SUPPLEMENTAL DATA

Table S I
Table S II
Table S III
Table S IV

Table S I. Strains and plasmids used in the yeast experiments

Strain	Relevant Genotype	Source or reference
BY4742	Mat**α** his3Δ1 leu2Δ0 lys2Δ0 ura3Δ0	Open Biosystems (Brachmann et al, 1998)
BY4741	Mat**a** his3Δ1 leu2Δ0 met15Δ0 ura3Δ0	Open Biosystems (Brachmann et al, 1998)
BY4743	BY4741/BY4742	Open Biosystems (Brachmann et al, 1998)
BY4739-10399	*spo7*Δ*::Kan*^{r} (in BY4739)	Open Biosystems (Winzeler et al, 1999)
BY4742-16601	*nem1*Δ*:: Kan*^{r} (in BY4742)	Open Biosystems (Winzeler et al, 1999)
SH003	*spo7Δ:: HIS3* (in BY4742)	This study
SH004	*nem1*Δ*:: Kan*^{r} *spo7Δ:: HIS3* (in BY4742)	This study
SH005	*spo7Δ:: HIS3* (in BY4741)	This study
SH009	Diploid spo7Δ generated by mating haploid *spo7*Δ (SH003 and SH005)	This study
Plasmid	**Relevant characteristics**	**Source or reference**
pRS313	Yeast centromeric plasmid (YCp) with *HIS3* marker	(Sikorski & Hieter, 1989)
pRS315	Yeast centromeric plasmid (YCp) with *LEU2* marker	(Sikorski & Hieter, 1989)
pRS316	Yeast centromeric plasmid (YCp) with *URA3* marker	(Sikorski & Hieter, 1989)
pRS317	Yeast centromeric plasmid (YCp) with *LYS2* marker	(Sikorski & Boeke, 1991)
pRS315-PGK1	pRS315 containing *PGK1* promoter and terminator	(Binns et al, 2006)
pRS316-PGK1	pRS316 containing *PGK1* promoter and terminator	(Binns et al, 2006)
ID: 5266084	Full length cDNA clone encoding NEP1-R1 (accession: BC036683)	Open Biosystems
ID: 4123279	cDNA clone encoding CTDNEP1 (accession: BC009295)	Open Biosystems
pRS315-PGK1-NEP1-R1	pRS315 expressing NEP1-R1 under control of *PGK1* promoter/terminator	This study
pRS316-PGK1-CTDNEP1	pRS316 expressing CTDNEP1 under control of *PGK1* promoter/terminator	This study
BG1805	MORF (movable ORF) vector, which was used as a template for HA-protA construct	(Gelperin et al, 2005)
pRS317-PGK1-Kar2-CFP-HDEL	pRS317 expressing ER luminal CFP marker subcloned from pRS315 PGK1-CFP-HDEL	(Szymanski et al, 2007) This study
pFA6-HygroMX	pFA6 vector containing *hph* (hygromycin drug cassettes)	(Longtine et al, 1998; Goldstein & McCusker, 1999) Gift from B.Tu lab
pFA6-CFP HygroMX	pFA6 vector encoding CFP which can be used to insert CFP in C-terminal of target ORF with hygromycin drug cassettes (*hph*)	This study
pRS315-PGK1-	pRS315 expressing C-terminally tagged NEP1-R1-	(Szymanski et al, 2007)

NEP1-R1-HA-protA	HA-protA under control of *PGK1* promoter/terminator	This study
pRS316-PGK1-CTDNEP1-2xFLAG	pRS315 expressing C-terminally tagged CTDNEP1-2xFLAG under control of *PGK1* promoter/terminator	This study
pRS315-PGK1-SPO7	pRS315 expressing Spo7p under control of *PGK1* promoter/terminator	This study
pRS317-PGK1-SEC63-CFP	pRS317 expressing Sec63p under control of *PGK1* promoter and *ADH1* terminator	This study

Binns D, Januszewski T, Chen Y, Hill J, Markin VS, Zhao Y, Gilpin C, Chapman KD, Anderson RG, Goodman JM (2006) An intimate collaboration between peroxisomes and lipid bodies. *J Cell Biol* **173:** 719-731

Brachmann CB, Davies A, Cost GJ, Caputo E, Li J, Hieter P, Boeke JD (1998) Designer deletion strains derived from Saccharomyces cerevisiae S288C: a useful set of strains and plasmids for PCR-mediated gene disruption and other applications. *Yeast* **14:** 115-132

Gelperin DM, White MA, Wilkinson ML, Kon Y, Kung LA, Wise KJ, Lopez-Hoyo N, Jiang L, Piccirillo S, Yu H, Gerstein M, Dumont ME, Phizicky EM, Snyder M, Grayhack EJ (2005) Biochemical and genetic analysis of the yeast proteome with a movable ORF collection. *Genes Dev* **19:** 2816-2826

Goldstein AL, McCusker JH (1999) Three new dominant drug resistance cassettes for gene disruption in Saccharomyces cerevisiae. *Yeast* **15:** 1541-1553

