Peroxisomes

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MEETING REPORT

Peroxisomes: new insights into protein sorting, dynamics, quality control, signalling and roles in health and disease

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Introduction

The 6th Open European peroxisome meeting (OEPM) was held on the 26th and 27th of October (2018) in Groningen, the Netherlands. OEPM is a biannual meeting organized by a European peroxisome research group. Previous meetings were held in Leuven, BE (2006), Lunteren, NL (2010), Dijon, FR (2012), Neuss, GER (2014) and Vienna, AU (2016). Over 120 participants were registered from 14 European countries, as well as Israel, Canada, the USA and South Korea. A large number of European research groups participated, including established and younger groups, showing that peroxisome research is blooming in Europe. This will further expand with the EU Marie Curie Innovative training network PERICO (PERoxisome Interactions and COmmunication; http://www.itn-PERICO.eu; coordinated by Ida van der Klei), which recently started and aims to train the next generation of peroxisome researchers.

At OEPM young peroxisome researchers (PhD students, junior Post-docs) present their work to an international audience, with a total of 27 talks and 40 posters being presented.

Meeting Report of the 6th Open European Peroxisome Meeting, Groningen, The Netherlands.

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During the meeting the OEPM 2018 Peroxisome Research Award (sponsored by BBA Molecular Cell Research) was awarded. This award is given to the early career researcher who was first author of the best publication in the peroxisome field over the last 2 years. Members of the jury were Myriam Baes (Leuven), Marten Veenhuis (Groningen) and Ron Wanders (Amsterdam). The awarded paper describes a novel peroxisome–ER contact site in mammalian cells (Costello et al. 2017a). The first author of this paper, Joe Costello (University of Exeter, UK), received the prize during the meeting and presented the Peroxisome Research Award lecture in the 2nd session (see below).

Peroxisomes are cell organelles consisting of a single membrane that encloses a proteinaceous matrix. Their enzyme content is highly variable and determines the function of these versatile organelles. Common functions are hydrogen peroxide and lipid metabolism, but peroxisomes can also contain highly specialized pathways, for instance enzymes involved in ether lipid or penicillin biosynthesis. Peroxisomes are also implicated in non-metabolic functions, such as signalling, innate immunity and ageing. There is now substantial evidence that peroxisomes actively contribute to cell signalling and that their functions are required for human health. Peroxisome deficiency or functional impairment result in devastating genetic conditions known as peroxisome biogenesis disorders (PBDs); but also recently peroxisomes were found to contribute to the pathology of Alzheimer's and Parkinson's diseases, ageing, cancer, type 2 diabetes, and heart failure; and affect immune responses against pathogens (Beach et al. 2012; Colasante et al. 2015; Di Cara et al. 2017; Dixit et al. 2010; Fransen et al. 2013; Trompier et al. 2014).

The interest in peroxisome research is still growing. Various aspects of these intriguing organelles are being studied, including their function, biogenesis, dynamics (fission, transport), degradation by autophagy (pexophagy) and quality control. Many different organisms are studied, including

yeasts, *Caenorhabditis elegans*, plants and mammals. The latter includes also medical research aiming at identifying novel leads for therapy and drug design. Another medically relevant topic is the biology of glycosomes in Trypanosomes, human parasites that cause sleeping sickness. Glycosomes are highly specialized peroxisomes containing glycolytic enzymes, rendering them very attractive as drug targets.

Below is an overview of the sessions and talks of this very successful and inspiring OEPM.

Session 1: peroxisomal protein sorting

Chair: Einat Zalckvar, Weizmann Institute of Science, Israel

Protein targeting to peroxisomes is a subject that has been extensively studied in the last years. The studies led to exciting results and enabled a good understanding of how proteins are targeted to peroxisomes. However, while the basic understanding is currently known, new questions are coming up such as: What is the exact molecular mechanism by which the targeting receptors of the matrix proteins are recycled? What is the targeting priority of the different peroxisomal proteins? How are proteins, which are dually localized to peroxisomes and to another organelle, targeted to both compartments? Are Peroxisome Membrane Proteins (PMPs) translated in proximity to peroxisomes? In the first session of the OEPM meeting some of these topics were discussed.

