1978	Ph.D. - Ludwig-Maximilians-Universität München, Germany (Max Planck Institute of Biochemistry, Martinsried)
1978 - 1986	PostDoc and Group Leader - University of Regensburg, Germany
1986 - 1988	Visiting Scientist - Dept. of Biochemistry, Stanford University, USA
1988 - 1997	Full Professor and Chairman of Biochemistry I - University of Heidelberg, Germany
1991 - 2003	Chairman SFB 352
1997 - 2002	Director - BZH
since 2001	Managing Editor FEBS Letters
2005 - 2007	President of the GBM
since 2003	Chairman SFB 638

Felix Wieland

Molecular mechanisms of COPI transport

Goal

Our research interests comprise two converging fields:

i) Molecular mechanisms of coated vesicle formation and uncoating, and

ii) Specificity and structural basis for protein - lipid interactions within a bilayer that regulate membrane protein activity.

These research interests have led us to develop and maintain

iii) A platform for quantitative lipidomics.

We are characterizing the components and their coordinate action that allow formation, fission and uncoating of Golgi-derived COPI-coated vesicles. This includes proteomics and lipidomics, functional in vitro assays and reconstitution of individual functional steps in a chemically defined liposomal system.

Background

In the eukaryotic cell, vesicular transport represents the basic mechanism for i) maintaining the homeostasis of the endomembrane system, ii) biosynthetic transport of newly synthesized proteins and lipids, and iii) the uptake and intracellular transport of exogenous macromolecules. The mechanism of fusion of vesicles as well as their role in neurotransmission has been recognized by the 2013 Nobel Prize for Physiology and Medicine. Three classes of coated vesicles are well established to mediate transport in the exo- and endocytic pathway: COPII vesicles for ER export, COPI vesicles for retrograde Golgi-ER and bidirectional intra-Golgi transport, and clathrin-coated vesicles operating in the late secretory and endocytic pathway. Coat components are involved in multiple tasks such as cargo selection, curvature formation at the donor membrane, vesicle fission and initiation of uncoating.

We are interested in the molecular mechanisms underlying vesicular transport by COPI vesicles. In contrast to COPII and clathrin coats, the heptameric large COPI coat component coatomer is recruited en bloc to the membrane, so that both the inner and outer shell of the vesicle are formed at the same time.

In our view, the formation of a COPI transport vesicle involves the following minimal set of components: donor membranes with transmembrane proteins acting as coat and/or cargo receptors (e.g. members of the p24 family), cytosolic Arf1, cytosolic coatomer and auxiliary enzymes that serve activation on the membrane of Arf1 (GBF1) and the activation of GTP hydrolysis by Arf1 (Arf GAPs).

Ph.D. - University of Frankfurt, Germany	1998
PostDoc - Memorial Sloan Kettering Cancer Center, New York, USA (Prof. James E. Rothman)	1998 - 2000
PostDoc - BZH (Prof. Felix Wieland)	2000 - 2002
Research Fellow - BZH	since 2002
Habilitation in Biochemistry, University of Heidelberg, Medical Faculty	2007

