3D Imaging and Analysis

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3D reconstruction of lipid droplets in lipase deficient macrophages

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Macrophages are a very good model system for studying lipid metabolism due to their high capacity for lipid accumulation. Ultrastructural analysis of lipid droplet formation in macrophages might help understanding foam cells formation, a major player in development of atherosclerosis.
We analyzed lipid droplet ultrastructure in macrophages obtained from different lipase deficient mouse models: ATGL -/- (Adiopse triglyceride lipase), HSL -/ - (Hormone sensitive lipase), ATGL –/- HSL -/- double knock out and LAL -/- (Lysosomal acid lipase).
Without exogenous lipid loading we saw accumulation of lipids in macrophages of all strains compared to wild type. In ATGL - /- we investigated many classical large lipid droplets with a leaflet. A small number of lipid droplets was observed in HSL -/- however this seems to be normal because in WT we also see similar lipid accumulation. In comparison to these single knock outs electron microscopy (EM) revealed significant ultrastructural differences in double knock outs. Lipid droplets appeared to cluster thereby losing their spherical shape. In LAL -/- neutral lipids generally accumulate in lysosomes which can readily be distinguished from LD´s due to double layer membranes around the accumulated lipids. After loading with VLDL we see normal lipid droplet formation and many clusters of lipid loaded lysosomes. By using different EM approaches and especially serial sectioning we were able to visualize heterogenous ultrastructure in the lysosomes.
High resolution analysis of lipid droplets in macrophages deficient of different lipases revealed different ways of lipid accumulation.