

# Lewy pathology in Parkinson's disease consists of a crowded organellar, membranous medley

**Authors:** Sarah H. Shahmoradian<sup>1,§</sup>, Amanda J. Lewis<sup>1</sup>, Christel Genoud<sup>2</sup>, Jürgen Hench<sup>3</sup>, Tim Moors<sup>4</sup>, Paula P. Navarro<sup>1</sup>, Daniel Castaño-Díez<sup>1</sup>, Gabriel Schweighauser<sup>3</sup>, Alexandra Graff-Meyer<sup>2</sup>, Kenneth N. Goldie<sup>1</sup>, Rosmarie Sütterlin<sup>1</sup>, Evelien Huisman<sup>4</sup>, Angela Ingrassia<sup>4</sup>, Yvonne de Gier<sup>4</sup>, Annemieke J.M. Rozemuller<sup>5</sup>, Jing Wang<sup>1</sup>, Anne De Paepe<sup>6</sup>, Johannes Ery<sup>7</sup>, Andreas Staempfli<sup>7</sup>, Joerg Hoernschemeyer<sup>7</sup>, Frederik Großerüschkamp<sup>8</sup>, Daniel Niedieker<sup>8</sup>, Samir F. El-Mashtoly<sup>8</sup>, Marialuisa Quadri<sup>9</sup>, Wilfred F.J. van IJcken<sup>10</sup>, Vincenzo Bonifati<sup>9</sup>, Klaus Gerwert<sup>8</sup>, Bernd Bohrmann<sup>11</sup>, Stephan Frank<sup>3</sup>, Markus Britschgi<sup>11\*</sup>, Henning Stahlberg<sup>1\*</sup>, Wilma D. J. van de Berg<sup>4\*</sup>, Matthias E. Lauer<sup>6\*</sup>

## Affiliations:

- <sup>1</sup> Center for Cellular Imaging and NanoAnalytics (C-CINA), Biozentrum, University of Basel, Switzerland
  - <sup>2</sup> Friedrich Miescher Institute for Biomedical Research, Switzerland
  - <sup>3</sup> Division of Neuropathology, Institute of Pathology, University Hospital Basel, Switzerland.
  - <sup>4</sup> Amsterdam Neuroscience, VU University Medical Center, Department of Anatomy and Neurosciences, section Clinical Neuroanatomy, Amsterdam, The Netherlands
  - <sup>5</sup> Amsterdam Neuroscience, VU University Medical Center, Department of Pathology, Amsterdam, The Netherlands
  - <sup>6</sup> Roche Pharma Research and Early Development, Chemical Biology, Roche Innovation Center Basel, Basel, Switzerland
  - <sup>7</sup> Roche Pharma Research and Early Development, Preclinical CMC, Roche Innovation Center Basel, Basel, Switzerland
  - <sup>8</sup> Department of Biophysics, Ruhr University Bochum, Germany
  - <sup>9</sup> Department of Clinical Genetics, Erasmus Medical Center, Rotterdam, Netherlands
  - <sup>10</sup> Center for Biomics, Erasmus Medical Center, Rotterdam, Netherlands
  - <sup>11</sup> Roche Pharma Research and Early Development, Neuroscience, Ophthalmology, and Rare Diseases Discovery and Translational Area/Neuroscience Discovery, Roche Innovation Center Basel, Basel, Switzerland
- § Current Address: Department of Biology and Chemistry, Paul Scherrer Institute, Villigen, Switzerland
- \* Shared senior authors

## Correspondence to:

Matthias Lauer ([matthias.lauer@roche.com](mailto:matthias.lauer@roche.com)),  
Wilma van de Berg ([wdj.vandenberg@vumc.nl](mailto:wvj.vandenberg@vumc.nl)),  
Henning Stahlberg ([henning.stahlberg@unibas.ch](mailto:henning.stahlberg@unibas.ch)), and  
Markus Britschgi ([markus.britschgi@roche.com](mailto:markus.britschgi@roche.com))

## Movies

**Movies are available on YouTube, at**

**<https://www.youtube.com/playlist?list=PLteVxMX-rlQu3HIZtObzPRspOr9uvA4s9>**

**and individually at the links provided below.**

**Movie 1:** Reconstructed and color-segmented 3D transmission electron tomogram of aSyn-immunopositive inclusion (LB). Corresponds to Fig. 1a. Thickness of tissue section imaged  $\approx$  150 nm.  
<https://youtu.be/StUx7Gxp3tI>

**Movie 2:** Reconstructed and color-segmented 3D transmission electron tomogram of aSyn-immunopositive inclusion (LB). Corresponds to Fig. 1b. Thickness of tissue section imaged  $\approx$  150 nm.  
<https://youtu.be/FwZX3X6QkjQ>

**Movie 3:** Reconstructed and color-segmented 3D transmission electron tomogram of aSyn-immunopositive inclusion (LB). Corresponds to Fig. 1c. Thickness of tissue section imaged  $\approx$  150 nm.  
<https://youtu.be/QRX73sGSbt4>

**Movie 4:** Reconstructed and color-segmented 3D transmission electron tomogram of aSyn-immunopositive inclusion (LB). Corresponds to Figs. 1d, S5. Thickness of tissue section imaged  $\approx$  150 nm.  
<https://youtu.be/ROQ6mbj8VIA>