Longtine MS, McKenzie A, 3rd, Demarini DJ, Shah NG, Wach A, Brachat A, Philippsen P, Pringle JR (1998) Additional modules for versatile and economical PCR-based gene deletion and modification in Saccharomyces cerevisiae. *Yeast* **14:** 953-961

Sikorski RS, Boeke JD (1991) In vitro mutagenesis and plasmid shuffling: from cloned gene to mutant yeast. *Methods Enzymol* **194:** 302-318

Sikorski RS, Hieter P (1989) A system of shuttle vectors and yeast host strains designed for efficient manipulation of DNA in Saccharomyces cerevisiae. *Genetics* **122:** 19-27

Winzeler EA, Shoemaker DD, Astromoff A, Liang H, Anderson K, Andre B, Bangham R, Benito R, Boeke JD, Bussey H, Chu AM, Connelly C, Davis K, Dietrich F, Dow SW, El Bakkoury M, Foury F, Friend SH, Gentalen E, Giaever G, Hegemann JH, Jones T, Laub M, Liao H, Liebundguth N, Lockhart DJ, Lucau-Danila A, Lussier M, M'Rabet N, Menard P, Mittmann M, Pai C, Rebischung C, Revuelta JL, Riles L, Roberts CJ, Ross-MacDonald P, Scherens B, Snyder M, Sookhai-Mahadeo S, Storms RK, Veronneau S, Voet M, Volckaert G, Ward TR, Wysocki R, Yen GS, Yu K, Zimmermann K, Philippsen P, Johnston M, Davis RW (1999) Functional characterization of the S. cerevisiae genome by gene deletion and parallel analysis. *Science* **285:** 901-906

Table S II. Primers used in the yeast study

Primer name	Sequence [a]	Remark
XbaI_NEP1-R1_F	5′-GCTCTAGAATGAACTCGCTGGAGCAGGC-3′	NEP1-R1 subcloning
NEP1-R1_HindIII_R	5′-CCCAAGCTTTCATTGAACATGAGGCCTAGGTTTC-3′	
XbaI_CTDNEP1_F	5′-GCTCTAGAATGATGCGGACGCAGTGTC-3′	CTDNEP1 subcloning
CTDNEP1_HindIII_R	5′-CCCAAGCTTTCACCAGAGCCGATGTTGGT-3′	
XbaI_SPO7_F	5′-GCTCTAGAATGGAGCCAGAGAGCATAGG-3′	SPO7 cloning into pRS315-PGK1
SPO7_XmaI_R	5′-TATCCCGGGTCATTCTGATTTAGGTCGGA-3′	
XbaI_Nem1_F	5′-GCTCTAGAATGAATGCCCTAAAATATTTCTCAAATCA-3′	Nem1 cloning into pRS316-PGK1
Nem1_SalI_R	5′-CCCCGTCGACTCAGTTTATGTTGAATGCCTTCTCT-3′	
BamHI_HA-ProtA_F	5′-GGCGGATCCTATCCATACGATGTTCCTGA-3′	For subcloning HA-protA into pRS315-PGK1
HA-ProtA-PstI_R	5′-GGGCTGCAGTCACTGATGATTCGCGTCT-3′	
5phos HindIII-2xFLAG_F	5′Phos-AGCTTGATTATAAAGATGACGATGACAAGGATTATAAAGATGACGATGACAAGTAAG-3′	5'end is phosphorylated. Hybridized each other to make ds DNA
5phos 2xFLAG-SalI_R	5'Phos-TCGACTTACTTGTCATCGTCATCTTTATAATCCTTGTCATCGTCATCTTTATAATCA-3′	
NEP1-R1 C-ter BamHI_R	5′-GCGGGATCCTTGAACATGAGGCCTAGGTTTCAAA-3′	For NEP1-R1-HA-protA construct
CTDNEP1 C-ter HindIII_R	5′-CCCAAGCTTCCAGAGCCGATGTTGGTGAA-3′	For CTDNEP1-2xFlag construct
CTDNEP1 D67N_F [b]	5'-GGAAGATCCTGGTGCTGAATCTGGATGAGACACTT-3'	(Asn to Asp)
CTDNEP1 D67N_R [b]	5'-AAGTGTCTCATCCAGATTCAGCACCAGGATCTTCC-3'	
PacI-C-ter CFP_F1	5′-CACCTTAATTAACGTGAGCAAGGGCGAGGAGC-3′	Used for pFA6-CFP-HygroMX
C-ter CFP-AscI_R1	5′-TGGCGCGCCTTACTTGTACAGCTCGTCCATGC-3′	
SEC63-KI_F [c]	*5′*-**CGATACGGATACAGAAGCTGAAGATGATGAATCACCAGAA**CGGATCCCCGGGTTAATTAA-3′	Used for CFP tagging into SEC63 C-terminus
SEC63-KI_R [c]	*5′*-**TCTAAGAGCTAAAATGAAAAACTATACTAATCACTTATAT**GAATTCGAGCTCGTTTAAAC-3′	
BglII_SEC63_F	5′-CACCAGATCTATGCCTACAAATTACGAGTATG-3′	
CFP-BglII_R	5'-CACCAGATCTTTACTTGTACAGCTCGTCCATGC-3′	

a: Restriction enzyme sites are underlined.
b: Site directed mutated sequences are colored red.
c: The upstream or downstream sequence of the stop codon of SEC63 are in bold.

