Institution:
University of Cambridge

Unit of Assessment:
UoA1

Title of case study:
Minichromosome maintenance proteins as biomarkers for improving the early detection of common cancers

1. Summary of the impact (indicative maximum 100 words)

An antibody screening test for early detection of cancer was developed in the laboratories of Prof Nick Coleman (Pathology) and Prof Ron Laskey (Zoology; UoA5) and Patent applications arising from their research were filed by Cancer Research Technology (CRT) and licensed to multiple diagnostic companies, including Becton Dickinson (BD). The BD ProEx^{TM} C reagent is in use internationally, including for the triage of cervical smears and biopsies showing 'borderline' abnormalities (~5-7% of cervical smears in developed countries). Additional licensing deals have been negotiated for screening in a range of other cancers, including bladder, pancreas and prostate. The licences have generated in excess of £800K for CRT and the University to date.

2. Underpinning research (indicative maximum 500 words)

Nick Coleman is in the Department of Pathology (from 1992, Associate Senior Lecturer 2001-2011, Professor of Molecular Pathology 2011-present and also previously Programme Leader in the Medical Research Council Cancer Cell Unit, MRC CCU).

Ron Laskey was Honorary Director of the MRC CCU in Cambridge, from its commencement in 2001 to his retirement in 2010. Throughout this period he remained Charles Darwin Professor of Embryology in Cambridge University Department of Zoology. He is now Emeritus Professor in the Dept. Zoology.

Laskey has a long-standing interest in the control of DNA replication. In 1994, he showed that MCM (Mini-Chromosome Maintenance) proteins coupled DNA synthesis to the cell cycle, by forming an active complex in the G1 phase of the cell cycle, which was dissociated in the G2 phase. Laskey's group raised polyclonal and monoclonal antibodies against human MCM proteins and showed that they are absent from non-proliferating cells[1].

In 1997 and 1998, Coleman collaborated with Laskey in a study of cell lines and human tissue sections to show that MCMs were abundant in nuclei throughout the cell cycle, but were lost following cell cycle exit into differentiation, quiescence or senescence[2]. They concluded that MCMs would make excellent biomarkers of the abnormal cell proliferation that characterises malignancy and pre-malignancy.

In 1999, the two groups used immunohistochemistry to demonstrate that, whereas MCMs were confined to the basal cells of normal stratified epithelia, MCMs were expressed in the full thickness of equivalent epithelia showing malignant or pre-malignant changes. This observation applied to most common cancers (e.g. cervix, large bowel, lung, bladder, etc.)[3].

Importantly, many cancer-screening tests use cells sampled from epithelial surfaces (e.g. in cervical smears, stool, sputum, urine, etc.). As MCM proteins were present in the surface cells of cancers/pre-cancers, but absent from the surface cells of normal epithelia, they represented biomarkers with the potential to identify cancer/pre-cancer cells in screening samples[3]. Laskey and Coleman hypothesized that an objective assay based on biomarkers would offer improved accuracy, throughput and affordability for many of the tests used to screen for common cancers. In all tumours tested, MCM proteins were significantly more abundant at the epithelial surface than other markers of cycling cells, such as Ki-67 and PCNA[2,3].

Between 1997 and 2003, the two groups tested whether MCMs could accurately detect malignant/pre-malignant cells in cervical smears[4]. In a clinical evaluation study of over 1,500 women, MCM testing showed very high (>95%) sensitivity for cancer and high-grade pre-cancer, at good levels of specificity (>90%), and detected several cases that were missed by conventional testing.

In 2007, Coleman and Laskey collaborated with Professor Geeta Mukherjee (Kidwai Institute, Bangalore, India) in an immunocytochemical study, which showed that MCM detection was also suitable for cervical screening in developing countries, as a low-cost, objective approach that substantially increased accuracy and reduced time, expertise and costs required for slide assessment[5].

From 2001, the two groups also collaborated to assess the suitability of MCM testing for other common cancers. For bowel cancer screening, they studied colonocytes retrieved from the surface of stool, in a 'proof-of-principle' study. MCM testing distinguished between normal and malignant cells, with positive staining in 37/40 cancers, including all nine early-stage tumours, but in none of 25 control subjects[6]. A novel method for retrieving stool-derived mucus, developed between 2005 and 2009, improved cell yield over 30-fold.

Between 2003 and 2008 the two groups also determined the performance of MCM testing in detecting lung cancer in sputum, using over 800 samples from patients referred for investigation of possible lung cancer. MCMs provided similar sensitivity for detecting lung cancer as a screening check by consultant pathologists (27-31%) and offered the advantage of automated detection.

