Purification of peroxisomes – a bibliography

There are several OptiPrep™ Application Sheets that are relevant to the isolation and analysis of peroxisomes using iodixanol gradients and three types of gradient have been used:

Continuous iodixanol gradients: principally for the purification of peroxisomes from mammalian liver, but they have also been used for organelles from fungi, mainly yeast and also from plants.
Discontinuous iodixanol gradients: for mammalian liver and kidney, some marine organisms and plants.
Self-generated iodixanol gradients: for mammalian liver and cultured cells

- **Application Sheet S11** describes the use of a pre-formed continuous iodixanol gradient
- **Application Sheet S12** describes the use of a discontinuous iodixanol gradient
- **Application Sheet S13** describes the use of a self-generated iodixanol gradient
- **Application Sheet S55** describes the use of a continuous iodixanol gradient for yeast peroxisomes

In addition there are two Application Sheets that are devoted to the analysis of the light mitochondrial fraction (LMF) and although these are not devoted specifically to peroxisomes, the latter are analyzed as part of a more general analysis of the LMF organelles, which include mitochondria, lysosomes and sometimes Golgi in addition to peroxisomes.

- **Application Sheet S15** describes the use of pre-formed continuous gradient
- **Application Sheet S16** describes the use of self-generated gradient

The bibliography below is divided into gradient type, then tissue or cell source. References are listed alphabetically according to **first author** and then, if required, chronologically. To aid identification of research topics, these are highlighted in blue.

1. Continuous gradients

1a. Brain (rodent)

Nawrotzki, R., Islinger, M., Vogel, I., Völkl, A. and Kirsch, J. (2012) *Expression and subcellular distribution of gephyrin in non-neuronal tissues and cells* Histochem. Cell. Biol., **137**, 471–482

1b. Fat pad (mammary)

Vapola, M.H., Rokka, A., Sormunen, R.T., Alhonen, L., Schmitz W., Conzelmann, E., Wärri, A., Grunau, S., Antonenkov, V.D. and Hiltunen, J.K. (2014) *Peroxisomal membrane channel Pxmp2 in the mammary fat pad is essential for stromal lipid homeostasis and for development of mammary gland epithelium in mice* Dev. Biol., **391**, 66–80

1c. Fibroblasts

Wiesinger, C., Kunze, M., Regelsberger, G., Forss-Petter, S. and Berger, J. (2013) *Impaired very long-chain Acyl-CoA β-oxidation in human X-linked adrenoleukodystrophy fibroblasts is a direct consequence of ABCD1 transporter dyfunction* J. Biol. Chem., **288**, 19269-19279

1d. Fungi

1d-1 *Paracoccidioides brasiliensis*

Brito, W.deA., Rezende, T.C.V., Parente, A.F., Ricart, C.A.O., de Sousa, M.V., Báo, N. and Soares, C.M.deA. (2011) *Identification, characterization and regulation studies of the aconitase of Paracoccidioides brasiliensis* Fungal Biol., **115**, 697-707

1d-2 Yeast

Antonenkov, V.D., Mindthoff, S., Grunau, S., Erdmann, R. and Hiltunen, J.K. (2009) *An involvement of yeast peroxisomal channels in transmembrane transfer of glyoxylate cycle intermediates* Int., J. Biochem. Cell Biol., **41**, 2546–2554

Cramer, J., Effelsberg, D., Girzalsky, W. and Erdmann, R. (2015) *Isolation of peroxisomes from yeast* Cold Spring Harb. Protoc; doi:10.1101/pdb.top074500

Cramer, J., Effelsberg, D., Girzalsky, W. and Erdmann, R. (2015) *Small-scale purification of peroxisomes for analytical applications* Cold Spring Harb. Protoc; doi:10.1101/pdb.prot083717

Debelyy, M.O., Platta, H.W., Saffian, D., Hensel, A., Thoms, S., Meyer, H.E., Warscheid, B., Girzalsky, W. and Erdmann, R. (2011) *Ubp15p, a ubiquitin hydrolase associated with the peroxisomal export machinery* J. Biol. Chem., **286**, 28223–28234