Noa Dahan (Maya Schuldiner laboratory, Weizmann Institute of Science) presented her recent findings suggesting localized translation as a mechanism for targeting of PMPs to their final destination. By applying proximity specific ribosome profiling Noa found that ribosomes interact with the surface of peroxisomes while translating a specific subset of PMPs which are not translated by ribosomes that interact with the ER. Noa also showed that the interaction between ribosomes and peroxisomes is abolished or reduced upon specific PMP deletion suggesting that these proteins anchor or regulate the anchoring of ribosomes to peroxisomes.

Two presentations discussed Pex5 ubiquitination: Ana Pedrosa (Jorge Azevedo laboratory, i3S/University of Porto) talked about their recent findings on the recognition and extraction of monoubiquitinated PEX5 (Ub-PEX5) by the AAA ATPases PEX1/PEX6. Using their established cell-free in vitro system, coupled with photoaffinity crosslinking and PEG-maleimide labelling, they showed that the ubiquitin moiety of Ub-PEX5 interacts with both PEX1 and PEX6, and that the polypeptide chain of PEX5 is globally unfolded during the extraction step (Pedrosa et al. 2018). These observations establish peroxisomal Ub-PEX5 as a bona fide substrate of these AAA ATPases. Rebecca Brinkmeier (Harald Platta laboratory, Ruhr-University, Bochum) presented their recent findings that chimeric Peroxisome Targeting Signal 1 (PTS1) receptor molecules, consisting of ubiquitin genetically fused to a Pex5p variant that lacks the monoubiquitination site, can facilitate peroxisomal matrix protein import (El Magraoui et al. 2019). Interestingly, when the ubiquitin moiety is mutated in order to slow down deubiquitination, the peroxisomes exhibit a complete PTS1-import defect, which is caused by a disturbed association with the import pore protein Pex14. The data indicate that balanced ubiquitination and deubiquitination dynamics regulate PTS1-protein import and that deubiquitination is a novel essential step in the receptor cycle of the PTS1-receptor Pex5 in the yeast *Saccharomyces cerevisiae*.

Two following presentations discussed targeting priority to peroxisomes by the Pex5 targeting receptor. In the first talk Sven Fischer (Bettina Warscheid laboratory, University of Freiburg) presented data on phosphorylation of Pex5 in *S. cerevisiae* using phosphoproteomic methodology. He found that phosphorylation of Pex5 at distinct sites partially impairs import of GFP-SKL into peroxisomes. Using native mass spectrometry and isothermal calorimetry, Sven investigated phosphorylation-dependent changes in receptor-cargo binding. His findings demonstrate that phosphorylation of Pex5 provides a new mechanism for modulating matrix protein import into peroxisomes at the posttranslational level. In the second presentation Mira Rosenthal (Maya Schuldiner laboratory, Weizmann Institute of Science) presented a new approach to study peroxisomal protein targeting priority in yeast. Mira expressed different levels of a mCherry protein with a PTS1 signal as targeting competitor for Pex5 cargo proteins. Using this approach, Mira found that only PTS1 proteins were affected in localization by the presence of the mCherry–PTS1 competitor. Surprisingly, there was a subset of PTS1 proteins that were not affected by the competitor indicating that they have a targeting priority. Mira is now investigating the mechanisms that enable priority such as the use of an alternate targeting receptor (such as Pex9), utilizing an alternative Pex5 binding site or having a strong affinity to Pex5.

Last, but definitely not least, Thorsten Stehlik (Michael Bölker laboratory, University of Marburg) talked about the discovery that the type 2C protein phosphatase Ptc5p from *S. cerevisiae* transits from mitochondria to peroxisomes. Thorsten found that Ptc5 dually localizes to mitochondria and peroxisomes. The peroxisomal localization requires both the mitochondrial inner membrane peptidase Imp1 as well as the peroxisomal import receptor Pex5. Thorsten showed that the phosphatase is first directed to mitochondria via an N-terminal bipartite mitochondrial targeting signal. Then the protein is cleaved by the inner membrane protease Imp1 and subsequently it is sorted to peroxisomes. Taken together, his

results point towards a novel protein sorting pathway from mitochondria to peroxisomes.

