

## **Supplemental Figure legends:**

**Supplemental Figure 1. Mass Spectrometry analysis of LONP1 interacting partners.** The LONP1 immunoprecipitation (IP) experiment was done using DSG-cross-linked control mitochondria followed by mass spectrometry (MS) analysis of the elution fraction. Table shows only proteins exclusively identified by MS in LONP1 IP and not in control.

**Supplemental Figure 2. Mass Spectrometry analysis of the insoluble fraction in the LONP1-depleted cell line.** Mitochondria from control and LONP1-depleted cells (4 days) were treated with DDM (n-Dodecyl- $\beta$ -D-Maltoside). The pellet fraction was further extracted with ProteaseMAX<sup>™</sup> surfactant and analyzed in solution by mass spectrometry. Table shows only proteins exclusively identified by MS in LONP1-depleted cell line (N/A) or at least 3-fold enriched compare to control.

**Supplemental Figure 3. Mass Spectrometry analysis of MPP $\beta$  interacting partners.** The MPP $\beta$  immunoprecipitation (IP) experiment was done in control mitochondria followed by mass spectrometry (MS) analysis of the elution fraction.

Supplemental Figure 1:

Accession number	Official name	Protein mascot score
gil83318181	LONP1	2115
gil119621956	AFG3L2	1152
gil119619869	IMMT	1099
gil21040386	HSPA9	958
gil862457	HADHA	775
gil14043666	ATAD3A	679
gil15991829	HK1	599
gil193783699	GOT2	579
gil37589015	TRAP1	507
gil7657581	SLC25A13	478
gil119606477	YME1L1	475
gil21411235	NDUFS1	449
gil6002671	GLS	465
gil269914128	OCIA1D	441
gil119617402	SHMT2	441
gil14290586	TIMM50	416
gil180928	COREII	411
gil3559910	SLC25A12	406
gil119604208	CHCHD3	388
gil7019477	HTRA2	379
gil119590169	PYCR2	327
gil49902674	OPA1	321
gil156416003	SDHA	318
gil801893	LRPPRC	283
gil499719	DLST	277
gil4929571	SAMM50	276
gil62897195	GLUD1	268
gil7242140	CPLX	268
gil4503607	EFTA	265
gil49456495	PYCR1	258
gil224451073	EXOG	252
gil74355155	ALDH1L2	244
gil12751119	PHB2	215
gil33636719	TIMM44	213
gil1718502	ACO2	212
gil20270303	RHOT2	197
gil22202629	AIFM1	194
gil28575023	MRPL43	190
gil32483377	PRDX3	185
gil163644321	UQCRCF1	183
gil156142176	TIMM21	178
gil12655193	PCK2	177
gil27886570	MRPL39	175
gil119607088	PRKDC	175
gil4502491	C1QBP (p32)	166
gil4557237	ACAT1	158

gil12597661	MRPL44	156
gil11321583	SUCLA2	151
gil15553127	HK2	149
gil119587871	NLRX1	142
gil5174723	TOMM40	139
gil62896795	NDUFS2	135
gil5454122	TIMM23	131
gil4758774	NDUFB10	129
gil119568145	MTHFD1L	129
gil21619683	OPTN	125
gil11545863	MCCC2	123
gil15080505	CCDC51	123
gil114325408	IARS2	120
gil2078327	HADH	120
gil38683855	PTCD3	119
gil25187965	RHOT1	118
gil151328807	COX2	116
gil30841303	SOD2	114
gil119599084	GFM1	112
gil188036020	DDX3X	112
gil31711992	DLTA	110
gil21687102	TIMM14	110
gil181238	CYC1	109
gil55663095	HAX1	108
gil550062	RAB2A	107
gil22027538	PDCD61P	107
gil119603999	NDUFA5	105
gil4758152	TIMM8	102
gil119609244	NDUFA9	102
gil15213479	POLDIP2	100
gil46250003	TMEM214	98
gil11024700	TIMM13	98
gil141797011	IQGAP1	97
gil7661806	MRPL15	96
gil4557657	ICT1	95
gil21265040	MRPL22	95
gil180937	COX5B	94
gil4557353	BCKDHB	94
gil28422556	SUCLG2	94
gil15559568	DNAJC11	93
gil62088434	ACAD9	89
gil62202498	UQCRH	88
gil15342081	MRPL38	86
gil4507231	SSBP1	85
gil20631958	BAX	83
gil15778942	NT5DC2	81
gil223468571	MRPL23	80
gil11139093	GRPEL1	80
gil42542702	CHCHD2	78