Effelsberg, D., Cruz-Zaragoza, L.D, Tonillo, J., Schliebs, W. and Erdmann, R. (2015) *Role of Pex21p for piggyback import of Gpd1p and Pnc1p into peroxisomes of Saccharomyces cerevisiae* J. Biol. Chem., **290**, 25333–25342

Einwachter, H., Sowinski, S., Kunau, W-H. and Schliebs, W. (2001) *Yarrowia lipolytica Pex20p, Saccharomyces cerevisiae Pex18p/Pex 21p and mammalian Pex5pL fulfil a common function in the early steps of the peroxisomal PTS2 import pathway* EMBO Rep., **2**, 1035-1039

Grunau, S., Mindthoff, S., Rottensteiner, H., Sormunen, R.T., Hiltunen, J.K., Erdmann, R. and Antonenkov, V.D. (2009) *Channel-forming activities of peroxisomal membrane proteins from the yeast Saccharomyces cerevisiae* FEBS J., **276**, 1698–1708

Grunau, S., Lay, D., Mindthoff, S., Platta, H.W., Girzalsky, W., Just, W.W. and Erdmann, R. (2011) *The phosphoinositide 3-kinase Vps34p is required for pexophagy in Saccharomyces cerevisiae* Biochem. J. **434**, 161–170

Kerssen, D., Hambruch, E., Klaas, W., Platta, H.W., de Kruijff, B., Erdmann, R., Kunau, W-H. and Schleibs, W. *Membrane association of the cycling peroxisome import receptor Pex5p* J. Biol. Chem., **281**, 27003-27015

Mindthoff, S., Grunau, S., Steinfort, L.L., Girzalsky, W., Hiltunen, J.K., Erdmann, R. and Antonenkov, V.D. (2016) *Peroxisomal Pex11 is a pore-forming protein homologous to TRPM channels* Biochim. Biophys. Acta, **1863**, 271–283

Oeljeklaus, S., Reinartz, B.S., Wolf, J., Wiese, S., Tonillo, J., Podwojski, K., Kuhlmann, K., Stephan, C., Meyer, H.E., Schliebs, W., Brocard, C., Erdmann, R. and Warscheid, B. (2012) *Identification of core components and transient interactors of the peroxisomal importomer by dual-track stable isotope labeling with amino acids in cell culture analysis* J. Proteome Res. 2012, 11, 2567−2580

Platta, H.W., Grunau, S., Rosenkrantz, K., Girzalsky, W. and Erdmann, R. (2005) *Functional role of the AAA peroxins in dislocation of the cycling PTS1 receptor back to the cytosol* Nat. Cell Biol., **7**, 817-822

Schäfer, A., Kerssen, D., Veenhuis, M., Kunau, W-H. and Schliebs, W. (2004) *Functional similarity between the peroxisomal PTS2 receptor binding protein Pex18p and the N-terminal half of the PTS1 receptor Pex5p* Mol. Cell Biol., **24**, 8895-8906

Thoms, S., Debelyy, M.O., Nau, K., Meyer, H.E. and Erdmann, R. (2008) *Lpx1p is a peroxisomal lipase required for normal peroxisome morphology* FEBS J., **275**, 504-514

Welker, S., Rudolph, B., Frenzel, E., Hagn, F., Liebisch, G., Schmitz, G., Scheuring, J., Kerth, A., Blume, A., Weinkauf, S., Haslbeck, M., Kessler, H. and Buchner, J. (2010) *Hsp12 is an intrinsically unstructured stress protein that folds upon membrane association and modulates membrane function* Mol. Cell, **39**, 507–520

1e. Liver (rodent)

Antonenkov, V.D., Sormunen, R.T. and Hiltunen, J.K. (2004) *The behavior of peroxisomes in vitro: mammalian peroxisomes are osmotically sensitive particles* Am. J. Physiol., **287**, C1623-C16350

Antonenkov, V.D., Rokka, A., Sormunen, R.T., Benz, R. and Hiltunen, J.K. (2005) *Solute traffic across mammalian peroxisomal membrane – single-channel conductance monitoring reveals pore-forming activities in peroxisomes* Cell. Mol. Life Sci., **62**, 2886-2895

Antonenkov, V.D., Sormunen, R.T., Ohlmeier, S., Amery, L., Fransen, M., Mannaerts, G.P. and Hiltunen, J.K. (2006) *Localization of a portion of the liver isoform of fatty-acid-binding protein (L-FABP) to peroxisomes* Biochem. J., **394**, 475-484

