oropharyngeal cancer in New Zealand, and investigate diagnostic methods and possibilities for early detection in a pre-vaccine population. This will be undertaken by addressing each of the following specific aims:

1. To determine the prevalence and genotypes associated with HPV positive oropharyngeal cancer in New Zealand (Chapter 2).
2. To compare HPV detection using a range of biomarkers in oropharyngeal cancer biopsies with variable p16 results (Chapter 3).
3. To describe the demographics and exposures of a pre-vaccine population of oropharyngeal cancer patients (Chapter 4).
4. To use brushings from conscious oropharyngeal cancer patients to investigate the detection of HPV 16, and cytological abnormalities (Chapter 5).

This thesis is written as a series of six interrelated Chapters, one of which (Chapter 2) has been published in a peer reviewed journal (Lucas-Roxburgh *et al*, 2017). Each Chapter is presented in the format of a manuscript for a peer reviewed journal and as a consequence there is some repetition between Chapters. A literature review (Chapter 1) introduces the thesis topic by discussing human papillomavirus and its role in human disease with a focus on oropharyngeal cancer. The thesis concludes with a

general discussion (Chapter 6) which summarizes the results of the research and puts these into context. The relevance of the findings and future directions for research are also described.

All references are listed at the end of the thesis in order to minimise repetition. All cited literature uses the format of PLoS ONE.

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