Institution: University of Sussex

Unit of Assessment: UoA 05 Biological Sciences

Title of case study: Diagnosis of genetic diseases with immune or neurological dysfunction

1. Summary of the impact

The Caldecott/Jeggo/O'Driscoll laboratories have identified human genetic diseases that are caused by defects in genes involved in DNA strand-break repair. Many of these diseases are associated with neurological pathologies such as cerebellar ataxia (resulting in poor balance, movement control, and patients often being wheelchair bound), microcephaly (smaller-than-normal head circumference), and developmental delay. The Caldecott/Jeggo/O'Driscoll laboratories have engaged in identifying/diagnosing patients with such diseases as a service to clinicians/clinical geneticists in the UK National Health Service (NHS) and worldwide. Since 2008, these laboratories have identified the underlying genetic defect in more than 150 patients with a range of hereditary DNA damage-related disorders. In particular, these laboratories have diagnosed patients with genetic defects in the DNA damage response genes *Lig4*, *NHEJ1*-XLF, *DCLRE1C*-Artemis, *PRKDC*-*DNA-PKcs*, *PCNT, ORC1*, *ATRIP, ATR,* and *TDP2*. These diagnoses benefit both the clinical geneticist and the patient; identifying not only the cause of the patient's disease but also enabling better disease management. For example, if not first diagnosed, standard chemotherapeutic regimes can be fatal in cancer patients who harbour homozygous TDP2 mutations, and standard conditioning regimes used during bone-marrow transplantation can be fatal in LIG4 Syndrome patients. These diagnoses can therefore translate into increased patient survival.

2. Underpinning research

Neurological disease is crippling and often life-threatening, and can result from a variety of hereditary genetic defects. Some of these defects reflect an inability of the affected individuals to process and/or repair DNA damage effectively. As a result of their work into fundamental mechanisms of DNA damage-response and repair, the Caldecott/Jeggo/O'Driscoll laboratories have identified a number of human genes that, if mutated, result in neurodegeneration and/or neurodevelopmental defects.

For example, the Caldecott laboratory has identified *TDP1*, *APTX*, *APLF* and *TDP2* as novel human genes involved in the repair of DNA single- and double-strand breaks, and has established that mutations in three of these cause neurodegenerative disease [see Section 3, R1–R4]. Intriguingly, three of these four genes are required for a single stage of DNA repair, in which the ends of DNA breaks are processed and chemically 'tidied' in readiness for their rejoining.

Similarly, the O'Driscoll and Jeggo laboratories have identified genetic defects that cause microcephaly (in which affected individuals possess a smaller-than-normal head circumference) and dwarfism, including Seckel Syndrome (due to mutations in the genes *ATR* and *ATRIP*), Microcephalic Osteodysplasic Primordial Dwarfism type-II (due to mutations in the gene *PCNT*) and Meier-Gorlin Syndrome (due to mutations in *ORC1*) [R5–R7]. These genes are normally involved in regulating cell duplication and division, and so result in growth defects if mutated.

In addition, the Jeggo laboratory has identified the genetic basis of disorders associated with combined immunodeficiency and with elevated sensitivity to ionising radiation (RS-SCID), such as LIG4 Syndrome (due to mutations in *Lig4*) [R8].

These genes are central components of the biochemical pathway by which chromosomal double-strand breaks are repaired; a process that is critical to the development of the immune system and

to resistance to X-rays and some types of chemotherapy. As a direct result of these fundamental discoveries in the field of DNA damage-response and repair, these laboratories are now frequently approached by clinical geneticists in the UK and abroad for advice and for diagnostic input, with the aim of identifying the underlying genetic cause of hereditary disease in which specific DNA damage-response mechanisms are defective.

Key researchers

- Professor Keith Caldecott, Deputy Director of the Genome Damage and Stability Centre, University of Sussex, 2002–present.
- Dr Mark O'Driscoll, Principal Investigator, Genome Damage and Stability Centre, University of Sussex, 1999–present.
- Professor Penelope Jeggo, Principal Investigator, Genome Damage and Stability Centre, University of Sussex, 1988–present.