Antonenkov, V.D., Ohlmeier, S., Sormunen, R.T. and Hiltunen, J.K. (2007) *UK114, a YjgF/Yer057p/UK114 family protein highly conserved from bacteria to mammals, is localized in rat liver peroxisomes* Biochem. Biophys. Res. Commun., **357**, 252-257

Gijsbers, S., Van der Hoeven, G. and Van Veldhoven, P.P. (2001) *Subcellular study of sphingoid base phosphorylation in rat tissues: evidence for multiple sphingosine kinases* Biochim. Biophys. Acta, **1532**, 37-50

Graham, J., Ford, T. and Rickwood, D (1994) *The preparation of subcellular organelles from mouse liver in self-generated gradients of iodixanol* Anal. Biochem., **220**, 367-373

Islinger, M., Lüers, G.H., Zischka, H., Ueffing, M. and Völkl, A. (2006) *Insights into the membrane proteome of rat liver peroxisomes: Microsomal glutathione-S-transferase is shared by both subcellular compartments* Proteomics, **6,** 804-816
Islinger, M. and Weber, G. (2008) *Free flow isoelectric focusing: a method for the separation of both hydrophilic and hydrophobic proteins of rat liver peroxisomes* In Methods Mol. Biol., **432**, Organelle Proteomics (ed. Pflieger, D, and Rossier, J.) Humana Press, Totowa, NJ, pp 199-215
Islinger, M., Li, K.W., Seitz, J., Völkl, A. and Lüers, G.H. (2009) *Hitchhiking of Cu/Zn superoxide dismutase to peroxisomes – evidence for a natural piggyback import mechanism in mammals* Traffic, **10**, 1711–1721
Islinger, M., Li, K.W., Loos, M., Liebler, S., Angermüller, S., Eckerskorn, C., Weber, G., Abdolzade, A. and Völkl, A. (2010) *Peroxisomes from the heavy mitochondrial fraction: isolation by zonal free flow electrophoresis and quantitative mass spectrometrical characterization* J. Proteome Res., **9**, 113–124
Islinger, M., Abdolzade-Bavil, A., Liebler, S., Weber, G. and Völkl, A. (2012) *Assessing heterogeneity of peroxisomes: isolation of two subpopulations from rat liver* In Liver Proteomics: Methods and Protocols, Methods Mol. Biol., **909** (eds Josic, D. and Hixson, D.C.) Springer Science+Business Media, New York 2012
Joly, E., Bendyan, M., Roduit, R., Saha, A.K., Ruderman, N.B. and Prentki, M. (2005) *Malonyl-CoA decarboxylase is present in the cytosolic, mitochondrial and peroxisomal compartments of rat hepatocytes* FEBS Lett., **579**, 6581-6586
Koch, A., Thiemann, M., Grabenbauer, M., Yoon, Y., McNiven, M.A. and Schrader, M. (2003) *Dynamin-like protein 1 is involved in peroxisomal fission* J. Biol. Chem., **278**, 8597-8605
Koch, A., Yoon, Y., Bonekamp, N.A., McNiven, M.A. and Schrader, M. (2005) *A role for Fis1 in both mitochondrial and peroxisomal fission in mammalian cells* Mol. Biol. Cell, **16**, 5077-5086
Lewin T.M., Van Horn, C.G., Krisans, S.K. and Coelman, R.A. (2002) *Rat liver acyl-CoA synthetase 4 is a peripheral-membrane protein located in two distinct subcellular organelles, peroxisomes and mitochondrial-associated membranes* Arch. Biochem. Biophys., **404**, 263-270
Mustacich, D.J., Leonard, S.W., Patel N.K. and Traber, M.G. (2010) *α-Tocopherol β-oxidation localized to rat liver mitochondria* Free Radic. Biol. Med., **48**, 73–81
Nawrotzki, R., Islinger, M., Vogel, I., Völkl, A. and Kirsch, J. (2012) *Expression and subcellular distribution of gephyrin in non-neuronal tissues and cells* Histochem. Cell. Biol., **137**, 471–482
Priore, P., Giudetti, A.M., Natali, F., Gnoni, G.V. and Geelen, M.J.H. (2007) *Metabolism and short-term metabolic effects of conjugated linoleic acids in rat hepatocytes* Biochim. Biophys. Acta, **1771**, 1299-1307
Van Veldhoven, P.P., Baumgart, E. and Mannaerts, G.P. (1996) *Iodixanol (OptiPrep), an improved density gradient medium for the iso-osmotic isolation of rat liver peroxisomes* Anal. Biochem. **237**, 17-23
Westin, M.A.K., Hunt, M.C. and Alexson, S.E.H. (2007) *Peroxisomes contain a specific phytanoyl-CoA/Pristanoyl-CoA thioesterase acting as a novel auxiliary enzyme in α- and β-oxidation of methyl-branched fatty acids in mouse* J. Biol. Chem., **282**, 26707-26716
Zhang, D., Yu, W., Geisbrecht, B.V., Gould, S.J., Sprecher, H. and Schulz, H. (2002) *Functional characterization of Δ^3, Δ^2-enoyl-CoA isomerase from rat liver* J. Biol. Chem., **277**, 9127-9132