Britta Brügger

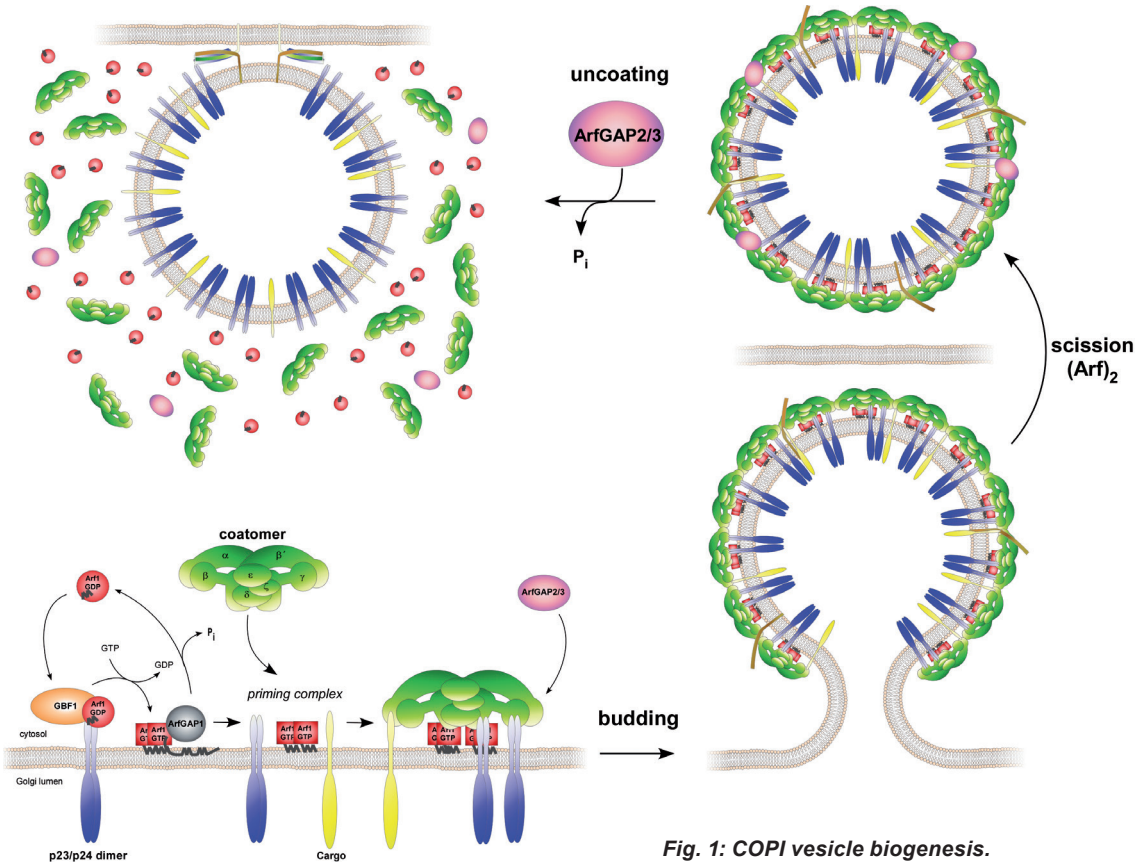

Fig. 1: COPI vesicle biogenesis.

A schematic view of individual steps in COPI vesicle biogenesis is given in Fig. 1.

Research Highlights

Molecular mechanisms of COPI vesicle biogenesis

1) Roles of Arf1 in vesicle formation and fission

Formation of coated vesicles requires two striking manipulations of the lipid bilayer. First, membrane curvature is induced to drive bud formation. Second, a scission reaction at the bud neck releases the vesicle. Using a reconstituted system for COPI vesicle formation from purified components we find that a non-dimerizing Arf1 mutant, which does not display the ability to modulate membrane curvature in vitro or to drive formation of coated vesicles, is able to recruit coatomer to allow formation of COPI-coated buds, but does not support scission. Chemical cross-linking of this Arf1 mutant restores vesicle release. These studies show that initial curvature of the bud is driven primarily by coatomer, whereas the mem-