Table S III. Oligonucleotide primers used for real-time PCR quantitation of gene expression. m, mouse; h, human; f, forward; r, reverse; TBP, TATA box binding protein; HPRT, hypoxanthine phosphoribosyltransferase; B2M, beta-2-microglobulin.

Primer	Sequence
mLipin-1	5′-CCTTCTATGCTGCTTTTGGGAACC-3′ 5′-GTGATCGACCACTTCGCAGAGC-3′
mNEP1-R1	5′-AGCTGCTCGATGTCGAACGGTA-3′ 5′-CCGTCACATGTCCGACTCCAAA-3′
mCTDNEP1	5′-AGATCCGCACGGTAATTCAG-3′ 5′-GGTGTCCCAGGTCTCACTGT-3′
mDsytrophin	5′-AGTCCTCCCCAGGACACAAGCA-3′ 5′-CCATCGCTCTGCCCAAATCATC-3′
mATP2A2 (Serca2)	5′-ATCTCCTTGCCTGTGATCCTC-3′ 5′-AGTCATGCAGAGGGCTGGTAG-3′
mTBP	5′-ACCCTTCACCAATGACTCCTATG-3′ 5′-ATGATGACTGCAGCAAATCGC-3′
hNEP1-R11 (TMEM188)	5′-GCTGCTCGATGTCGAACGGTAT-3′ 5′-CATATCCAACCGCAAAAGGAATC-3′
hCTDNEP1 (Dullard)	5′-TACCTTCTGCGGAGGCAGATCC-3′ 5′-TGTCTCATCCAGATCCAGCACCA-3′
hLipin-1	5′-TGCTGGAGAGCAGCAGAACTC-3′ 5′-TAGGGTATGAGGCTGACTGAG-3′
hlipin-2	5′-CCTCTCCTCAGACCAGATCG-3′ 5′-GGAGAATCTGTCCCAAAGCA-3′
hHPRT	5′-TATGGCGACCCGCAGCCCT-3′ 5′-CATCTCGAGCAAGACGTTCAG-3′
hβ2M	5′-GTCTTTCAGCAAGGACTGGTC-3′ 5′-CAAATGCGGCATCTTCAAACC-3′
Oligos for knockdown studies	
ASO control	5′-CCTTCCCTGAAGGTTCCTCC-3′
Lipin1-ASO	5′-GGGATGAGATGCAGCCTCTC-3′
siNEP1-R1 (mouse TMEM188)	ON-TARGETplus SMART pool, Dharmacon Inc., Chicago, IL

Table S IV. Plasmids used in the human cell study

Plasmid	Relevant characteristics	Source or reference
CTDNEP1-V5 His	Mammalian expression vector (pcDNA3.1/V5-His, Invitrogen) containing of CTDNEP1 sequence before tag	(Kim et al, 2007)
CTDNEP1-2xFLAG	Mammalian expression vector (pcDNA3.1, Invitrogen) containing C-terminally FLAG tagged CTDNEP1, subcloned from pRS316-PGK1-CTDNEP1-2xFLAG (see Table S I)	This study
NEP1-R1-FLAG	Mammalian expression vector (pcDNA3.1) encoding C-terminal FLAG tagged NEP1-R1, which was PCR amplified from pRS315-PGK1-NEP1-R1 (see Table S I)	This study
NEP1-R1-HA-ProtA	Mammalian expression vector (pcDNA3.1) containing C-terminally tagged NEP1-R1-HA-protA, subcloned from pRS316-PGK1-NEP1-R1-HA-ProtA (see Table S I)	This study
HA-Lipin1b	Mammalian expression vector (pcDNA3.1) containing N-terminally HA-tagged mouse Lpin1b	(Kim et al, 2007)
Lipin1a-V5 His	Mammalian expression vector (pcDNA3.1/V5-His) containing C-terminally tagged mouse Lpin1a	(Donkor et al, 2007)
Lipin1b-V5 His	Mammalian expression vector (pcDNA3.1/V5-His) containing C-terminally tagged mouse Lpin1b	(Donkor et al, 2007)
Lipin 2-V5 His	Mammalian expression vector (pcDNA3.1/V5-His) containing C-terminally tagged mouse Lpin2	(Donkor et al, 2007)

Donkor, J, Sariahmetoglu, M, Dewald, J, Brindley, DN, Reue, K (2007) Three mammalian lipins act as phosphatidate phosphatases with distinct tissue patterns. *J Biol Chem* **282**:3450-3457

Kim, Y, Gentry, MS, Harris, TE, Wiley, SE, Lawrence, JC, Dixon JE (2007) A conserved phosphatase cascade that regulates nuclear membrane biogenesis. *Proc Natl Acad Sci U S A* **104**:6596-6601