1f. Mammary fat pad

Vapola, M.H., Rokka, A., Sormunen, R.T., Alhonen, L., Schmitz W., Conzelmann, E., Wärri, A., Grunau, S., Antonenkov, V.D. and Hiltunen, J.K. (2014) *Peroxisomal membrane channel Pxmp2 in the mammary fat pad is essential for stromal lipid homeostasis and for development of mammary gland epithelium in mice* Dev. Biol., 391, 66–80

1g. Plant tissues

Arai, Y., Hayashi, M. and Nishimura, M. (2008) *Proteomic analysis of highly purified peroxisomes from etiolated Soybean cotyledons* Plant Cell Physiol., **49**, 526-539
Hossain, Z. and Komatsu, S. (2014) *Soybean proteomics* In Plant Proteomics: Methods Mol. Biol., **1072** (ed. Jorrin-Novo, J.V. et al), Springer Science+Business Media, LLC, pp 315-331
Komatsu, S. and Ahsan, N. (2009) *Soybean proteomics and its application to functional analysis* J. Proteom., **72**, 325-336
Palma, J.M., Corpas, F.J. and del Rio, L.A. (2009) *Proteome of plant peroxisomes: new perspectives on the role of these organelles in cell biology* Proteomics, **9**, 2301-2312
Reumann, S. (2011) *Toward a definition of the complete proteome of plant peroxisomes: Where experimental proteomics must be complemented by bioinformatics* Proteomics **11**, 1764–1779

1h. Review

Antonenkov, V.D. and Hiltunen, J.K. (2006) *Peroxisomal membrane permeability and solute transfer* Biochim. Biophys. Acta, Mol. Cell Res., **1763**, 1697-1706

2. Discontinuous gradients

2a. HEK cells

Ge, L., Melville, D., Zhang, M. and Schekman, R. (2013) *The ER–Golgi intermediate compartment is a key membrane source for the LC3 lipidation step of autophagosome biogenesis* eLife, **2**: e00947

Zhang, J., Kim, J., Alexander, A., Cai, S., Tripathi, D.N., Dere, R., Tee, A.R., Tait-Mulder, J., Di Nardo, A., Han, J.M., Kwiatkowski, E., Dunlop, E.A., Dodd, K.M., Folkerth, R.D., Faust, P.L., Kastan, M.B., Sahin, M. and Walker, C.L. (2013) *A tuberous sclerosis complex signalling node at the peroxisome regulates mTORC1 and autophagy in response to ROS* Nat. Cell Biol., 15, 1186-1196

2b. Human fibroblasts

Beltran, P.M.J., Mathias, R.A. and Cristea, I.M. (2016) *A portrait of the human organelle proteome in space and time during cytomegalovirus infection* Cell Systems **3**, 361–373

2c. Kidney (rodent)

Mi, J., Garcia-Arcos, I., Alvarez, R., and Cristobal, S. (2007) *Age-related subproteomic analysis of mouse liver and kidney peroxisomes* Proteome Sci., **5:19**

Mi, J., Kirchner, E. and Cristobal, S. (2007) *Quantitative proteomic comparison of mouse peroxisomes from liver and kidney* Proteomics, **7**, 1916-1928