Session 2: peroxisome dynamics and transport

Chair: Stephan Kemp, Amsterdam UMC, the Netherlands

Peroxisomes are extremely dynamic organelles that constantly adapt their number and size in response to cellular demand, but also in response to environmental conditions. How do they do that? What are the underlying processes that regulate peroxisome abundance and maintenance? How do peroxisomes grow, what triggers their growth, and where do the lipids needed for membrane synthesis come from? How do peroxisomes segregate during cells division?

The researchers that presented their work in this 2nd session on "peroxisome dynamics and transport" are using various model systems, like yeast, plant, mammalian cells, but also in silico models, to gain insight into these fundamental questions.

Plant immune-associated nucleotide-binding (IAN) proteins belong to the family of small GTPases and are associated with immunity-related functions. Saugat Pokhrel (Sigrun Reumann laboratory, Institute of Plant Science and Microbiology, Hamburg) presented a newly identified role of 1 of the 13 *Arabidopsis thaliana* IAN proteins in peroxisome cluster formation. The presented data supports that post-translational modification of AtIAN protein determines protein targeting to the peroxisomal membrane.

Peroxisomes are extremely dynamic. They constantly adapt their number in response to cellular demand, remodeling their membrane while undergoing growth and division. Josiah Passmore (Michael Schrader laboratory, University of Exeter) presented a mathematical, biophysical model of peroxisome growth and division dynamics. This model describes what key parameters regulate peroxisome proliferation and morphology, and how these parameters can be manipulated to give rise to different patient peroxisome phenotypes. In the future, it is hoped that the model will be useful to understand what governs peroxisome proliferation, linking the morphological observations with the physical processes underlying regulation of peroxisome abundance.

Rising ambient temperature is considered to be one of the most detrimental stress conditions for plants. Heat stress causes several alterations in plant growth and development, mostly due to the excess production of reactive oxygen species (ROS). Lennart Charton (Nicole Linka laboratory, Heinrich Heine University, Düsseldorf) presented a newly identified transport protein of the mitochondrial carrier family in the model plant *Arabidopsis thaliana* that is located to the peroxisome. The protein is highly upregulated upon heat stress and mutant plants are more susceptible to elevated temperatures. In vivo redox measurements unveiled a disturbed cytosolic glutathione homeostasis during times of elevated ROS production. Therefore, they named the protein "peroxisomal heat stress responsive carrier 1" (PHS1). It is hypothesized that PHS1 plays an important role in ROS scavenging during times of increased oxidative stress by transporting substrates that are directly or indirectly involved in plant stress response.

Stem cells can maintain homeostasis by dividing asymmetrically to produce a new stem cell and a progenitor cell that gives rise to the differentiated cell types of a tissue. Hien Bui (Pekka Katajisto laboratory, Institute of Biotechnology, University of Helsinki) showed that peroxisomes are age-selectively and asymmetrically segregated during division of stem-like cells of the human mammary gland. Interestingly, the daughter that becomes the new stem-like cell receives older peroxisomes. During mitosis, the majority of peroxisomes bind to the astral microtubules of the mitotic spindle and the two poles of the mitotic spindle demonstrate different selectivity for old and young peroxisomes. These data indicate that spindle pole associated peroxisomes define the level of age-selective apportioning of peroxisomes between daughter cells and suggest that peroxisomes regulate self-renewal and differentiation via two spindle-dependent mechanisms: by orienting the spindle and by their own age-selective inheritance discovered here.

Yeast peroxisomes lack lipid biosynthetic enzymes. Therefore, growth of the peroxisomal membrane needs transport of membrane lipids from other organelles. Intracellular lipid transport can occur at membrane contact sites (MCSs), which are regions of close apposition between two organelles. Proteins that physically bridge two organelles at MCSs are called tethers. Fei Wu (Ida van der Klei laboratory, University of Groningen) presented data on Pex23, Pex24 and Pex32 in the yeast *Hansenula polymorpha*. By making single deletion mutants their role in endoplasmic reticulum (ER)-peroxisome contact sites was investigated. Deletion of Pex23, Pex24 or Pex32 resulted in a reduction in the number of peroxisomes, an increase in their size and an increased distance between the ER and peroxisomes. These peroxisome morphology deficiencies could be rescued by the introduction of an artificial peroxisome-ER tether. Based on these data Fei proposed that Pex32 is a candidate tether protein at the ER-peroxisome contact sites in *H. polymorpha*.