3. References to the research

R1 Ahel, I., Rass, U., El-Khamisy, S.F., Katyal, S., Clements, P.M., McKinnon, P.J., Caldecott, K.W. and West, S.C. (2006) 'The neurodegenerative disease protein aprataxin resolves abortive DNA ligation intermediates', *Nature*, 443(7112): 713–16.

R2 Cortes Ledesma, F., El-Khamisy, S.F., Zuma, M.C., Osborn, K. and Caldecott, K.W. (2009) 'A human 5'-tyrosyl DNA phosphodiesterase that repairs topoisomerase-mediated DNA damage', *Nature*, 461(7264): 674–8.

R3 El-Khamisy, S.F., Saifi Gulam, M., Weinfeld, M., Helleday, T., Lupski., J.R. and Caldecott, K.W. (2005) 'Defective DNA single-strand break repair in spinocerebellar ataxia with axonal neuropathy-1', *Nature*, 434(7029): 108–13.

R4 O'Driscoll, M., Ruiz-Perez, V.L., Woods, C.G., Jeggo, P.A. and Goodship, J.A. (2003) 'A splicing mutation affecting expression of ataxia-telangiectasia and Rad3-related protein (ATR) results in Seckel syndrome', *Nature Genetics*, 33(4): 497–501.

R5 Iles, N., Rulten, S., El-Khamisy, S.F. and Caldecott, K.W. (2007) 'APLF (C2orf13) is a novel human protein involved in the cellular response to chromosomal DNA strand breaks', *Molecular Cellular Biology*, 27(10): 3793–803.

R6 Griffith, E., Walker, S., Martin, C., Vagnarelli, P., Stiff, T., Vernay, B., Al Sanna, N., Saggar, A., Hamel, B., Earnshaw, W.C., Jeggo, P.A., Jackson, A.P. and O'Driscoll, M. (2008) 'Mutations in Pericentrin cause Seckel syndrome with defective ATR-dependent DNA damage signalling', *Nature Genetics*, 40(2): 232–6.

R7 Bicknell, L.S., Walker, S., Klingseisen, A., Stiff, T., Leitch, A., Kerzendorfer, C., Martin, C.A., Yeyati, P., Al Sanna, N., Bober. M., Johnson, D., Wise, C., Jackson, A.P., O'Driscoll, M. and Jeggo, P.A. (2011) 'Mutations in ORC1, encoding the largest subunit of the origin recognition complex, cause microcephalic primordial dwarfism resembling Meier-Gorlin syndrome', *Nature Genetics,* 43(4): 350–5.

R8 O'Driscoll, M., Cerosaletti, K.M., Girard, P.-M., Dai, Y., Stumm, M., Kysela, B., Hirsch, B., Gennery, A., Palmer, S.E., Seidel, J., Gatti, R.A., Varon, R., Oettinger, M.A., Neitzel, H., Jeggo, P.A. and Concannon, P. (2001) 'DNA Ligase IV mutations identified in patients exhibiting development delay and immunodeficiency', *Molecular Cell*, 8(6): 1175–85.

Outputs can be supplied by the University on request.

Grants supporting the research or awarded based on the research

- Caldecott: Molecular characterisation of single-strand break repair and related responses and their role in neuroprotection. MRC Programme, 1 March 2007–28 February 2012; £1,800,000.
- O'Driscoll: How aberrant ataxia telangiestasia and rad3-related (ATR) pathway function affects genomic stability. CR-UK Senior Fellowship, 1 July 2007–31 June 2013; £1,600,000.
- Jeggo: DNA damage responses in mammalian cells and their contribution to human health disorders. MRC Programme, 1 October 2006–30 September 2011; £1,498,355.