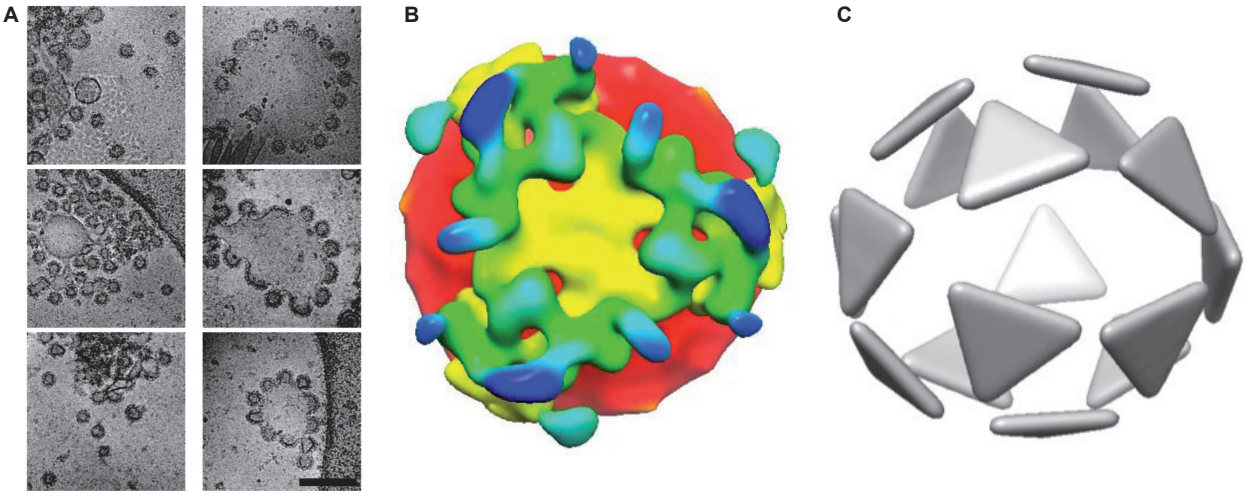

Fig. 2: COPI vesicle biogenesis. A) *Cryo-electron microscopy of COPI-coated vesicles generated with Arf wt (left hand panels) and COPI-coated buds generated with a scission arrest Arf mutant (right hand panels).* **B)** *Three coatomer complexes form a triade, the basic structure of the COPI lattice,* ***C)*** *Arrangement of triads to form the COPI lattice.*

brane curvature potentiating activity of dimeric Arf1 is required for membrane scission. Using a semi-intact cell system we have shown that during the scission reaction Arf does not hydrolyse its bound GTP.

2) Structures of coatomer and of the COPI coat

Together with John Briggs' group at the EMBL we investigate the structure of soluble coatomer by single particle electron microscopy, and of the coatomer shell on coated vesicles. With the first data of a coat on a membrane, a structure emerges that is strikingly different from those of the COPII and the clathrin systems as delineated from protein assemblies. The basic unit of the lattice is a coatomer triade. Triades can be arranged on the vesicular membrane in various patterns that are defined by variable vertices at the contact sites of triades.

3) Regulation of COPI transport by a unique sphingolipid/cargo-receptor complex

We have discovered a specific binding of the sphingomyelin molecular species SM 18:0 to the transmembrane domain of one member of the p24 family, p24. SM 18:0 binding favors dimerization of p24. Dimeric p24, in turn, recruits coatomer and triggers a conformational change of the complex resulting in polymerization, initiating COPI bud formation. Thus, a membrane lipid molecular species can serve as a cofactor in controlling vesicle budding. We have defined steps in vesicular transport *in vivo* that depend on this specific interaction.

Structural principles of transmembrane protein/membrane lipid interactions

1) A signature within the p24 transmembrane domain for recognition of a sphingolipid molecular species

We have discovered a peptide signature for sphingolipid binding within the transmembrane span of p24. When transplanted, the signature confers sphingolipid binding to a non-sphingolipid binding transmembrane domain. Results from a data mining approach indicate that this signature represents a conserved binding site for sphingolipids in several transmembrane proteins.

2) We found that a functional sphingomyelin-binding signature in the influenza neuraminidase is necessary for optimal transport of this protein to the host's plasma membrane.

3) Capitalizing on fluorescent sphingomyelin species containing pentaenyl fatty acids we have developed a simple assay to assess specific lipid-lipid recognition in liposomal membranes.

Lipidomics platform

Using our lipidomics platform, we have elucidated lipidoms of organisms and subcellular systems (see lipidomics by Britta Brügger on page 52).

Our investigations are based on a wide range of methods, including live cell imaging (with Rainer Pepperkok; EMBL), bioinformatics (with Gunnar von Heijne and Arne Elofsson, Stockholm), molecular dynamics simulations (with Erik Lindahl, Stockholm), *in vivo* and *in vitro* FRET studies, cryo-electron microscopy (with John Briggs, EMBL), protein chemistry, molecular biology, and quantitative nano-mass spectrometry of lipids, as well as chemical biology approaches.

Our research is supported by the German Research Council (SFB 638: Dynamics of macromolecular complexes in biosynthetic transport, SFB/TRR 83: Molecular architecture and cellular functions of protein/lipid assemblies, GRK 1188: Quantitative analysis of dynamic processes in membrane transport and translocation, and CellNetworks Heidelberg.