In the Peroxisome Research Award lecture Joe Costello (Michael Schrader laboratory, University of Exeter) described the work which led to the discovery of the first peroxisome-ER tethering components in mammalian cells (Costello et al. 2017a, b). For many years peroxisomes and the ER had been known to form close physical associations and exchange lipids but the molecular players involved

had remained elusive. Joe identified two peroxisomal acyl-CoA binding proteins, ACBD4 and ACBD5, which use a FFAT-like motif to interact with the ER membrane protein VAPB. These interactions mediate physical contacts between peroxisomes and the ER, bringing the two organelles into close proximity. The ACBD5-VAPB complex is required for peroxisomal membrane expansion and also to regulate the movement of peroxisomes. Patients with mutations in ACBD5 have recently been identified and the role peroxisome contact sites play in disease is currently being investigated.

Session 3: peroxisomes in health and disease

Chair: Francesca di Cara, Department of Microbiology and Immunology, Dalhousie University

Understanding how peroxisomes are regulated physiologically and how peroxisomes participate in signal transduction remains an unexplored and challenging problem with broad potential to illuminate novel cellular processes in health and disease. Talks in the 3rd session "Peroxisomes in Health and disease" of this vibrant meeting discussed new and exciting findings regarding the different roles that peroxisomes play in organism health and their potential role as a therapeutic target for drug discovery in various disease.

Neglected tropical diseases (NTD) are a group of 17 insect-transmitted infectious diseases, which affect more than 1 billion people worldwide. Among these, three diseases are caused by trypanosomatid parasites; African sleeping sickness, Chagas disease and Leishmaniasis. More than 20 million people are infected with one of these parasites, leading to over 30,000 deaths annually. Currently used therapies against these diseases are ineffective and therefore identification of new drugs and drug targets is necessary. Unlike in all other organisms, trypanosomes compartmentalize glycolytic enzymes inside a unique organelle called glycosome, which are evolutionary related to peroxisomes. Glycosomes are essential for the survival of trypanosomes, and therefore considered as an attractive drug target. Ann-Britt Schäfer (Ralf Erdmann laboratory, Ruhr-University, Bochum) summarized their work on glycosomes for the development of new therapies against trypanosomatid parasites. In cooperation with the Sattler laboratory (Helmholtz Zentrum Munich, Germany), they previously exploited the glycosomal matrix protein import as drug target by designing small molecule inhibitors of *Trypanosoma* Pex5-Pex14 interaction. Currently, the *Trypanosoma* Pex19 interactome is being characterized to utilize the glycosomal membrane protein import machinery as new drug target. They identified two new glycosomal membrane proteins, ATAD1 and an unusual long Pex11-like protein in trypanosomes. RNAi knockdown studies showed that these proteins are essential for the parasite growth, indicating their potential as novel drug targets. These studies will enable the design of efficient therapies against trypanosomiasis and leishmaniasis by using glycosomes as therapeutic targets.

Peroxisomes have been demonstrated to be essential to modulate innate immune responses acting as signal platform in the immune cells (Di Cara et al. 2017; Dixit et al. 2010). Inflammation represents one of the main ways of innate immune cells to deal with and combat early phase infections. However, high or persistent inflammation brings detrimental effects for the body and promotes disease. Innate immune cells possess intracellular receptors that sense homeostasis imbalance. Juan Francisco Rodríguez-Alcázar (Eicke Latz laboratory, University of Bonn) reported his latest finding that demonstrated the importance of peroxisomes to regulate inflammation. Using a genetical model, he found that macrophages lacking peroxisomes showed dysregulated innate immune activation towards several pro-inflammatory stimuli.

