

SUPPLEMENTAL DATA

PPAR α -INDEPENDENT ACTIVATION OF THE SREBP-2 PATHWAY IN NEWBORN PEROXISOME-DEFICIENT *PEX2* KNOCK-OUT MICE

Werner J. Kovacs^{1,2,3}, Khanichi N. Charles^{2,7}, Katharina M. Walter^{1,3}, Janis E. Shackelford², Thomas M. Wikander⁴, Michael J. Richards⁵, Steven J. Fliesler^{5,6}, Skaidrite K. Krisans² and Phyllis L. Faust⁴

Table S1. Specific activity of HMGCR normalized to HMGCR protein content in the livers of P0 129 control and *Pex2*^{-/-} mice

Genotype	Sp. act. (pmol/min/mg)	Vol. density (per mg of protein)	Sp. act./vol. density (fmol/min)	Normalized average sp. act./vol. density
+/+	141	224	631	
+/+	326	581	561	
+/+	215	1064	202	
+/-	423	919	460	1.0 ± 0.4
+/-	386	1061	364	
+/-	228	744	306	
-/-	511	2063	248	
-/-	596	2067	288	
-/-	401	1958	205	0.6 ± 0.1 *
-/-	718	3589	200	

Measurements of HMGCR activities and immunoblots were done on the same liver samples. For Western blot analysis, 200 µg of protein was separated on a SDS-7.5% polyacrylamide gel, transferred to a nitrocellulose membrane, and subjected to immunoblot analysis using a polyclonal anti-HMGCR antibody. *, $P = 0.05$ (Student's *t*-test). Sp. act., specific activity.

Table S2. Specific activity of IDI1 normalized to IDI1 protein content in the livers of P0 129 control and *Pex2*^{-/-} mice

Genotype	Sp. act. (pmol/min/mg)	Vol. density (per mg of protein)	Sp. act./vol. density (fmol/min)	Normalized average sp. act./vol. density
+/+	1037	377	2753	
+/+	1894	696	2722	
+/+	1706	1004	1699	
+/-	2551	892	2860	1.0 ± 0.2
+/-	2509	790	3177	
+/-	1778	566	3142	
-/-	2718	1931	1408	
-/-	3568	3500	1019	
-/-	3180	2251	1413	0.4 ± 0.1 *
-/-	3002	3572	840	

Measurements of IDI1 activities and immunoblots were done on the same liver samples. For Western blot analysis, 200 µg of protein was separated on a SDS-13% polyacrylamide gel, transferred to a nitrocellulose membrane, and subjected to immunoblot analysis using a polyclonal anti-IDI1 antibody. *, $P < 0.001$ (Student's *t*-test). Sp. act., specific activity.

Table S3. Relative expression of SREBP-regulated genes in the liver of P0 control and *Pex2*^{-/-} mice on the 129 and SW/129 genetic background

Gene	129		SW/129	
	Control	<i>Pex2</i> ^{-/-}	Control	<i>Pex2</i> ^{-/-}
HMGCR	1.00 ± 0.19	3.60 ± 1.20 **	1.00 ± 0.31	4.36 ± 1.54 *
FPPS	1.00 ± 0.61	2.25 ± 0.99 *	1.00 ± 0.25	1.97 ± 1.28
SREBP-2	1.00 ± 0.20	1.62 ± 0.20 ***	1.00 ± 0.13	2.05 ± 0.58 *
SREBP-1c	1.00 ± 0.11	1.73 ± 0.20 ***	1.00 ± 0.31	0.39 ± 0.15 *
Insig-1	1.00 ± 0.43	3.51 ± 1.73 *	1.00 ± 0.22	2.06 ± 0.97
Insig-2a	1.00 ± 0.75	1.63 ± 0.87	1.00 ± 0.53	5.19 ± 1.12 **

Each value represents the amount of mRNA relative to that in control mice, which was arbitrarily defined as 1. Values are mean ± S.D. from RNA samples of six (129 genetic background) and four (SW/129 genetic background) mice. *, *P* < 0.05; **, *P* < 0.01; ***, *P* < 0.001 vs. control mice (Student's *t*-test). Expression data obtained from mice on the SW/129 genetic background are reproduced, with permission, from Kovacs *et al.* [15], © the American Society for Biochemistry and Molecular Biology. [*J. Biol. Chem.* 2009; 284:7232-7245.]

Table S4. Relative expression of ER stress markers in the liver of P0 control and *Pex2*^{-/-} mice on the 129 and SW/129 genetic background

Gene	129		SW/129	
	Control	<i>Pex2</i> ^{-/-}	Control	<i>Pex2</i> ^{-/-}
Herpud1	1.00 ± 0.41	1.27 ± 0.26	1.00 ± 0.14	3.54 ± 0.10 ***
GADD45	1.00 ± 0.22	2.15 ± 0.90 *	1.00 ± 0.18	0.91 ± 0.06
Grp78	1.00 ± 0.10	1.22 ± 0.29	1.00 ± 0.11	0.82 ± 0.30
Grp94	1.00 ± 0.13	1.02 ± 0.20	1.00 ± 0.06	0.65 ± 0.21 *
p58 ^{IPK}	1.00 ± 0.39	0.91 ± 0.21	1.00 ± 0.25	1.28 ± 0.31
IRE1α	1.00 ± 0.24	1.62 ± 0.46 *	1.00 ± 0.26	2.15 ± 0.20 ***
p8	1.00 ± 0.22	7.40 ± 3.43 **	1.00 ± 0.34	11.18 ± 2.63 ***
ATF4	1.00 ± 0.08	1.62 ± 0.25 ***	1.00 ± 0.26	1.32 ± 0.29
CHOP	1.00 ± 0.28	1.10 ± 0.32	1.00 ± 0.23	1.67 ± 0.31 *
ATF3	1.00 ± 0.84	9.58 ± 8.99 *	1.00 ± 0.04	15.70 ± 8.52 *
TRIB3	1.00 ± 0.32	4.33 ± 1.69 **	1.00 ± 0.27	9.77 ± 3.97 **

Each value represents the amount of mRNA relative to that in control mice, which was arbitrarily defined as 1. Values are mean ± S.D. from RNA samples of six (129 genetic background) and four (SW/129 genetic background) mice. *, *P* < 0.05; **, *P* < 0.01; ***, *P* < 0.001 vs. control mice (Student's *t*-test). Expression data obtained from mice on the SW/129 genetic background are reproduced, with permission, from Kovacs *et al.* [15], © the American Society for Biochemistry and Molecular Biology. [*J. Biol. Chem.* 2009; 284:7232-7245.]