3. References to the research (indicative maximum of six references)

1. Madine MA, Khoo C-Y, Mills AD and Laskey RA (1995). MCM3 complex required for cell cycle regulation of DNA replication in vertebrate cells. *Nature* 375*: 421-424* doi:10.1038/375421a0
2. Gonzalez MA, Tachibana KK, Laskey RA, Coleman N (2005) Control of DNA replication and its potential clinical exploitation. *Nature Reviews Cancer* 5:135-41 doi:10.1038/nrc1548
3. Freeman A, Morris LS, Mills AD, Stoeber K, Laskey RA, Williams GH, Coleman N (1999) The minichromosome maintenance proteins as biological markers of dysplasia and malignancy. *Clinical Cancer Research* 5: 2121-2132
4. Williams GH, Romanowski P, Morris L, Madine M, Mills AD, Stoeber K, Marr J, Laskey RA, Coleman N (1998) Improved cervical smear assessment using antibodies against proteins that regulate DNA replication. *Proceedings of the National Academy of Sciences USA* 95: 14932-14937
5. Mukherjee G, Muralidhar B, Bafna UD, Laskey RA, Coleman N (2007) MCM immunocytochemistry as a first line cervical screening test in developing countries: a prospective cohort study in a regional cancer centre in India. *British Journal of Cancer* 96:1107-11 doi:10.1038/sj.bjc.6603679
6. Davies RJ, Freeman A, Morris LS, Bingham S, Dilworth S, Scott IS, Laskey RA, Miller R, Coleman N (2002) Analysis of minichromosome maintenance proteins as a novel method for detecting colorectal cancer in stool. *Lancet* 359 1917-19 doi: 10.1016/S0140-6736(02)08739-1

Grant support

The MRC CCU (directed by Ron Laskey 2001 -2010) has received over £3M in funding each year since 2001. From 2001 to the present, Coleman and Laskey have been funded by Programme Grants within the MRC CCU and from Cancer Research UK, with a combined value of over £6.2M. The current funding round extends to November 2016. The principal grants are:

- 'Translational approaches to improving cancer screening and diagnosis' (Coleman) Medical Research Council Programme Grants, within MRC Cancer Cell Unit: (2006-2011) £1,190,722; (2001-2006) £1,230,385.
- 'Viral and host mechanisms in cervical carcinogenesis' (Coleman) Cancer Research UK Programme Grant (2011-16) £1,125,002
- 'Eukaryotic DNA replication and novel cancer markers' (jointly Laskey/Coleman) Cancer Research UK Programme Grants: (2006-2011) £1,431,085; (2001-6) £1,245,700

Intellectual property

The use of MCMs as biomarkers to detect cancer/pre-cancer is protected by multiple international patents, granted to Cancer Research Technology on behalf of Cancer Research UK, and Cambridge University. The principal patent is: 'Determination of Cellular Growth Abnormality', reference: US Patent Office 6,303,323 (16/10/2001) [and divisionals]; World Intellectual Property Organisation 1999/021014 (29/04/1999); Canada 2305872 (29/04/1999); China 98812478 (31/01/01); European Patent Office EP1025444 (09/08/00).

4. Details of the impact (indicative maximum 750 words)

The MCM technology and associated intellectual property has had impacts on commerce and on healthcare systems internationally. It has also improved public understanding of the importance of early cancer detection.

Impacts on commerce: industry has invested in research and development, a new product is in production

In 2004, the MCM technology was licensed by Cancer Research Technology to TriPath Imaging (U.S.) in the field of cervical cancer. In 2007, TriPath and its associated licences were bought by Becton Dickinson (BD), who developed the BD ProEx™ C reagent, based on antibodies against MCM2 and DNA-topoisomerase 2α. This has been in widespread use internationally since 2008, particularly in USA and Canada, as well as in European centres (most notably Scandinavia and Southern Europe). [Text removed for publication]

Impacts on commerce: highly skilled people have taken up specialist roles in companies, jobs have been created

The research of Laskey and Coleman led to the creation of the MRC Cancer Cell Unit (CCU), a new MRC Unit, in 2001, with Laskey as founding director. The purpose of the Unit was to build on and extend translational cancer research, particularly MCM-based cancer detection. The MRC CCU receives over £3M in funding p.a. and continues to provide over 100 new jobs in the UK. [Text removed for publication] In addition, Becton Dickinson has invested heavily in developing the BD ProEx™ C reagent (based on MCM detection), creating new highly-skilled jobs (number withheld by BD) in the USA.

