PROJECT SYNOPSIS, School of Life Sciences
Director of studies: Dr Ian Harmer

Project title: Molecular characterization of antibodies associated with heparin-induced thrombocytopenia. Is there a link between epitope specificity and clinical outcome?

Background and synopsis

Heparin induced thrombocytopaenia (HIT) is a severe adverse effect of heparin treatment, caused by platelet activating antibodies, usually recognising complexes of platelet factor 4 (PF4) and heparin. Up to 8% of patients treated with heparin develop HIT associated antibodies. 1-5% of those exposed for 5+ days develop HIT with thrombocytopenia and 1/3 of these suffer from venous and/or arterial thrombosis which can be fatal.

We do not yet understand why they arise in only a limited number of patients or why only some of these develop life threatening symptoms. Treatment of patients would be greatly improved if it were possible to predict which of those with HIT antibodies were likely to develop thrombocytopenia. Characterising HIT antibodies present in individuals who do and do not have thrombocytopenia will enhance understanding of the condition and offers the opportunity to develop diagnostic tests to identify those individuals at high risk.

Although HIT develops rapidly on first treatment with heparin, immune response and antibodies are characteristic of prior exposure to the antigen leading to the suggestion that the antibodies might be derived from B1- cells in a T-independent manner. HIT antibodies bind 2 or 3 distinct (unidentified) neoepitopes exposed on PF4 when it binds heparin, but not in all patients are these epitopes recognised. HIT antibodies do not recognise the heparin binding site of PF4. A murine monoclonal able to significantly inhibit binding in one third of patients has been generated but no human antibodies to it are yet available.

Antibodies representative of serum populations will be isolated by phage display using the heparin-PF4 complex. Their fine specificity will be determined and sequencing will identify any patterns in V-gene use or CDR3 structure, associated with specificity or B-cell population. Isolated antibodies will be used to determine the precise specificity of a large number of patient antisera in order to identify any correlation between epitope(s) recognised and clinical outcome. A positive correlation would offer the potential of development of a clinically and commercially important diagnostic test kit.

The student will take part in the School research training programme, gain knowledge of antibody engineering and molecular biology techniques, learn a range of immunological, protein expression and purification techniques, and gain experience of interaction between an academic research laboratory and a routine clinical diagnostic laboratory.

This project is a collaboration with the Royal Brompton Hospital.

Supervisory Team and Research Environment

Supervisor Name	Role (DoS, 2nd Supervisor, 3rd Supervisor)	No. of successful PhD/ MPhil supervisions	Current student load for 2010/11 (FTE)	School (for cross School projects)
Dr Ian Harmer	DoS	3	0	
Dr Angray Kang	2nd	4	2	

Recent publications by supervisors relevant to the project:

Harmer IJ, Jennings NS, Campbell K, Stafford P, Smith GA, Metcalfe P, Benton MA, Marsh JC and Ouwehand WH (2007) Transfusion **47** 499-510.

Jennings NS, **Harmer IJ**, Campbell K, Stafford P, Smith GA, Metcalfe P, Benton MA, Marsh JC and Ouwehand WH (2006) J. Immunol. Methods **316** 75-83.

Chappel JA, Rogers WO, Hoffman SL and **Kang AS**. (2004) Malar J. **3** 28-40

Informal enquiries: i.harmer@westminster.ac.uk

http://www.westminster.ac.uk/schools/science/research/research-groups/applied-biotechnology
http://www.heparininducedthrombocytopenia.com/index.asp
http://circ.ahajournals.org/cgi/content/full/114/8/e355
http://bloodjournal.hematologylibrary.org/cgi/content/full/91/3/916

Ethical issues

Molecular characterization of antibodies associated with heparin-induced thrombocytopenia

Blood samples will be provided by the Royal Brompton Hospital. Ethical approval will be sought through their local procedures and from the University of Westminster. All samples will be anonymous although it will be necessary to be able to link to patient records through a third party in order to correlate results with patient outcome.