

Professor David M Glover FRS FRSE
Arthur Balfour Professor of Genetics
Director Cancer Research UK
Cell Cycle Genetics Group



Cell Cycle Genetics Research Group



Department of Genetics

To whom it may concern:

I am writing to confirm our wish to continue our productive collaboration with the Laboratory of Mass Spectrometry of IBB PAS which has now been ongoing for some six or so years. Our group studies key regulators of the cell division cycle using *Drosophila* and the mouse as a model. We have a major interest in the organisation of multiprotein complexes at both the kinetochore and the centrosome and it is these areas that we have collaborated with the Warsaw group. Their mass spectrometric tools have been essential for us both in providing routine data about protein-protein interactions and in developing new ways of studying these protein complexes.

Examples of the application of the combined expertise of the two groups are our findings:

- that the centromeric protein CENP-C is critical to establish the mitotic kinetochore for chromosome segregation (Przewlaka, M.R., Venkei, Z., Bolanos-Garcia, V.M., Debski, J., Dadlez, M., and Glover, D.M. (2011) CENP-C is a structural platform for kinetochore assembly *Current Biology* 21: 399-405);
- by our characterisation of the phosphoprotein Endos as a key inhibitor of protein phosphatase 2A to oppose the mitotic-kinase Cdk1 (Rangone, H., Wegel, E., Gatt, M.K., Yeung, E., Flowers, A., Debski, J., Dadlez, M., Janssens, V., Carpenter, A.T.C., and Glover, D.M. (2011) Suppression of Scant Identifies Endos as a Substrate of Greatwall Kinase and a Negative Regulator of Protein Phosphatase 2A in Mitosis **PLoS Genet.** 2011 Aug;7(8):e1002225);
- and our recent demonstration of the sites on Ana2 required to be phosphorylated by Plk4 to initiate centriole duplication (Dzhindzhev, N.S., Tzolovsky, G., Lipinski, Z., Schneider, S., Lattao, R., Fu, J., Debski, J., Dadlez, M., and Glover D.M. (2014) Plk4 phosphorylates Ana2 to trigger Sas6 recruitment and procentriole formation *Current Biology*, October issue).

We would now like to take our studies of protein complexes in the centriole to a new level and apply mass spectrometry to study how its constituent proteins interact. This will utilise methodology successfully developed in Professor Dadlez' laboratory based on measurements of the rates of hydrogen-deuterium exchange on the protein - slow exchanging residues are buried within the structure of individual proteins or mark regions of physical interactions. Thus the proteomics lab will provide us with structural information that we can test using approaches of molecular cell biology. We are successfully using similar approaches to study proteins that constitute the kinetochore. Their application to the centriole has to our knowledge never before been attempted before but our experience with kinetochore proteins indicates that it will be extremely valuable. The proposed research will be of great benefit to both of our laboratories and we will be delighted to participate in it.

A handwritten signature in black ink, appearing to read "D M Glover".

David M. Glover
Cambridge, 12th September 2014

Downing Site
Downing Street
Cambridge CB2 3EH
Telephone: +44 (0)1223 333988 / 333986
Fax: +44 (0)1223 333968
email: d.glover@gen.cam.ac.uk
ajc1001@cam.ac.uk (John Crang PA)