

## MAJOR RESEARCH ACHIEVEMENT DURING THE FIRST YEAR OF THE 'RETNET' PROJECT (01/01/2004 – 31/12/2004)

- Fine mapping and cloning of a new gene causing adRP. A family of French origin with autosomal dominant mode of inheritance for retinitis pigmentosa (adRP) having thirty individuals was used for linkage mapping of the disease gene in the human genome.
- The work has focused on developing retinal therapies in animal models. siRNAs targeting Rhodopsin and rds-peripherin, which have attained greater than 70% suppression *in vitro* have been incorporated into bicistronic vectors co-expressing the marker gene EGFP.
- An *in silico* data processing scheme that enabled us to retrieve and cluster human retinal cDNA sequences and expressed sequence tags from public databases was established. For the identification and mapping of transcription initiation and termination sites we have established a routine bioinformatics analysis scheme and an experimental pipeline for the recovery of 5' ends based on the Cap Finder RACE protocol.
- Extensive analysis of both the human and the murine UniGene databases at NCBI and we selected a collection of murine and human cDNA clusters, which are predicted to be predominantly expressed in the eye. These transcripts will represent the main target of detailed expression studies that will be carried out in the second year of the project.
- The *rdl* congenic mice strain were treated at post-natal (PN) 15. Total RNA were purified from five treated animals sacrificed at PN17. The RNA were labeled and hybridized to mouse Affymetrix Genchips (37,000 probe sets).
- Transcriptomics data analysis of GDNF, CNTF and Diltiazem target genes in *rdl* mice. In order to provide high quality results, we established a specialized protocol with the aim to identify regulated target genes combined with a universal quality index. The construction of a relational database dedicated to quality indicators for present and future retina-related transcriptomics experiments and the integration of flexible query tools has been initiated.
- 14 smaller-sized families (with 2-6 affected individuals) with autosomal dominant rhegmatogenous retinal detachment (adRRD) were collected. Analysis was performed using microsatellite markers. Linkage analysis of these loci is in progress.
- Neuroprotection in animal models of retinal degeneration. The first year of the project has seen the start of an interesting line of research concerning the effectors responsible for the retinal degeneration in the *rdl* mouse, which serves as a model for the human disease *Retinitis pigmentosa* (RP).
- Methods to analyse the stoichiometry of protein interactions from primary retinal cells have been established. For enrichment of stoichiometrically physiological protein complexes native separation methods have been chosen and applied to rod outer and rod inner segments. The conceptual principle is to reduce the complexity of the subcellular proteomes to be analysed step by step without destroying their initial functional context by denaturation.