**Movie 5:** Reconstructed and color-segmented 3D transmission electron tomogram of aSyn-immunopositive inclusion (LB). Corresponds to Fig. S4a. Thickness of tissue section imaged  $\approx$  150 nm.  
<https://youtu.be/7-2CN-V7NKQ>

**Movie 6:** Reconstructed and color-segmented 3D transmission electron tomogram of aSyn-immunopositive inclusion (LB). Corresponds to Fig. S4b. Thickness of tissue section imaged  $\approx$  150 nm.  
<https://youtu.be/c82B7DCBIRE>

**Movie 7:** Reconstructed and color-segmented 3D transmission electron tomogram of aSyn-immunopositive inclusion (LB). Corresponds to Fig. S4c. Thickness of tissue section imaged  $\approx$  150 nm.  
<https://youtu.be/ec5BG-ItlxM>

**Movie 8:** Reconstructed and color-segmented 3D transmission electron tomogram of aSyn-immunopositive inclusion (LB). Corresponds to Fig. S4d. Thickness of tissue section imaged  $\approx$  150 nm.  
<https://youtu.be/iR6985Mp8qc>

**Movie 9:** Reconstructed and color-segmented 3D transmission electron tomogram of aSyn-immunopositive inclusion (LB). Corresponds to Fig. S4e. Thickness of tissue section imaged  $\approx$  150 nm.  
[https://youtu.be/wg7v7I\\_BGsQ](https://youtu.be/wg7v7I_BGsQ)

**Movie 10:** Reconstructed and color-segmented 3D transmission electron tomogram of aSyn-immunopositive inclusion in neurite (LN). Corresponds to Fig. S4f. Thickness of tissue section imaged  $\approx$  150 nm.  
<https://youtu.be/Y0FaBmbWvpQ>

**Movie 11:** Reconstructed and color-segmented 3D transmission electron tomogram of a region inside aSyn-immunopositive inclusion (LB, Fig. 1a) collected at higher magnification. Thickness of tissue

section imaged  $\approx 150$  nm.

<https://youtu.be/lwV0-xPiH5I>

**Movie 12:** Reconstructed and color-segmented 3D transmission electron tomogram of a region inside aSyn-immunopositive inclusion (LB, Fig. 1a) collected at higher magnification. Tailed membrane stacks are clearly visible, as indicated in Fig. 1a (two yellow arrow-heads on right-hand side).

Thickness of tissue section imaged  $\approx 150$  nm.

<https://youtu.be/9SDeEs5yJdQ>

**Movie 13:** Reconstructed and color-segmented 3D transmission electron tomogram of region at the edge of the aSyn-immunopositive inclusion (LB, Fig. 1a) collected at higher magnification. A mitochondrion is clearly visible, as indicated in Fig. 2c (white oval). Thickness of tissue section imaged  $\approx 150$  nm.

<https://youtu.be/2TwAJcmoH1g>

**Movie 14:** Reconstructed and color-segmented 3D transmission electron tomogram of region at the edge of the aSyn-immunopositive inclusion (LB, Fig. S4a) collected at higher magnification.

Thickness of tissue section imaged  $\approx 150$  nm.

<https://youtu.be/sgij8doJNzQ>

**Movie 15:** Reconstructed and color-segmented 3D transmission electron tomogram of region inside the aSyn-immunopositive inclusion (LB, Fig. S4a) collected at higher magnification. Cluster of vesicles in separate adjacent compartment to LB is visible as shown in Fig. 2d. Thickness of tissue section imaged  $\approx 150$  nm.

<https://youtu.be/lfcvCX133OU>

**Movie 16:** Reconstructed and color-segmented 3D transmission electron tomogram of region within an aSyn-immunopositive Lewy neurite (same as shown in Fig. 3a) collected at high magnification.

Thickness of tissue section imaged  $\approx 150$  nm.

[https://youtu.be/ogdMLPaz\\_T0](https://youtu.be/ogdMLPaz_T0)

**Movie 17:** Reconstructed and color-segmented 3D transmission electron tomogram of region within an aSyn-immunopositive Lewy neurite (same as shown in Fig. 3b) collected at high magnification.

Thickness of tissue section imaged  $\approx 150$  nm.

<https://youtu.be/D4r2PjtVy80>

**Movie 18:** Reconstructed and color-segmented 3D transmission electron tomogram of region within a 'control' neurite in brain tissue from a non-demented, age-matched donor (same as shown in Fig. 3c) collected at high magnification. Thickness of tissue section imaged  $\approx 150$  nm.

<https://youtu.be/YIn15OccuGs>

**Movie 19:** Reconstructed and color-segmented 3D transmission electron tomogram of region within a 'control' neurite in brain tissue from a non-demented, age-matched donor (same as shown in Fig. 3d) collected at high magnification. Thickness of tissue section imaged  $\approx 150$  nm.

<https://youtu.be/arL5GfyFgWM>

**Movie 20:** Reconstructed serial block-face scanning electron tomograms depicting three separate Lewy pathological inclusions within the *substantia nigra* of Donor B. Scale bar = 5  $\mu$ m.

<https://youtu.be/O1Xb2LaELMI>

**Movie 21:** Stimulated emission depletion microscopy showing a Lewy pathological inclusion in the same tissues (Donor B, *substantia nigra*) as taken from parallel blocks for the SBFSEM ultrastructural analysis (Fig. 3d). Thickness of tissue section = 20  $\mu$ m.

<https://youtu.be/dKO9HZqGTTI>