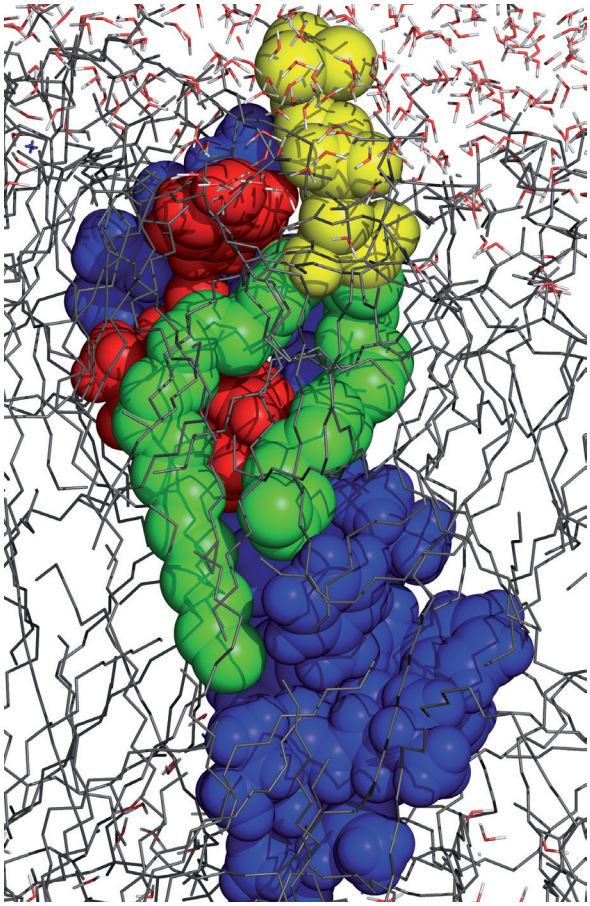

Fig. 3: Structure of a SM 18-binding motif. *Molecular dynamics simulation of p24 TMD (blue, with the motif highlighted in red) and SM 18:0 (green, hydrocarbon chains; yellow, headgroup of SM 18:0) in a POPC bilayer.*

Selected Publications 2011 - 2013

F. Adolf, A. Herrmann, A. Hellwig, R. Beck, B. Brügger and F. T. Wieland (2013) Scission of COPI and COPII vesicles is independent of GTP hydrolysis. Traffic 14(8):922-32.

A.M. Ernst, S. Zacherl, A. Herrmann, M. Hacke, W. Nickel, F.T. Wieland, B. Brügger (2013) Differential transport of Influenza A neuraminidase signal anchor peptides to the plasma membrane. FEBS Lett. 587(9):1411-7.

M. Faini, R. Beck, F.T. Wieland, J.A. Briggs (2013) Vesicle coats: structure, function, and general principles of assembly. Trends Cell Biol. Feb 12.

J.M. Duran, F. Campelo, J. van Galen, T. Sachsenheimer, J. Sot, M.V. Egorov, C. Rentero, C. Enrich, R.S. Polishchuk, F.M. Goñi, B. Brügger, F. Wieland, V. Malhotra (2012) Sphingomyelin organization is required for vesicle biogenesis at the Golgi complex. EMBO J. 12;31(24):4535-46.

M. Faini, S. Prinz, R. Beck, M. Schorb, J.D. Riches, K. Bacia, B. Brügger, F.T. Wieland* and J.A. Briggs* (shared corresponding authors) (2012) The Structures of COPI-coated vesicles reveal alternate Coatomer Conformations and Interactions. Science 336(6087):1451-4.

F.-X. Contreras, A.M. Ernst, P. Haberkant, P. Björkholm, E. Lindahl, B. Gönen, C. Tischer, A. Elofsson, G. von Heijne, C. Thiele, R. Pepperkok, F. Wieland, and B. Brügger (2012) Molecular recognition of a single sphingolipid species by a protein's transmembrane domain. Nature 481:525-529.

R. Beck, S. Prinz, P. Diestelkötter-Bachert, S. Röhling, F. Adolf, K. Hoehner, S. Welsch, P. Ronchi, B. Brügger, J.A. Briggs, and F. Wieland (2011) Coatomer and dimeric ADP ribosylation factor 1 (Arf1) promote distinct steps in membrane scission. J Cell Biol. 194(5):765-77.

F.X. Contreras, A.M. Ernst, F. Wieland and B. Brügger (2011). Specificity of intramembrane protein-lipid interactions. Cold Spring Harb Perspect Biol. Jun 1;3(6)

M.C. Sahlmüller, J.R. Strating, R. Beck, V. Popoff, M. Haag, A. Hellwig, I. Berger, B. Brügger, and F.T. Wieland (2011) Recombinant heptameric coatomer complexes: novel tools to study isoform-specific functions. Traffic 12(6):682-692.

Awards and Honors Felix Wieland

1993	Honorary Member of Charité, Medical Faculty of the Humboldt University, Berlin
since 2000	Elected EMBO Member
2001	Heinrich-Wieland Award
since 2003	Member of Deutsche Akademie der Naturforscher Leopoldina
2006	Feldberg Foundation Award
2011	Elected Member of the Academia Europea
2013	HMLS Award

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