The Sophie Cameron Trust Ph.D. Student: Priyamvada Dua

Priya was educated in New Delhi, India, until 2004, then took her B.Sc. in biomedical sciences at Newcastle upon Tyne University, followed by an M.Sc. in integrative neuroscience at Imperial College, London. In 2008, she was awarded the Sophie Cameron Trust Ph.D. Studentship under Professor Gavin Giovannoni at Queen Mary University, London.

EXPERIENCE

June-August 2005 **Professor J.E.Hesketh (University of Newcastle Upon Tyne) And The Wellcome Trust Vacation Scholarship**

- **Project title**: To study effects of knock-down of components of the mRNA localization apparatus on mouse embryonic stem (ES) cell differentiation.

- **Techniques learnt**:

Cell Culture: All experiments required IEC 6 cells so a major part of my work involved cell culture. I learnt routine methods for cell culture.

Western Blotting: The major analytical method involved in the project was western blotting of cell extracts. So I prepared cell extracts, measured their protein content and analysed them by western blotting using chemiluminescent detection and autoradiography.

Preparation of cell protein extract and Bradford assay for total protein measurement: This was done to prepare 30 microgram protein samples for western blotting

Immunocytochemistry: This was carried out to confirm expression of staufen in IEC6 cells and to show localization of the protein within these cells.

siRNA Transfection: Knockdown of staufen by siRNA was carried out. This required transfection of cells using lipofectamine.

Jan- March 2006 **Dr. Sasha Gartside (University of Newcastle Upon Tyne) BSc 3rd year project**

- **Project title**: Expression of nicotinic receptor subunits in 5-HT and non-5-HT cells of the dorsal raphe nucleus (DRN).

- **Techniques Learnt**:
  Tissue Collection and Fixation: The rat brain was sectioned using a vibratome and a freeze microtome after which the sections were fixed in 4% paraformaldehyde and then cryoprotected in 30% sucrose.

  Ligand Binding: The α7 nicotinic acetylcholine receptor subunit was labelled with fluorescent conjugated α-bungarotoxin.

Immunohistochemistry: The α4 nAChR was labelled using both fluorescent and biotinylated antibody.

Double labelling experiments: Performed for both subunits with tryptophan hydroxylase a marker for 5-HT neurones.

Analysis: Slides were examined using a fluorescence microscope.

March- Sept '07 **Dr. Egle Soilto and Prof. Julia Buckingham**
MSc Project

- **Project Title:** Role of annexin A1 in brain microvascular endothelial cells

- **Techniques used:**
  Cell Culture : The cell lines used in this study were human brain microvascular endotholial cells (HBMEC), mouse brain microvascular endothelial cells (b.End.3), human bone marrow endothelial cels (TrHBMEC), microglia cells (BV-2) and monocytic cells (U937).

  Immunohistochemistry : Performed on brain tissue from mice ANXA1 KO and littermate control treated with LPS (ip).

  FACS : It was carried out to analyse the expression ANXA1 (total and membrane) and the adhesion molecules expressed by the three cell lines.

  Western Blotting : To confirm the expression of annexin A1 in the endothelail and glial cells.

  qRT-PCR : To study the expression of leprecan 1 and protocadherin 6 (housekeeping gene) in the endothelial cell lines

  Cell Adhesion Assay : BV2 cells were co–cultured with endothelial cells stimulted with LPS,IL-1 AND TNF-α and vice versa to measure the percentage of adhesion using ELISA

  Flow Chamber : Was used to measure the adherence of BV2 cells to stimulated endothelial cells under shear stress condition at The William Harvey Institute.