repeats this cycle (self-renewal), the GMC divides once more to produce two terminally differentiated neurons and/or glial cells. During mitosis of the neuroblasts, several molecules have been shown to be absolutely required for asymmetric division to take place. These include molecules which are asymmetrically localised and partitioned into the GMC (including Prospero mRNA and Miranda, Brat and Prospero proteins), and molecules that set up and maintain the asymmetric potential of the cell (including Inscuteable, Par-3/6, Bicaudal D, Egalitarian and Rab6). In order for correct asymmetric cell division to occur, these molecules must interact with each other. However the mechanisms underlying these protein-protein interactions are unknown.

Here we focus on gaining structural insight into the mechanisms that drive asymmetric cell division using X-ray crystallographic techniques to determine high-resolution structures of the proteins involved. We are currently making progress in expressing, purifying and crystallising the proteins Miranda, Prospero and Inscuteable. We have also solved the crystal structure of *Drosophila melanogaster* Rab6 to 1.4 Å resolution. Rab6 is known to bind many effector proteins, including Bicaudal D, and we aim to use this structure to aid determination of Rab6-effector complex structures.

Keywords: cell division, protein-protein interactions

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Structural basis of dephosphorylating activity of PIP substrate in PTPRQ

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The PTPRQ belongs to a receptor-type classical protein tyrosine phosphatase (PTP) family with one catalytic domain in the cytoplasmic region. Unlike other classical PTPs, PTPRQ has de-phosphorylating activities toward phosphatidyl-inositol (PIP) substrates. We successfully cloned, over-expressed, purified and subsequently crystallized the catalytic domain of PTPRQ. Subsequently PTPRQ was crystallized in two forms. PTPRQ crystals belonged to hexagonal space group, $P6_4$ with cell dimensions of $a = b = 78$ Å, $c = 84$ Å, $\alpha = \beta = 90°$, $\gamma = 120°$ diffracted x-ray to 1.6 Å resolution. Structural solution was obtained by molecular replacement method using PTPRO structure. Overall, PTPRQ adopts a typical tertiary fold as other classical PTPs do. However, the disordered "E-loop" of PTPRQ surrounding catalytic core and concomitantly absence of its interactions with the residues in "P-loop" results in flat active site pocket. We propose that this structural feature might facilitate to accommodate the bigger substrates and be suitable for dephosphorylation of PIP.

Keywords: PTP, PIP, E-loop

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Structural investigation of RAP80; A novel BRCA1 interacting protein involved in the mediation of DNA damage repair function

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BRCA1 (Breast Cancer Susceptibility gene 1) is one of the central molecules that play crucial role in DNA damage repair process [1]. BRCA1 recruitment at the site of DNA damage depends on its interacting partners like RAP80 (Receptor Associated Protein 80), CCDC98, MERIT40. RAP80 comprises two tandems Ubiquitin-Interacting Motifs (UIM1, UIM2) at its N-terminus, ABRAXAS Interacting Region (AIR) at the central domain and two zinc-finger containing motifs (ZFD1, ZFD2) at C-terminus [2]. UIM motifs of RAP80 interact with K63 linked polyubiquitin chain(s) on H2AX and thereby assemble the RAP80-BRCA1 complex at the damage site for DNA repair [2]. CCDC98 (ABRAXAS) acts as a bridging molecule to mediate the interaction of RAP80 with BRCA1 [3]. MERIT40 helps in the stabilization of whole complex by favoring the interaction among various members[4].

To dissect the structural diversity, RAP80 functional domains (tandem repeat UIM, AIR) and CCDC98 were sub-cloned in bacterial expression vectors pGEX-kT and pMAL-c2 respectively. Purified recombinant protein(s) were obtained using two step purification involving affinity chromatography followed by gel filtration chromatography. Mass spectrometric analysis confirmed the identity and estimated the correct molecular mass of the purified proteins. Secondary and tertiary structures monitored using Far UV Circular Dichroism and Fluorescence spectroscopy suggested that the recombinant proteins have well folded structural conformation. Crystallization screening of RAP80 tandem UIM1 and UIM2 domain showed nucleation, further optimization is under process. CCDC98 fusion protein with Maltose binding protein tag has been purified and binding analysis using ITC and co-crystallization is under optimization. Comparative study of wild type and mutants of RAP80-BRCA1 complex will be helpful in basic understanding of DNA repair defect and hence the tumorogenesis.

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Cooperative DNA-binding mechanism of Aristaless and Clawless

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Homeodomain is one of the most widely spread superfamily of eukaryotic DNA binding domain that regulates transcriptions of various kinds of genes that are indispensable for development. Some homeodomain proteins bind cooperatively to specific DNA sequences to increase those binding affinities and site specificities for the target DNA. In this report, we examined a cooperativity of two homeodomain proteins, Aristaless (Al) and Clawless (Cll) from *Drosophila melanogaster*. These homeodomain proteins play an important role in *Drosophila* leg development. In the distal-most region, homeobox genes, *al*, *Lim1*, and *cll* are expressed to specify the region, while a pair of *Bar* homeobox genes are expressed in its immediate neighbour (distal tarsus). For the accurate differentiation of these regions, the