gil5729937	MTX2	76
gil10947135	ABCF1	75
gil14250760	PTRF	73
gil112421122	DNAJC13	73
gil22902126	PTK7	72
gil110169703	ND5	72
gil40225932	DNAJA3	70
gil14150134	CHCHD6	70
gil22547136	MRPL4	70
gil116283278	TFB1M	70
gil4758138	DDX5	70
gil1263008	ALDH1B1	69
gil14124984	CCT3	65
gil181486	YBX1	64
gil7706351	PTRH2	63
gil118142816	MRPL1	60
gil49457552	FARS1	60
gil21687127	CCDC127	59
gil48146471	MRPL17	57
gil21411521	NDUFA10	56
gil38569475	MTX1	55
gil40789249	DARS	54
gil7657369	NDUFA8	54
gil62089036	CCT6A	54
gil47115243	TFAM	52
gil70166645	PUS1	51
gil62988276	MRPP3	51
gil47678395	DDX17	46
gil34809544	NDUFA13	45
gil157384956	NUBPL	43
gil119627099	MRPL37	43
gil20336290	DHX30	42
gil311893396	ATP5J2-PTCD1	39
gil40226469	PMPCB	28

Supplemental Figure 2:

Accession number	Official name	LONP1-4D-KD (PELLET) Mascot score	CONTROL (PELLET) Mascot score	Score ratio
gil31621305	LRPPRC	2167	690	3.14
gil74355155	ALDH1L2	1562	N/A	N/A
gil21411235	NDUFS1	1243	152	8.18
gil20336290	DHX30	1125	315	3.57
gil36796743	MTHFD1L	870	248	3.51
gil1263008	ALDH1B1	804	130	6.18
gil20372922	PNPT1	725	52	13.94
gil373251164	GLS	725	33	21.97
gil12655193	PCK2	720	N/A	N/A
gil312812	PCCB	699	N/A	N/A
gil515634	COREI	682	128	5.33
gil119616108	MRPS27	672	137	4.91
gil21361322	TUBB4A	659	N/A	N/A
gil4503377	DPYSL2	657	162	4.06
gil119617634	CCT2	450	78	5.77
gil4503607	ETFA	618	109	5.67
gil4758788	NDUFS3	531	148	3.59
gil3184148	NDUFV1	530	N/A	N/A
gil347134	SDHA	525	64	8.20
gil1168056	OAT	480	N/A	N/A
gil55960510	CCT3	142	47	3.02
gil7242140	CLPX	479	N/A	N/A
gil509291	ATP5F1	463	124	3.73
gil49456495	PYCR1	460	112	4.11
gil187452	MUT	456	N/A	N/A
gil7661672	POLDIP2	450	N/A	N/A
gil4557235	ACADVL	448	46	9.74
gil12804973	ELAC2	444	N/A	N/A
gil51873036	OGDH	439	57	7.70
gil33350932	DYNC1H1	434	79	5.49
gil1082886	TRAP1	430	N/A	N/A
gil3342006	PMPCB	428	N/A	N/A
gil28178832	IDH2	426	23	18.52
gil4103602	TIMM44	415	35	11.86
gil11545863	MCCC2	415	N/A	N/A