Peroxisomal β-oxidation defects lead to the development of severe neurodegenerative disorders. X-linked adrenoleukodystrophy (X-ALD) is characterized by very long chain fatty acid (VLCFA) accumulation resulting from mutations in the ABCD1 gene. This gene encodes for a peroxisomal half ABC transporter which, like its closest homologue ABCD2, participates in the entry of VLCFA-CoA into the peroxisome. Progress in understanding the physio-pathogenesis of X-ALD suffers from the lack of appropriate cell and animal models.

Quentin Raas (Stéphane Savary laboratory, University of Bourgogne Franche-Comté) demonstrated that the peroxisomal defects in microglia are a key element of the onset of the disease. He generated BV-2 microglial cell lines deficient in ABCD1, ABCD2, both ABCD1 and ABCD2 or ACOX-1 (the first rate-limiting enzyme of the peroxisomal β-oxidation system) by CRISPR/Cas9 engineering. These cell lines present high inflammatory status (cytokine release and expression) in basal conditions or upon LPS stimulation and phagocytosis defects. Thus, these data suggest a clear link between peroxisomal defects and altered immune response in the microglial cell lines. Altogether, the novel mutant cell lines that he generated and characterized represent a promising model that should permit identification of new therapeutic targets for this complex neurodegenerative disease.

Zellweger Spectrum Disorder (ZSD) usually results from biallelic mutations in *PEX* genes required for peroxisome biogenesis. PEX1-p.G843D is a common hypomorphic allele that produces a misfolded, degraded protein. Catherine Argyriou (Nancy Braverman laboratory, McGill University) reported the characterization of a highly effective flavonoid,

diosmetin, as a potential drug to recover peroxisome functions in patients with at least one PEX1-p.G843D allele. To see if diosmetin could be useful for other 'exportomer' mutations, Catherine tested its effects on 20 different patient cell lines with *PEX1, PEX6*, or *PEX26* mutations. Interestingly, she found that diosmetin recovered peroxisome functions in PEX1-p.G593R, PEX6-p.G220V, PEX6-p.L946_T947delinsPro, and PEX26-p.R298W patient cell lines, but had no effect on null lines. Candidate drug testing on cells with additional *PEX* mutations is ongoing. She hypothesized that diosmetin acts as a pharmacological chaperone that improves the stability, conformation, and functions of PEX1/PEX6/PEX26 exportomer complexes, and that this recovery depends on the mutation's effect on complex structure. Interestingly she also reported that this drug has different effects in mice carrying equivalent PEX1-p.G843D mutation. Her work led to the important conclusion that the murine complex might behave differently, so caution is needed in moving candidate drugs from patient cell lines to testing in the mouse models.

Patients with peroxisome biogenesis disorders or a deficiency in peroxisomal β-oxidation, such as loss of ACOX1 or MFP2 (also called D-bifunctional protein), often present retinopathy. However, the importance of peroxisomal β-oxidation for the development and functioning of the retina remains obscure. Yannick Das (Miriam Baes laboratory, University of Leuven) presented the characterization of the retinal pathology in a previously developed mouse model with deficient peroxisomal β-oxidation (MFP2 knockout). Already at the age of 3 weeks, MFP2 knockout mice displayed a reduced size of the photoreceptor outer segments (POS) and protrusion of retinal pigment epithelium (RPE) cells into the POS layer. This resulted in impaired retinal function based on electroretinogram measurements. At the age of 8 weeks, reduced visual acuity was proven by an optomotor repsonse test. Both in the RPE and in photoreceptors ultrastructural abnormalities were noticed, including mitochondrial damage. Furthermore, accumulation of neutral lipid droplets was observed in the RPE. Thus, his data reveal that peroxisomal β-oxidation is essential for retinal homeostasis and function at an early age. Furthermore, the pathology in the mouse model resembles the pathological findings in man and is therefore an attractive model system for future mechanistic studies.

Session 4: peroxisomal protein degradation and autophagy

Chair: Peter Kim, Hospital for Sick Children, University of Toronto, Canada

An old proverb that "all good things must come to an end" stands true in biology. The session of peroxisomal protein degradation and autophagy focused on mechanisms by which the cell degrades peroxisomes and their proteins, and the disease consequences of a defect in peroxisome quality control.

