

Supporting Information

RP-CARS reveals molecular spatial order anomalies in myelin of an animal model of Krabbe disease.

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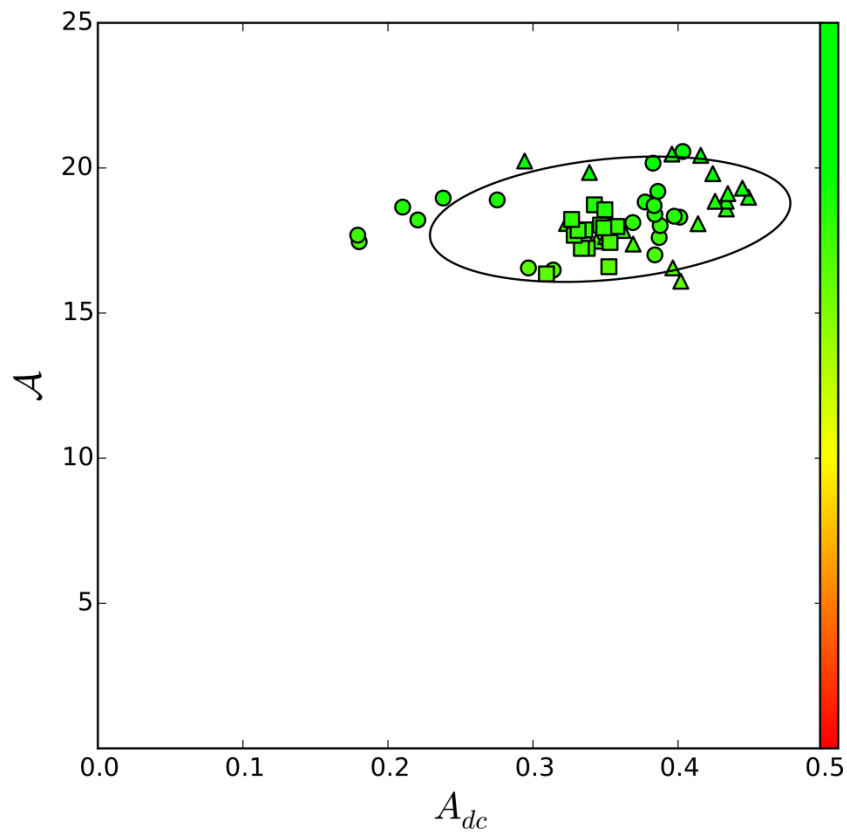


Figure S1. Correlation plot between the average alpha value of manually thresholded slices (A , on the y-axis) and their average A_{dc} value (on the x-axis). Each point represents a slice of a (WT mouse) z-stack (i.e. at different depth inside the tissue) and different shapes denote different z-stacks. The points are colour-coded according to the colour bar shown on the right and consistently with Figure 3. The ellipse shows the confidence interval at twice the standard deviation. The (Pearson product-moment) correlation coefficient ($R^2=0.25$) between A and A_{dc} is not statistically significant ($p>0.05$).