gil40789249	DARS2	410	76	5.39
gil14210536	TUBB6	410	N/A	N/A
gil261399877	CCT7	408	22	18.55
gil34147676	MTERFD1	397	N/A	N/A
gil3415109	ERAL1	397	N/A	N/A
gil62897551	MRPS7	393	N/A	N/A
gil12597661	MRPL44	393	40	9.83
gil62898103	GOT2	391	99	3.95
gil38683855	PTCD3	389	N/A	N/A
gil1000704	NNT	389	N/A	N/A
gil62988276	MRPP3	388	57	6.81
gil11596859	MRPL17	381	N/A	N/A
gil15928608	SLC25A6	377	N/A	N/A
gil45267832	FASTKD2	377	N/A	N/A
gil22547134	MRPL37	377	N/A	N/A
gil49168620	ACO2	374	N/A	N/A
gil20070344	TARS2	374	N/A	N/A
gil14043072	HNRNPA2B1	371	117	3.17
gil16950603	MRPS35	368	44	8.36
gil4502643	CCT6A	363	46	7.89
gil1922287	ECHS1	362	N/A	N/A
gil40217812	TBRG4	361	N/A	N/A
gil38492358	PTCD1	294	N/A	N/A
gil18028283	ACAD9	357	79	4.52
gil50417352	DPYSL3	356	66	5.39
gil7661806	MRPL15	348	N/A	N/A
gil14589864	ASPH	117	N/A	N/A
gil21361454	PYCR2	343	76	4.51
gil187281	HNRNPM	339	41	8.27
gil71052166	POLRMT	335	66	5.08
gil189049	NDUFA9	335	32	10.47
gil180917	SDHB	334	N/A	N/A
gil21626466	MATR3	324	24	13.50
gil14285174	GFM1	322	N/A	N/A
gil9800545	ACOT2	317	N/A	N/A
gil7661556	TRUB2	316	N/A	N/A
gil4758118	MRPS29	313	75	4.17
gil119617482	TSFM	312	N/A	N/A
gil57863257	TCP1	310	N/A	N/A
gil12804225	CCT5	304	76	4.00
gil62896685	RUVBL1	299	44	6.80

gil9910244	MRPS22	292	N/A	N/A
gil14150078	NOA1	290	N/A	N/A
gil8923421	SARS2	286	N/A	N/A
gil10864011	SQRDL	286	N/A	N/A
gil509676	ACAA2	275	N/A	N/A
gil211828181	HNRNPL	273	N/A	N/A
gil40225932	DNAJA3	272	70	3.89
gil119573109	COPA	271	30	9.03
gil38569417	ARS2	270	N/A	N/A
gil4557817	OXCT1	267	N/A	N/A
gil184427	HSPG2	267	44	6.07
gil119606898	PITRM1	261	N/A	N/A
gil116283278	TFB1	260	57	4.56
gil12584221	DDX28	257	N/A	N/A
gil5106956	FHOD1	255	69	3.70
gil38649415	SRPRB	253	78	3.24
gil379317177	DDX21	248	36	6.89
gil4506103	EIF2AK2	247	71	3.48
gil119595266	CLPB	245	67	3.66
gil387011	PDHA1	244	54	4.52
gil32358	HNRNPU	244	23	10.61
gil55769543	MRPL14	242	N/A	N/A
gil21361331	CPS1	236	N/A	N/A
gil4826649	MRPL49	232	N/A	N/A
gil70995422	NQO1	227	32	7.09
gil37572267	TRMT10C	224	N/A	N/A
gil4885377	HIST1H1D	222	N/A	N/A
gil22547140	MRPL4	152	N/A	N/A
gil181575	DLD	219	57	3.84
gil285945	MRPL19	219	N/A	N/A
gil119597555	TFB2M	210	N/A	N/A
gil16554607	MRPS10	209	N/A	N/A
gil5031877	LMNB1	209	N/A	N/A
gil24308400	MCU	209	N/A	N/A
gil10437178	MRPL1	206	N/A	N/A
gil62897643	MTHFD2	205	N/A	N/A
gil14028877	MRPS25	205	N/A	N/A
gil40254986	HSDL2	204	55	3.71
gil119582537	ETF1	202	N/A	N/A
gil13559363	MRPL9	200	N/A	N/A
gil211904152	SERPINE2	198	N/A	N/A