A common theme that reverberates in the mechanisms of peroxisome quality control is ubiquitin. Ubiquitin appears to act as a signalling molecule for the degradation not only for the turnover of PMPs but for the whole organelle itself. Previously, the PMPs Pex3 and Pex13 were shown to be degraded by the ubiquitin–proteasome system (UPS) (Bellu et al. 2002; Chen et al. 2018; Williams and van der Klei 2013).

To study the mechanisms of PMP degradation, Srishti Devarajan (Chris Williams laboratory, University of Groningen) presented their synthetic genetic array studies using tandem fluorescent protein timers to determine the rate of degradation of various PMPs. They found one of the most unstable PMPs in *S. cerevisiae* is Pxa1p, a fatty acid transporter that is homologue to the mammalian ALDP. Loss of ALDP is associated with X-linked Adrenoleukodystrophy. They showed that a mutant form of Pxa1p (Pxa1MUT), designed to mimic X-ALD causing ALDP mutations, is inhibited in function and therefore rapidly degraded from peroxisomes in an UPS dependent mechanism. Interestingly they show that preventing Pxa1MUT degradation restores Pxa1p function in vivo, suggesting that some of the X-ALD pathogenicity is not due to loss of protein function but caused by the instability of the protein.

Daniel Schwerter (Ralf Erdmann laboratory, Ruhr University, Bochum) described a potential regulator of UPS dependent PMP quality control. The Peroxisomal AAA ATPase composed of PEX1/PEX6 removes ubiquitinated proteins from the membranes of peroxisomes. Daniel presented a novel function of Msp1/ATAD1 on peroxisomes, a AAA ATPase that has previously been shown to remove tail-anchored proteins from the mitochondria outer membrane. They show that the peroxisomal localized ATAD1 removes ubiquitinated PEX5. In particular, they show that over-expression of Msp1p/ATAD1 in PEX1 deficient cells in yeast can reduce a number of ubiquitinated PMPs including Pex5 and Pex13. Similarly, removing Msp1/ATAD1 in mammalian cells stabilized PEX5 in the PEX1 deficient cells. Together, they suggest that Msp1p/ATAD1 may regulate PMP stability.

In mammalian cells, ubiquitination has been shown to signal peroxisome degradation by an autophagy process called pexophagy. Peter Kim's group showed evidence of two factors that may regulate pexophagy. One is the deubiquitinating enzyme called USP30, a deubiquitinating enzyme best known for its role in preventing mitophagy.

Victoria Riccio (Peter Kim laboratory, Hospital for Sick Children, University of Toronto) presented data that shows that USP30 prevents pexophagy by removing the ubiquitin on PMPs. They also show that the overexpression of USP30 can prevent starvation induced pexophagy by working against PEX2, the E3 ubiquitin ligase for pexophagy.

A number of peroxins, such as PEX1, PEX2, PEX3 and PEX14, have been shown to be involved in pexophagy in a number of model systems. Since most peroxins are involved in import of matrix proteins into peroxisomes, Nick Demers (Peter Kim laboratory, Hospital for Sick Children, University of Toronto) asked whether any matrix protein import peroxins were involved in pexophagy. Using a siRNA screen, he found that the depletion of PEX13 resulted in the activation of ubiquitin mediated pexophagy. In addition, he reported that the depletion of PEX13 induces general autophagy in a ROS dependent manner. These findings suggest that PEX13 is a novel pexophagy regulator, where in its absence pexophagy as well as general autophagy are induced.

Peroxisome levels have been associated with various different cancers. While high levels of peroxisome-derived ether-lipids are correlative with the proliferative capacity and tumorigenic potential of certain tumor cells, peroxisome abundance is frequently reduced in human clear cell renal cell carcinomas (ccRCC). Interestingly, peroxisome abundance is reduced more frequently in well-differentiated ccRCCs. However, the impact of peroxisome metabolism on tumor progression is still poorly understood and it is unknown whether cancer is cause or consequence of peroxisome dysfunction. Tanja Eberhart (Werner Kovacs laboratory, ETH, Zurich) set out to determine the role of decreased peroxisome abundance on tumorigenesis by establishing ccRCC cell lines where pexophagy is triggered by inducible ubiquitin signalling, leading to cells characterized by a few peroxisomal aggregates or complete lack of peroxisomes. Using these cell lines they xenograft them into mice to investigate the impact of peroxisome deficiency on tumor progression *in vivo*.