2d. Liver (chick embryo)

Labitzke, E.M., Diani-Moore, S. and Rifkind, A.B. (2007) *Mitochondrial P450-dependent arachidonic acid metabolism by TCDD-induced hepatic CYP1A5; conversion of EETs to DHETs by mitochondrial soluble epoxide hydrolase* Arch. Biochem. Biophys., **468**, 70-81

2e. Liver (rodent)

Amelina, H., Sjödin, M.O.D., Bergquist, J. and Cristobal, S. (2011) *Quantitative subproteomic analysis of age-related changes in mouse liver peroxisomes by iTRAQ LC–MS/MS* J.Chromatogr. B, **879**, 3393– 3400

Grant, P., Ahlemeyer, B., Karnati, S., Berg, T., Stelzig, I., Nenicu, A., Kuchelmeister, K., Crane, D.I. and Baumgart-Vogt, E. (2013) *The biogenesis protein PEX14 is an optimal marker for the identification and localization of peroxisomes in different cell types, tissues, and species in morphological studies* Histochem. Cell. Biol., **140**, 423–442

Karnati, S., Lüers, G., Pfreimer, S. and Baumgart-Vogt, E. (2013) *Mammalian SOD2 is exclusively located in mitochondria and not present in peroxisomes* Histochem. Cell Biol., **140**, 105–117

Lamhonwah, A-M., Skaug, J., Scherer, S. and Tein, I. (2003) *A third human carnitine/organic cation transporter (OCTN3) as a candidate for the 5q31 Crohn's disease locus (IBD5)* Biochem. Biophys. Res. Commun., **301**, 98-101

Mi, J., Garcia-Arcos, I., Alvarez, R., and Cristobal, S. (2007) *Age-related subproteomic analysis of mouse liver and kidney peroxisomes* Proteome Sci., **5:19**

Mi, J., Kirchner, E. and Cristobal, S. (2007) *Quantitative proteomic comparison of mouse peroxisomes from liver and kidney* Proteomics, **7**, 1916-1928

Salvi, M., Battaglia, V., Brunati, A.M., La Rocca, N., Tibaldi, E., Pietrangeli, P., Marcocci, L., Mondovi, B., Rossi, C.A. and Toninello, A. (2007) *Catalase takes part in rat liver mitochondria oxidative stress defense* J. Biol. Chem., **282**, 24407-24415

Spinazzola, A., Viscomi, C., Fernandez-Vizarra, E., Carrara, F., D'Adamo, P., Calvo, S., Marsano, R.M., Donnini, C., Weiher, H., Strisciuglio, P., Parini, R., Sarzi, E., Chan, A., DiMauro, S., Rotig, A., Gasparini, P., Ferrero, I., Mootha, V.K., Tiranti, V. and Zeviani, M. (2006) *MPV17 encodes an inner mitochondrial membrane protein and is mutated in infantile hepatic mitochondrial DNA depletion* Nat. Genet., **38**, 570-575

Weng, H., Ji, X., Naito, Y., Endo, K., Ma, X., Takahashi, R., Shen, C., Hirokawa, G., Fukushima, Y. and Iwai, N. (2013) *Pex11α deficiency impairs peroxisome elongation and division and contributes to nonalcoholic fatty liver in mice* Am. J. Physiol. Endocrinol. Metab., **304**, E187–E196

2f. Mouse embryo fibroblasts

Zhang, J., Kim, J., Alexander, A., Cai, S., Tripathi, D.N., Dere, R., Tee, A.R., Tait-Mulder, J., Di Nardo, A., Han, J.M., Kwiatkowski, E., Dunlop, E.A., Dodd, K.M., Folkerth, R.D., Faust, P.L., Kastan, M.B., Sahin, M. and Walker, C.L. (2013) *A tuberous sclerosis complex signalling node at the peroxisome regulates mTORC1 and autophagy in response to ROS* Nat. Cell Biol., 15, 1186-1196