gil48145545	NDUFA6	193	N/A	N/A
gil7662645	MRPS18B	192	N/A	N/A
gil15721937	MRPS24	191	N/A	N/A
gil116063536	COQ5	191	N/A	N/A
gil11139093	GRPEL1	190	N/A	N/A
gil7705626	MRPS16	189	N/A	N/A
gil34740329	HNRNPA3	186	N/A	N/A
gil29126836	MRPS9	185	44	4.20
gil28277071	IPO5	181	N/A	N/A
gil3941724	EIF4G1	179	50	3.58
gil2737886	IDH3B	176	N/A	N/A
gil7705851	CHCHD2	175	N/A	N/A
gil7022287	IARS2	174	N/A	N/A
gil7705618	MRPL11	172	51	3.37
gil4505571	SQSTM1	172	55	3.13
gil15011974	ARHGEF2	171	45	3.80
gil4505371	NDUFS8	171	36	4.75
gil968888	HMGB1	171	40	4.28
gil3342734	NDUFS7	168	N/A	N/A
gil7661548	MMADHC	168	N/A	N/A
gil5453555	RAN	168	49	3.43
gil3540239	NDUFS2	165	N/A	N/A
gil14149738	NLN	164	N/A	N/A
gil21265040	MRPL22	163	N/A	N/A
gil11141903	FASTKD5	163	N/A	N/A
gil27886570	MRPL39	160	N/A	N/A
gil56205490	ACADM	158	N/A	N/A
gil5931600	PPA2	157	N/A	N/A
gil4826848	NDUFA5	157	N/A	N/A
gil20149633	ECSIT	157	N/A	N/A
gil18314586	MRPS31	154	N/A	N/A
gil119588675	METTL15	153	N/A	N/A
gil28872734	MRPL43	153	N/A	N/A
gil4758138	DDX5	152	N/A	N/A
gil41327781	NDUFAF3	149	N/A	N/A
gil119598593	AHSG	144	N/A	N/A
gil26051229	MRPL28	143	N/A	N/A
gil11321601	PFKP	143	31	4.61
gil78070334	GTPBP10	140	N/A	N/A
gil28212272	NOP9	140	43	3.26
gil6274550	NDUFB9	138	N/A	N/A



gil4557657	ICT1	138	N/A	N/A
gil21265080	MRPL18	137	N/A	N/A
gil34709	SOD2	136	N/A	N/A
gil8922625	TMLHE	136	N/A	N/A
gil511635	ECI1	136	N/A	N/A
gil5453832	HYOU1	136	38	3.58
gil179360	BCKDHA	136	N/A	N/A
gil27597059	DNAJC9	135	N/A	N/A
gil40254992	CBR4	135	N/A	N/A
gil56549685	GTPBP8	134	N/A	N/A
gil119625894	PAICS	132	N/A	N/A
gil13654278	SLIRP	132	20	6.60
gil38648667	FASN	129	N/A	N/A
gil23308697	SRPR	127	42	3.02
gil39725942	TWINKLE	126	20	6.30
gil157426893	GUF1	123	28	4.39
gil292029	FPGS	121	N/A	N/A
gil292830	TOP2A	120	N/A	N/A
gil4732024	PDSS1	115	N/A	N/A
gil16554604	MRPS23	115	N/A	N/A
gil23273479	MTG1	114	N/A	N/A
gil119611391	MRPS14	114	N/A	N/A
gil42518070	TJP2	114	N/A	N/A
gil5453918	SLC25A17	112	N/A	N/A
gil7657118	GCAT	112	N/A	N/A
gil7661730	MRPS28	110	N/A	N/A
gil793763	MMP14	109	N/A	N/A
gil24308117	MTG2	108	N/A	N/A
gil3493529	H2AFY	108	N/A	N/A
gil21265070	MRPL2	108	N/A	N/A
gil444646	APP	108	N/A	N/A
gil22035588	MRPL24	107	N/A	N/A
gil695360	COX5A	106	20	5.30
gil223468571	MRPL23	106	N/A	N/A
gil288563	ITIH3	104	N/A	N/A
gil6912292	CBX5	104	N/A	N/A
gil13654274	FAHD1	104	N/A	N/A
gil62898171	LCP1	103	18	5.72
gil14141157	HNRNPH3	102	N/A	N/A
gil14602703	BCKDK	101	15	6.73
gil18079218	OSBPL8	101	30	3.37