Impacts on healthcare: a new diagnostic has been adopted; disease prevention has been enhanced

An important use of the BD ProEx™ C reagent is for triage of cervical smears and biopsies showing 'borderline' changes, referred to as atypical squamous cells of uncertain significance (ASCUS). These abnormalities are seen in 5-7% of cervical smears in developed countries, and provide a major clinical challenge, as they include cases of pre-cancer, as well as non-neoplastic processes such as repair and reaction to inflammation. BD ProEx™ C significantly improves the accuracy of pre-cancer detection in cervical smears in this sample group, reducing patient over-investigation and potentially therefore overall screening costs[7].

Since 2010, several independent groups in Europe and USA have evaluated the BD ProEx™ C reagent in combination with human papillomavirus (HPV) testing, as a more accurate and cost-effective replacement for cervical Papanicolaou (Pap) screening. There is an important clinical need for a biomarker-based approach to primary cervical screening, as the Pap test is based on subjective interpretation and a single test has a sensitivity for cancer/pre-cancer of only ~50%. A recent major study compared eight primary cervical screening strategies and concluded that the optimal combination was HPV testing followed by triage using the BD ProEx™ C reagent[8]; it had the highest level of accuracy and reduced the number of patient procedures required. As a result, BD pre-adapted its new screening machines worldwide to run BD ProEx™ C in combination with HPV testing (BD Totalys system).

To date, over 30 publications from independent research groups have demonstrated the value of the BD ProEx™ C reagent, in a variety of clinical settings. The product has also been used by histopathology services across the NHS in the UK[10]. It is not yet possible to state with accuracy the number of patients who have benefitted from BD ProEx™ C in USA/Canada and Europe since 2008, as BD is still in the process of commercialising the products and providing local support to cytology laboratories worldwide.

To date, licenses based on the MCM technology have generated income in excess of £800K for Cancer Research Technology on behalf of Cancer Research UK, and the University of Cambridge[11].

Impacts on health and welfare: public awareness of a health benefit has been raised

The MCM work has featured prominently in the national press, including several articles since 2008[12,13]. Coleman and Laskey have given numerous invited talks on MCM testing, to both specialist and lay audiences, thereby improving public and professional understanding of the importance of early cancer detection[14].

Due to the impacts of this work, both Laskey and Coleman have received major scientific awards. In 2009, Laskey was awarded the Royal Medal of the Royal Society for 'his pivotal contributions to our understanding of the control of DNA replication and nuclear protein transport, which has led to

a novel screening method for cancer diagnosis'. In 2010, Coleman received the Goudie Medal, The Pathological Society of Great Britain and Ireland, which is awarded 'to a distinguished active scientist who is making seminal contributions to pathological science'.

5. Sources to corroborate the impact (indicative maximum of 10 references)

7. Badr RE et al, 2008, BD ProEx C: a sensitive and specific marker of HPV-associated squamous lesions of the cervix. *Am J Surg Pathol.* 32:899-906
8. Depuydt et al, 2011, BD-ProExC as adjunct molecular marker for improved detection of CIN2+ after HPV primary screening. *Cancer Epidemiol Biomarkers Prev.* 20:628-37
9. BD ProEx™ C reagent website: www.bd.com/tripath/products/proexc/index.asp
10. Histopathology Reporting in Cervical Screening – an integrated approach. NHS Cervical Screening Programme. NHSCSP Publication Number 10 (second edition). September 2012 www.cancerscreening.nhs.uk/cervical/publications/nhscsp10.pdf
11. Personal communication, Business Management Director, Cancer Research Technology
12. 24 October 2008, Virtual Medical Centre, 'A new test to prevent anal cancer'. http://www.virtualmedicalcentre.com/news/a-new-smear-test-to-prevent-anal-cancer/12707
13. 10 November 2008, Pink News, 'Thousands of lives could be saved by new anal cancer test'. http://www.pinknews.co.uk/2008/11/10/thousands-of-lives-could-be-saved-by-new-anal-cancer-test/
14. Talks by Coleman include European Cancer Organisation and European Society for Medical Oncology [ECCO15:ESMO34] Teaching lecture jointly with Laskey (Berlin 2009); Goudie Medal Lecture, The Pathological Society of GB and Ireland (London 2010); Plenary lecture Society for General Microbiology Human Papillomavirus UK meeting, (Lake District, 2012)