

## Microscopy Group



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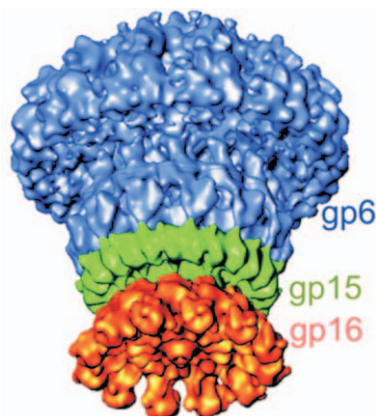
### Scientific overview

The microscopy group provides a central scientific service unit of the institute. Therefore, we collaborate on various subjects with different groups from this institute but also from outside. Beside general service support and smaller projects, the main focus of our methods is on:

- immuno labelling on ultra-thin sections and of isolated structures;
- nucleic acid - protein interactions;
- fine structural analysis of oligomeric proteins, organelles or phages after negative staining or cryo preparation of the samples in vitreous ice.

The lab is running two transmission electron microscopes (EM400 and CM100). The CM100 is designed for cryo work and equipped with a slow scan CCD camera that in addition has widely replaced traditional photographic film for routine work. A confocal microscope (Zeiss LSM510) for 3D analysis of samples labelled with fluorescent dyes is attended.

In collaboration with AG Lanka we are analysing different aspects in conjugation and DNA replication. Only to mention a few topics: The components of the MPF complex were analysed by immuno labelling, structure and binding properties to DNA. Pili of different incompatibility groups like IncP, IncW and IncF were visualized after negative staining. Linear plasmids derived from Phages N15 or PY54 have covalently closed hairpins at their ends which we have demonstrated by showing them denatured as ssDNA rings. The binding site of the telomerase was mapped by binding to the DNA.



The group of E. Wanker (Berlin-Buch) is focused on the molecular pathomechanism of Huntington's disease and related illnesses, which are caused by formation of insoluble aggregates in neuronal cells. We are monitoring by EM the effect of selected drugs on the aggregation of huntingtin,  $\alpha$ -synuclein or  $\beta$ -amyloid fibrils. Proteins formed aggregates in transformed cells that were localized on thin sections by immuno-gold labelling.

The bacteriophage SPP1 was investigated by T. A. Trautner for many years in our institute. We are still co-operating in different aspects of the morphogen-



esis and DNA packaging of this phage with two former members of his group (P. Tavares, Paris; J.C. Alonso, Madrid). One project was the structure of the head to tail interface at one vertex of the phage capsid. The main protein of this connector structure is the portal (gp6) which has a 13fold symmetry in solution but is 12fold in the phage. We have solved the 3D-structures of the portal protein and the connector (see figure) from negatively stained and from in vitrified ice embedded samples in collaboration with M. van Heel's group (FHI in Berlin, now London).

Determining the 3D structure of complex biological structures by cryo EM and image processing will be the main focus for the next years. The UltraStrukturNetzwerk (USN) has started end of last year. First work has already started but at the time the main focus this year is in planning and organizing the setup and facilities for the new cryo EM.

## General information

### Selected Publications 1998-2003

Jensen, RB, **Lurz R** & Gerdes K (1998). *Mechanism of DNA segregation in prokaryotes: Replicon pairing by parC of plasmid R1*. PNAS USA 95:8550-8555

**Deneke J, Ziegelin G, Lurz R & Lanka E** (2000). *The protelomerase of temperate Escherichia coli phage N15 has cleaving-joining activity*. PNAS USA 97: 7721-7726

Friedhoff P, **Lurz R, Lueder G** & Pingoud A (2001). *Sau3AI, a monomeric type II restriction endonuclease that dimerizes on the DNA and thereby induces DNA loops*. J Biol Chem 276:23581-88

**Lurz R, Orlova E, Günther D, Dube P, Dröge A, Weise F, van Heel M & Tavares P** (2001). *Structural organisation of the head-to-tail interface of a bacterial virus*. J Mol Biol 310:1027-37

**Sittler A, Lurz R, Lueder G, Priller J, Lehrach H, Hayer-Hartl MK, Hartl FU & Wanker EE** (2001). *Geldanamycin activates a heat shock response and inhibits huntingtin aggregation in a cell culture model of Huntington's disease*. Hum Mol Genet 10: 1307-1315

**Waelter S, Boeddrich A, Lurz R, Scherzinger E, Lueder G, Lehrach H & Wanker EE** (2001). *Accumulation of mutant huntingtin fragments in aggresomes-like inclusion bodies as a result of insufficient protein degradation*. Mol Biol of the Cell 12:1393-1407

**Deneke J, Ziegelin G, Lurz R & Lanka E** (2002). *Phage N15 telomere resolution. Target requirements for recognition and processing by the protelomerase*. J Biol Chem 277: 10410-19

**Kalkum M, Eisenbrandt R, Lurz R & Lanka E** (2002). *Tying rings for sex*. Trends in Microbiol 10:382-387

Hertwig S, Klein I, **Lurz R, Lanka E** & Appel B (2003). *PY54, a linear plasmid prophage of Yersinia enterocolitica with covalently closed ends*. Mol Microbiol 48:989-1003

Orlova E, Gowen B, **Dröge A, Stiege AC, Weise F, Lurz R, van Heel M & Tavares P** (2003). *Structure of a viral DNA gatekeeper at 10 Å resolution by cryo-electron microscopy*. J Mol Biol 22:1255-1262

Pingoud V, Conzelmann C, Kinzebach S, Sudina A, Metelev V, Kubareva E, Bujnicki JM, **Lurz R, Lüder G, Xu S-Y** & Pingoud A (2003). *PspGI, a type II restriction endonuclease from the extreme thermophile Pyrococcus sp.: structural and functional studies to investigate an evolutionary relationship with several mesophilic restriction enzymes*. J Mol Biol 329:913-929

### Co-operations

Traditionally electron microscopists co-operate with different groups. Many of our co-operations started within this institute and were continued when the scientists moved to new positions. At the time we are working on projects with following external groups:

1. Alonso JC, CSIC, Madrid, Spain
2. Carazo JM, CNB, Madrid, Spain
3. Orlova EV, Birkbeck College, London, UK
4. Pingoud A, Justus Liebig University, Giessen
5. Reeve JN, Ohio State University, Columbus, USA
6. Surayanarayana T, University of Hyderabad, India
7. Tavares P, CNRS, GIF-sur-Yvette, France
8. Weinhold E, RWTH Aachen