gil31652228	MRPL21	99	N/A	N/A
gil37594471	HIBCH	97	N/A	N/A
gil62088696	SRSF1	96	N/A	N/A
gil28839782	COX4L1	96	N/A	N/A
gil14602513	MRPL48	96	N/A	N/A
gil16877964	IVD	95	N/A	N/A
gil7705369	COPB1	94	N/A	N/A
gil62898139	GJA1	92	N/A	N/A
gil48146851	IPO4	91	N/A	N/A
gil48976061	ACAD10	91	N/A	N/A
gil22749443	MICU2	91	N/A	N/A
gil40255109	GRPEL2	90	N/A	N/A
gil62420877	ETFB	90	N/A	N/A
gil1292941	HMGCL2	89	N/A	N/A
gil4826862	NME4	88	N/A	N/A
gil3249735	KIFC3	88	N/A	N/A
gil8922631	EXD2	88	N/A	N/A
gil181238	CYC1	87	28	3.11
gil180937	COX5B	87	N/A	N/A
gil21040371	DDX39A	86	N/A	N/A
gil55665272	MGST3	85	N/A	N/A
gil119622401	MRPL46	85	N/A	N/A
gil21618497	MRPL47	84	N/A	N/A
gil21265096	MRPL50	84	N/A	N/A
gil8922534	RNMTL1	83	N/A	N/A
gil4758792	NDUFS6	83	N/A	N/A
gil19923640	GFM2	83	N/A	N/A
gil14269586	MRPS26	83	N/A	N/A
gil6959304	MYBBP1A	82	N/A	N/A
gil10092657	NDUFA12	82	N/A	N/A
gil9719409	PDSS2	82	N/A	N/A
gil18375501	APEX1	81	N/A	N/A
gil62088544	DIAPH1	81	N/A	N/A
gil119591303	TRIP12	81	N/A	N/A
gil56605994	CISD2	80	N/A	N/A
gil34783041	MRPS5	80	N/A	N/A
gil30524	DLAT	80	N/A	N/A
gil51476873	MCCC1	79	N/A	N/A
gil119580947	LASPI	79	N/A	N/A
gil913393	KPNA1	78	N/A	N/A
gil61835204	MPST	78	N/A	N/A

gil16877641	PRRC1	78	N/A	N/A
gil15489339	VASN	78	N/A	N/A
gil72534692	NT5DC3	78	N/A	N/A
gil4506001	PPOX	77	N/A	N/A
gil71680896	COX7A2	77	N/A	N/A
gil4099880	MYO5A	77	18	4.28
gil4503915	GART	76	N/A	N/A
gil14044070	DDRGK1	76	N/A	N/A
gil124297151	LARS	76	N/A	N/A
gil38176424	CUL4B	76	N/A	N/A
gil12803113	TUFM	76	N/A	N/A
gil32528306	RFC1	76	N/A	N/A
gil20149688	TRMT2B	75	N/A	N/A
gil12804901	IDH3G	73	N/A	N/A
gil31543415	GADD45GIP1	73	N/A	N/A
gil21619683	OPTN	72	N/A	N/A
gil119582193	ABLIM3	72	N/A	N/A
gil16554609	MRPS11	72	N/A	N/A
gil122937211	ECM29	71	N/A	N/A
gil41388987	RPUSD3	71	N/A	N/A
gil4505621	PEBP1	71	N/A	N/A
gil55961491	MTO1	70	N/A	N/A
gil124378039	SEC16A	70	N/A	N/A
gil333470683	MRPL45	69	N/A	N/A
gil70166599	PUS1	68	N/A	N/A
gil119601354	RDH11	68	N/A	N/A
gil2624702	SSBP1	49	N/A	N/A

Supplemental Figure 3:

Accession number	Official name	MPP $\beta$ Mascot score	Control Mascot score	Score ratio
gil94538354	PMPCB	1953	39	50.08
gil74746874	CDC42BPA	1068	#N/A	#N/A
gil67678154	RPS9	434	#N/A	#N/A
gil729475	FEN1	398	#N/A	#N/A
gil62906882	ATP5L	346	53	6.53
gil83305924	TIMM50	310	36	8.61
gil84708830	TUBB6	309	#N/A	#N/A
gil62822438	AGPS	303	#N/A	#N/A
gil83318181	LONP1	286	120	2.38
gil62897685	TIMM44	282	#N/A	#N/A
gil54696944	RPL10a	275	20	13.75
gil987870	OAPB	254	#N/A	#N/A
gil78099790	PLOD1	247	#N/A	#N/A
gil83755037	ACAT1	236	#N/A	#N/A
gil7670842	ECI2	218	#N/A	#N/A
gil62945797	H2AFX	211	#N/A	#N/A
gil578798836	MACF1	204	#N/A	#N/A
gil74355155	ALDH1L2	192	#N/A	#N/A
gil74733765	NT5DC2	180	#N/A	#N/A
gil94730427	SUCLA2	177	#N/A	#N/A
gil94721261	CNP	159	#N/A	#N/A
gil62422557	PAK4	154	#N/A	#N/A
gil74733244	APOO	133	#N/A	#N/A
gil71151875	COX20	131	#N/A	#N/A
gil74759261	PXK	125	19	6.58
gil7705618	MRPL11	116	#N/A	#N/A
gil9665218	CLPX	115	#N/A	#N/A
gil790447	ACADV1	114	#N/A	#N/A
gil7023534	AK3	113	#N/A	#N/A
gil74750555	PTRH1	112	#N/A	#N/A
gil58257735	ARHGEF2	110	#N/A	#N/A
gil74762790	FADS3	99	#N/A	#N/A
gil68533031	FASN	96	#N/A	#N/A
gil86197962	FLOT1	95	#N/A	#N/A
gil7706349	MRPS33	94	#N/A	#N/A
gil68052264	UFL1	93	#N/A	#N/A
gil7920147	NSF	86	#N/A	#N/A

gil83753568	RAB18	86	#N/A	#N/A
gil9247171	FYN	85	#N/A	#N/A
gil548937720	IQGAP3	82	#N/A	#N/A
gil83715985	HSD17B10	78	#N/A	#N/A
gil8923708	FOXRED1	78	#N/A	#N/A
gil571026676	SLC39A7	75	#N/A	#N/A
gil97046413	CCDC93	73	#N/A	#N/A
gil85701348	FECH	72	#N/A	#N/A
gil74762551	ATAD1	71	#N/A	#N/A
gil9755051	TM9SF3	69	#N/A	#N/A
gil62822421	MAP4K4	68	#N/A	#N/A
gil98986464	TMED10	67	#N/A	#N/A
gil9622124	RDH11	66	#N/A	#N/A
gil7657345	MTCH1	65	#N/A	#N/A
gil68299593	JAK1	65	#N/A	#N/A
gil81175200	TALIN1	62	#N/A	#N/A
gil74355449	DPM3	61	#N/A	#N/A
gil62897865	MRPS11	61	#N/A	#N/A
gil7705274	ARMCX3	60	#N/A	#N/A
gil8922601	ARL8B	60	#N/A	#N/A
gil6273493	SPAST	60	#N/A	#N/A
gil801893	LRPPRC	60	#N/A	#N/A
gil6980694	3HAD_B	59	#N/A	#N/A
gil74749360	MTHFD1L	59	#N/A	#N/A
gil74761587	CARS2	59	#N/A	#N/A
gil64654324	GNA11	58	#N/A	#N/A
gil914935	DAD-1	58	#N/A	#N/A
gil8515718	CALU	55	#N/A	#N/A
gil8886871	PLSCR1	55	#N/A	#N/A
gil578804559	DOCK10	55	#N/A	#N/A
gil91208273	D2HGDH	53	#N/A	#N/A
gil8650412	ABCB10	53	#N/A	#N/A
gil7023042	ELOVL1	53	#N/A	#N/A
gil8923565	AURKAIP1	52	#N/A	#N/A
gil8922511	MRPS18A	52	#N/A	#N/A
gil7688673	RTCB	52	#N/A	#N/A
gil578815274	SLC39A14	52	#N/A	#N/A
gil7705805	MRPS2	52	#N/A	#N/A
gil84105433	GNA12	49	#N/A	#N/A
gil62089396	MRPS15	42	#N/A	#N/A
gil90112017	PMPCA	24	#N/A	#N/A