2g. Mussels

Apraiz, I., Mi, J. and Cristobal, S. (2006) *Identification of proteomic signatures of exposure to marine pollutants in mussels (Mytilus edulis)* Mol. Cell. Proteom., **5**, 1274-1285
Apraiz, I., Cajaraville, M.P. and Cristobal, S. (2009) *Peroxisomal proteomics: Biomonitoring in mussels after the Prestige's oil spill* Mar. Pollut. Bull., **58**, 1815–1826
Cristobal, S. (2007) *Proteomics-based method for risk assessment of peroxisome proliferating pollutants in the marine environment* Methods Mol. Biol., **410**, 123-135
Mi, J., Orbea, A., Syme, N., Ahmed, M., Cajaraville, M.P. and Cristobal, S. (2005) *Peroxisomal proteomics, a new tool for risk assessment of peroxisome proliferating pollutants in the marine environment* Proteomics, **5**, 3954-2965

2h. Yeast

Nyathi, Y., De Marcos Lousa, C., van Roermund, C.W., Wanders, R.J.A., Johnson, B., Baldwin, S.A., Theodoulou, F.L. and Baker, A. (2010) *The Arabidopsis peroxisomal ABC transporter, Comatose, complements the Saccharomyces cerevisiae pxa1 pxa2Δ mutant for metabolism of long-chain fatty acids and exhibits fatty acyl-CoA-stimulated ATPase activity* J. Biol., Chem., **285**, 29892–29902
Nyathi, Y., Zhang, X., Baldwin, J.M., Bernhardt, K., Johnson, B., Baldwin, S.A., Theodoulou, F.L. and Baker, A. (2012) *Pseudo half-molecules of the ABC transporter, COMATOSE, bind Pex19 and target to peroxisomes independently but are both required for activity* FEBS Lett., **586**, 2280–2286

3. Self-generated gradient

3a. CHO cells

Honsho, M., Yagita, Y., Kinoshita, N. and Fujuki, Y. (2008) *Isolation and characterization of mutant animal cell line defective in alkyl-dihydroxyacetonephosphate synthase: Localization and transport of plasmalogens to post-Golgi compartments* Biochim. Biophys. Acta, **1783**, 1857-1865
Kobayashi, S., Tanaka, A. and Fujiki, Y. (2007) *Fis1, DLP1 and Pex11p coordinately regulate peroxisome morphogenesis* Exp. Cell Res., **313**, 1675-1686
Matsuzaki, T. and Fujiki, Y. (2008) *The peroxisomal membrane protein import receptor Pex3p is directly transported to peroxisomes by a novel Pex19p- and Pex16p-dependent pathway* J. Cell Biol. **183**, 1275–1286

3b. Hep-G2 cells

Morel, F., Rauch, C., Petit, E., Piton, A., Theret, N., Coles, B. and Guillouzo, A. (2004) *Gene and protein characterization of the human glutathione S-transferase kappa and evidence for a peroxisomal localization* J. Biol. Chem., **279**, 16246-16253

3c. Liver (rodent)

He, D., Barnes, S. and Falany, C.N. (2003) *Rat liver bile acid CoA:amino acid N-acyltransferase: expression, characterization, and peroxisomal localization* J. Lipid Res., **44**, 2242-2249
Graham, J., Ford, T. and Rickwood, D (1994) *The preparation of subcellular organelles from mouse liver in self-generated gradients of iodixanol* Anal. Biochem., **220**, 367-373
Kurochkin, I.V., Mizuno, Y., Konagaya, A., Sakaki, Y., Schonbach, C. and Okazaki, Y. (2007) *Novel peroxisomal protease Tysnd1 processes PTS1- and PTS2-containing enzymes involved in β-oxidation of fatty acids* EMBO J., **26**, 835-845
McClelland, G.B., Khanna, S., Gonzales, G.F., Butz, C.E. and Brooks, G.A. (2003) *Peroxisomal membrane monocarboxylate transporters: evidence for a redox shuttle system* Biochem. Biophys. Res. Commun., **304**, 130-135
Miyata, N., Hosoi, K-i., Mukai, S. and Fujiki, Y. (2009) *In vitro import of peroxisome-targeting signal type 2 (PTS2) receptor Pex7p into peroxisomes* Biochim. Biophys. Acta, **1793**, 860–870
Styles, N.A., Falany, J.L., Barnes, S. and Falany, C.N. (2007) *Quantification and regulation of the subcellular distribution of bile acid coenzyme A:amino acid N-acyltransferase activity in rat liver* J. Lipid Res., **48**, 1305-1315

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