Session 5: signalling and regulation

Chair: Nicole Linka, Heinrich-Heine-University Düsseldorf, Germany

Matt Anderson-Baron (Andrew J. Simmond laboratory, Department of Cell Biology, University of Alberta) introduced that components of the peroxisome interact with lipid droplets during periods of elevated lipid metabolism in *Drosophila melanogaster*. This interaction is necessary to regulate the level of lipolysis, the process by which fatty acids are cleaved from triglyceride stores in the lipid droplets. In his talk, Matt presented that in *Drosophila* S2 cells, Pex14 localizes to the surface of lipid droplets under certain metabolic conditions. Pex14 functions to regulate lipolysis by blocking the lipase, Hsl, from accessing the lipid droplet surface. He also showed that the lipid storage droplet 1 (Lsd1), the regulator of lipolysis in *Drosophila*, blocks the recruitment of Pex14 to the lipid droplet. Besides Pex14, other Pex proteins, like Pex3 and Pex13, localize to the surface of lipid droplets under the same conditions, indicating that peroxisomes are able to influence the mobilization of lipid droplets.

Using the model *C. elegans*, Elisabeth Rackles (Stephane G. Rolland laboratory, Department of Cell and Development Biology, Ludwig Maximilian University Munich) investigated a peroxisomal stress response which is induced upon perturbation of peroxisomal biogenesis. Quality control of other organelles (such as mitochondria or ER) has been shown to involve a retrograde signalling from the organelle to the nucleus to activate the production of organelle-specific proteases and chaperones in response to organelle-specific stress. Her results indicate that peroxisomal stress also induces a retrograde signalling which leads to the transcriptional up-regulation of peroxisomal quality control genes.

Evidence that peroxisomes are involved in immune defense was presented by Francesca Di Cara from the Department of Cell Biology of the University of Alberta in Canada (now Assistant Professor at the Department of Microbiology & Immunology of the Dalhousie University in Canada).

In the model organism *Drosophila melanogaster* peroxisomes are highly abundant in the gut epithelium. This tissue facilitates the absorption of nutrients, but also serves in defending against pathogens. Francesca showed that dysfunctional peroxisomes cause lipotoxicity and redox unbalance in these epithelial cells, which in turn promotes Tor kinase-dependent autophagy. As a consequence of an increased cell death the epithelial cell layer gets unstable and more susceptible to pathogens, indicating that the immune defense is compromised in the gut (Caraet al. 2018).

Julia Sellin and Margret Bülow from the Life and Medical Sciences Institute at the University of Bonn in Germany presented results obtained in a joint project. They could show that in the absence of peroxisomes, like in Drosophila *Pex19* mutants, lipid metabolism is dysregulated. In this situation the Lipase 3 is specifically up-regulated. The increased lipase activity leads to an accumulation of free medium and long chain fatty acids (but not very long chain fatty acids), that in turn causes mitochondrial damage. Julia presented

evidence that the increased Lipase 3 activity is due to the action of two transcription factors. Hnf4, the *Drosophila* master regulator of lipolysis, is hyperactive and as a result induces Lipase 3 expression (Bulow et al. 2018). In contrast, Schlank, a new type of transcriptional factor, functions as repressor for the Lipase 3 gene. In the Pex19 mutant, Schlank is excluded from the nucleus and localized to the ER membrane, where it fulfills its enzymatic function as ceramide synthase (Sellin et al. 2018).

Conclusion of the meeting

After a short intermezzo, in which the participants were challenged on their knowledge of the *PEX* gene nomenclature, the prizes for the best oral and poster presentations were awarded. Eden Yifrach (Weizmann Institute) won the prize for the best poster and Juan Fransisco Ridriguez Alcazar (University of Bonn) the prize for the best oral presentation. Finally, the next edition of OEPM, which will be organized by Ralf Erdmann (2020 in Germany), was announced.

Overall, the 6th OEPM was very successful in bringing together the peroxisome research community, offering ample opportunities for exchanging ideas and starting up new collaborations.

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