Liquid biopsy for Head and Neck Cancers
Arutha Kulasinghe 1, Liz Kenny 2, Chris Perry 2, Majid Warkiani 4, Lidiia Jovanovic 5, Tony Blick 1, Ken O’Byrne 6, Jean-Paul Thierry 7, Ian Vela 5,8, Erik Thompson 1, Colleen Nelson 3, Chamindie Punyadeera 3

1) The School of Biomedical Sciences, Institute of Health and Biomedical Innovation, Queensland University of Technology, Kelvin Grove, Qld, Australia.
2) School of Medicine, University of Queensland; Royal Brisbane and Women’s Hospital; Central Integrative Cancer Service, Queensland Health, Qld, Australia.
3) Department of Otorhinolaryngology, Princess Alexandra Hospital, Woolloongabba, Qld, Australia.
4) School of Mechanical and Manufacturing Engineering, Australian Centre for Non-Medicine, University of New South Wales, Sydney, Australia.
5) Australian Prostate Cancer Research Centre Queensland (IPRP) / Queensland University of Technology, Transitional Research Institute, Woolloongabba, Qld, Australia.
6) Translational Cell Imaging Queensland, Institute of Health and Biomedical Innovation, Queensland University of Technology, Qld, Australia.
7) Department of Biochemistry, Yong Loo Lin School of Medicine, National University of Singapore, Singapore.
8) Department of Urology, Princess Alexandra Hospital, Woolloongabba, Qld, Australia.

Head and Neck Cancer (HNC): Circulating Tumour Cells

* 7th most common cancer, 900 000 new cases, 300 000 deaths (1)
* Less than 50% survive beyond 5 years
* Metastatic disease is responsible for 88% of HNSCC patient deaths within 12 months of diagnosis
* Tumour cells are shed by primary and metastatic cancers.
* Circulating tumour cells are a hallmark of invasive cancer cells and key to metastasis.

Advantages of blood to determine tumour burden
- Minimally invasive blood test vs multiple tumour biopsies.
- Serial sampling (intratumour heterogeneity & tumour evolution).
- Real time monitoring (metastatic progression & treatment response).

Aims
1. Compare CTC enrichment platforms (CellSearch®, ScreenCell®, RosetteSep™, Miltenyi Beads®, Microfluidic Technologies)
2. Characterize patient CTCs (IHC, Immunofluorescence, DNA FISH)
3. Expand patient CTCs ex vivo in MSK (2D) media and Happy Cell (3D)
4. Perform drug sensitivity testing on cultured CTCs
5. Develop a single CTC picking strategy for sequencing

Materials & Methods

![Image](https://example.com/image1)

**Figure 1.** Patient CTCs captured using: (A) CellSearch® (18.6%), (B) ScreenCell® (46.4%), and (C) RosetteSep™ (64.0%) enrichment platforms.

**Figure 2.** Short term ex vivo culture (2D/3D).

**Figure 3a.** CTC count at baseline vs short term culture success samples, P < 0.0002

**Figure 3b.** Correlation between HPV status and short term culture success, P = 0.007

**Figure 4.** Suspicious HNC patient lung lesions.

**Discussion**

- In a paired HNC patient cohort, CTCs were detected in 8/43 (18.6%) by CellSearch®, 13/28 (46.4%) by ScreenCell® and 16/25 (64.0%) by RosetteSep™ (including CTC clusters). Patients were clinically and radiographically M0. In a few patients, suspicious lesions and metastasis were found in the lungs after 6 months (Figure 4).
- Low numbers of CTCs remains a bottleneck in the field of HNC.
- Ex-vivo culture allows for the expansion of CTCs in the short term in defined MSK media + Happy Cell (2D/3D formats)
- Short term CTC cultures were successfully generated in 725 HNC patients (57% of these cultures were from HPV-positive patients). Cultures remained more viable in 3D formats than in 2D (63 days vs 50 days).
- Blood samples with higher CTC counts had a higher success rate of culture (p = 0.0002; Mann-Whitney test, Figure 3a), as did those from HPV+ patients (p = 0.007; Fisher’s exact test, Figure 3b)
- There are currently no methods to predict which patients with a higher disease burden will develop metastases. The ability to do so would lend itself to escalation at diagnosis.

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References