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RACP Foundation Research Awards

FINAL REPORT

Project / Program Title		Massively parallel sequencing for the detection of uniparental disomy in the peripheral blood of patients with myeloproliferative neoplasms.
Name		Dr David Ross
Award Received		2015 RACP GlaxoSmithKline Research Establishment Fellowship
Report Date		8 August 2016
Chief Investigator / Supervisor		David Ross Co-investigators: Prof Richard D'Andrea, Prof Hamish Scott, Dr Chris Hahn
Administering Institution		SA Pathology
Funding Period	Start Date:	15 February 2015
	Finish Date:	15 March 2016

PROJECT SUMMARY

Polycythaemia vera (PV) is a myeloproliferative neoplasm (MPN), a type of blood cancer that results in increased production of blood cells and an increase in the risk of bleeding and clotting complications. Some PV patients later progress to myelofibrosis or acute leukaemia. Most PV patients are aged over 60 at diagnosis.

Most MPN patients have an acquired mutation in the JAK2 gene. JAK2 is an enzyme that plays a key role in controlling the production of red cells and platelets. The mutated enzyme has increased activity resulting in increased cell numbers. One of the unanswered questions in MPN research is why the same mutation occurs in different MPNs, but with different clinical features in PV, myelofibrosis, and essential thrombocythaemia.

An early observation after the discovery of the JAK2 mutation was that around 80% of PV patients have two copies of the mutated gene. This is thought to result from abnormal copying of the mutant gene during cell division, giving a growth advantage to cells with two copies. We are interested to study the differences between PV patients with one copy and patients with two copies of the mutant gene. In order to do that we first need a rapid screening test so that we can tell which patients fall into which group.

The RACP GlaxoSmithKline Research Establishment Grant provided me with funding to develop a Next Generation Sequencing test to measure the proportion of cells in a sample that contain two copies of the mutated JAK2 gene. We are currently using this test to screen a large number of PV patients so that we can better understand the genetic diversity of the disease.

PROJECT AIMS / OBJECTIVES

The aim of this project was to develop and validate a PCR-based method for the detection of LOH in MPN.

A research student worked on this project for 6 months under my supervision, and a working method was developed using massively parallel sequencing of hundreds of amp/icons, each containing a chromosome 9 SNP. Additional work is ongoing.

SIGNIFICANCE AND OUTCOMES

The research has established the feasibility of a novel method for the detection of UPD in MPN, but with broader applicability in other neoplasms. In MPN biology it will greatly simplify the study of JAK2 clonal progression and clonal expansion, particularly in relation to what drives the phenotypic distinction between ET and PV.

In clinical MPN management the detection of JAK2 LOH by MPS may have diagnostic and prognostic value, a question which was explored in a paper that was recently accepted for publication (see below: Impact Factor4.411). We compared UPD in patients with ET, masked PV, and overt PV. The MPS method was not used in that paper, but half of those patients were the control group for the MPS project. For those patients we have results to enable us to compare MPS with the traditional method of sequencing erythroid colonies.

Method optimization is ongoing and when complete we will screen all of our PV patients for UPD, and expect a further publication to arise from that work.

PUBLICATIONS / PRESENTATIONS

Tiong IS, Casolari DA, Nguyen T, Van Velzen MJM, Ambler K, D'Andrea RJ, and Ross OM. Masked polycythaemia vera is genetically intermediate between JAK2V617F mutated essential thrombocythaemia and overt polycythaemia vera Blood Cancer Journal 2016 accepted for publication July 11.

Abstract (same authors and title) submitted to Haematology Society of Australia and New Zealand Annual Meeting 2016.