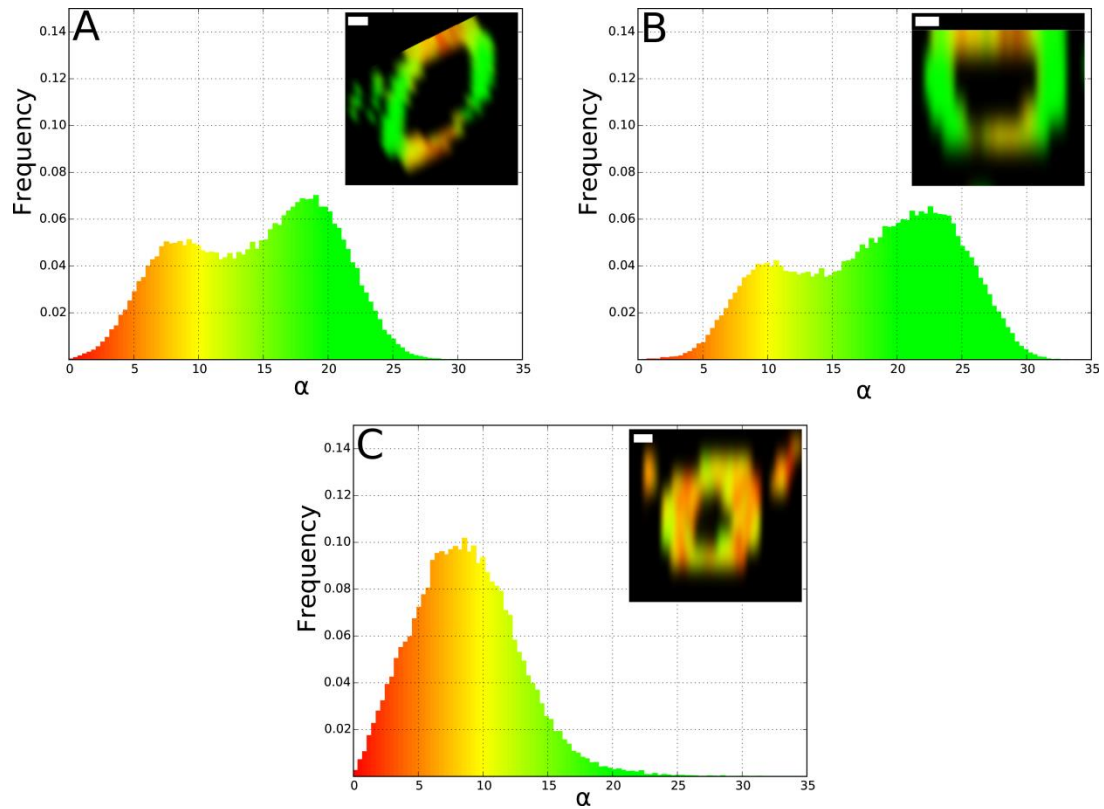


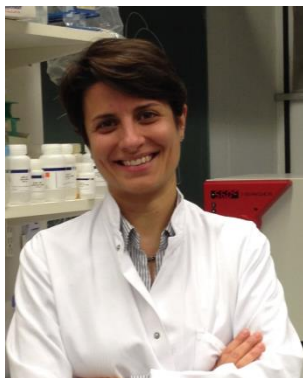
Figure S2. Normalised frequency histograms of the α values in a z-stack representative of the WT condition (A), of the presymptomatic TWI condition (B) and of the symptomatic TWI condition (C). Insets: virtual reconstructions of the myelin walls in the optical transverse plane, constructed from the z-stacks corresponding to the same respective histograms. The images are constructed as in Figure 3: by mapping the value of α onto the hue (from red, $\alpha = 0$, to green, $\alpha \geq 20$, as displayed on the histograms bars), the $A_{2\omega}$ signal onto the saturation and the A_{dc} signal onto the value. However, differently from Figure 3, the areas below the automatically-chosen threshold (as described in the “Materials and methods” section) and the areas corresponding to non-acquired volumes are displayed in black. Scale bar: 1 micron.



Giuseppe de Vito studied Biology at University of Pisa and Scuola Normale Superiore (Italy). Former Junior Specialist at University of California-Irvine (USA), he is currently completing his Ph.D. in Molecular Biophysics at Scuola Normale Superiore in collaboration with Istituto Italiano di Tecnologia at the NEST institute in Pisa (Italy). The main topic of his Ph.D. thesis is the label-free polarization-resolved optical imaging of biological samples.



Valentina Cappello studied Biology at the University of Milan (Italy) working on neurodegenerative disorders based on ultrastructural characterization. After graduation she continued to work several years in the same field focusing on ALS and other neuro/muscular disorders. At present, she works in CNI centre of IIT (NEST Pisa, Italy) where she is working in the characterization of the interaction between nanostructured material and biological samples and in the development of new methods for the correlation of different imaging techniques and transmission electron microscopy



Ilaria Tonazzini graduated in Pharmaceutical Chemistry and Technology and got a PhD in Sciences of Drugs and Bioactive Substances at the University of Pisa, Italy. She then worked as postdoc at the Institute for Nanostructured Materials Studies-CNR in Bologna and from 2010 she works at NEST (National Enterprise for nanoScience and nanoTechnology), Istituto Nanoscienze-CNR and Scuola Normale Superiore, in Pisa. From 2015 she is fellow of the Fondazione Umberto Veronesi. Her main research interests focus on the study of neuronal environmental sensing and cytoskeleton dynamics in patho-physiological conditions.



Marco Cecchini graduated in Physics at the University of Pisa (Italy) and got a PhD in Condensed Matter Physics at the Scuola Normale Superiore of Pisa (Italy). Currently he is a researcher at the Nanoscience Institute of the National Research Council (CNR, Italy). His main research interests focus on peripheral nervous system tissue engineering, study of pathogenesis in Krabbe disease, and microfluidics.



Vincenzo Piazza graduated in Physics at University of Pisa and Scuola Normale Superiore (Italy), where he also received his PhD in Condensed Matter Physics. He is currently Team Leader at Fondazione Istituto Italiano di Tecnologia (Pisa, Italy). He was director of the Center of Nanotechnology Innovation, IIT@NEST. His research activity included coherent transport in mesoscopic systems, electronic transport in the quantum-Hall regime, and single-photon sources for quantum cryptography. He is now focused on the development and applications of non-linear microscopy techniques.