4. Details of the impact

As indicated above, the Caldecott/Jeggo/O'Driscoll laboratories have identified a number of human genes which, if mutated, result in neurological disease. As a result of these discoveries, blood and tissue samples from patients with the relevant disease pathology are routinely tested for mutations in these and closely related genes by the Caldecott/Jeggo/O'Driscoll laboratories, at the request of clinicians in hospitals within the UK National Health Service (NHS) and Europe (e.g. Great Ormond Street, Newcastle General Hospital, Bristol Genetics Laboratory, Nijmegen Medical Centre), or within external diagnostic laboratories within the UK and Europe directly (e.g. Birmingham).

For example, in 2011/2012, based on its identification that APLF and TDP2 are novel human DNA repair genes, the Caldecott laboratory conducted genetic/biochemical analyses for defects in APLF, TDP2, and XRCC1-dependent DNA damage responses in blood samples and fibroblasts from patients with neurological/developmental delay, at the request of NHS clinicians in Bristol (St Michael's Hospital) [see Section 5, C1] and London (Great Ormond Street) [C2], and by clinical geneticists in Nijmegen (Medical Centre). These diagnoses uncovered mutations in TDP2 as a cause of epilepsy and spinocerebellar ataxia in four patients [C3].

In like manner, during the current REF period, the Jeggo laboratory has conducted similar diagnostic testing at the request of clinicians from a variety of hospitals in the UK and abroad, including Great Ormond Street, identifying 11 individuals with RS-SCID out of 140 who were screened [C4, C5]. Some of these hospitals have recently established sequencing procedures/ assays based on guidance from the Jeggo laboratory to aid diagnosis, and the Jeggo laboratory continues to advise and carry out a diagnostic service for those patients in whom sequencing fails to reveal a mutation or who display a pathology that is suggestive of a novel or unusual DNA damage-response-related genetic defect [C5]

In addition, these diagnostic tests are now also conducted externally at a variety of medical/clinical genetics centres worldwide, independent of our laboratories, thereby expanding the reach of our impact. For example, as a direct result of novel genetic defects identified by the O'Driscoll/Jeggo laboratories, multiple clinical genetics centres in the UK (Birmingham, Great Ormond Street, Newcastle General Hospital), in North America/Canada (University of Chicago Genetics Services, Greenwood Genetics Centre South Carolina, Baylor College of Medicine Texas, Nemours Children's Hospital Delaware, and Children's Hospital of Eastern Ontario, Canada) and in Europe (Nijmegen Clinical Genetics Center, Dept of Human Genetics Belgium, Neckar-Enfants Malades Hospital Paris, Institut für Humangenetik Erlangen and Institut für Humangenetik Freiburg) provide molecular diagnostic analyses of causative genes in a clinical genetics context, and are registered on the GeneTests service [C6].

As a result of this work, since 2008 the Caldecott/Jeggo/O'Driscoll laboratories have identified the underlying genetic defect in more than 150 patients with a range of hereditary DNA damage-related disorders. In terms of beneficiaries of this impact, the results of diagnostic tests are provided to the relevant clinicians in the form of written reports, which are employed to inform additional diagnostic tests and patient care and management. These diagnoses thus benefit both

the clinical geneticist and the patient; identifying not only the cause of the patient's disease but also enabling better disease management. For example, if not first diagnosed, standard chemotherapeutic regimes can be fatal in cancer patients who harbour homozygous TDP2 mutations, and standard conditioning regimes used during bone-marrow transplantation can be fatal in LIG4 Syndrome patients. These diagnoses can therefore translate into increased patient survival.

5. Sources to corroborate the impact

Corroborating evidence for Caldecott/O'Driscoll/Jeggo: letters from clinical geneticists confirming diagnoses and impact on patient healthcare.

C1 Email from Specialist Registrar (St Michael Hospital).

C2 Email from clinician (Great Ormond Street).

C3 Email from clinical geneticist (Nijmegen Medical Centre).

C4 Emails from Great Ormond Street, Manchester Children's Hospital, the Beatson Institute and Gleneagles Medical Centre, Singapore.

C5 Statement from clinician, Peter MacCallum Cancer Centre, Melbourne, Australia.

C6 http://www.ncbi.nlm.nih.gov/sites/GeneTests/.