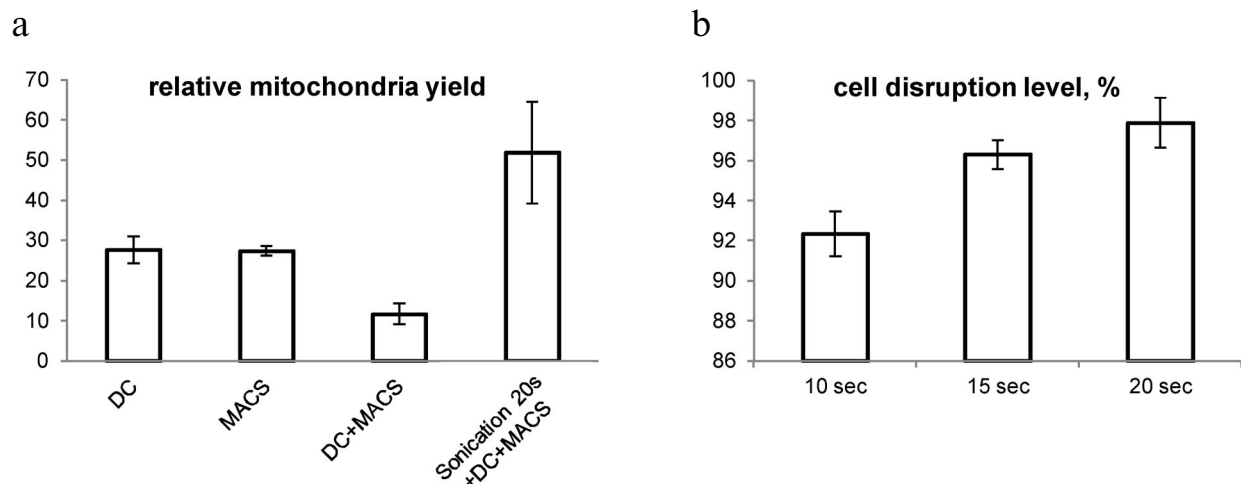
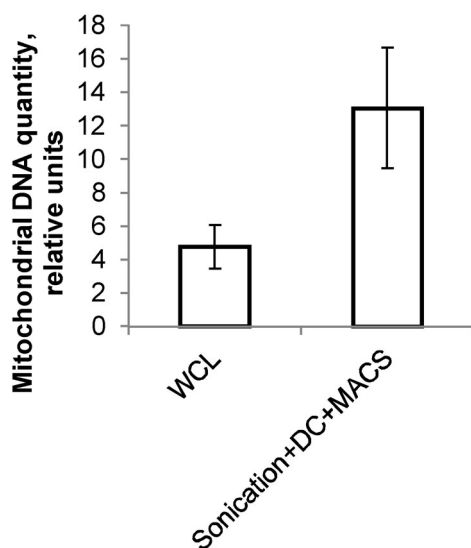


**Supplement to:** M. A. Afanasyeva, A. S. Ustiugova, S. A. Golyshev, A. T. Kopylov, A. V. Bogolyubova, D. E. Demin, P. V. Belousov, and A. M. Schwartz, Isolation of Large Amounts of Highly Pure Mitochondria for "Omics" Studies (ISSN 0006-2979, *Biochemistry (Moscow)*, 2018, Vol. 83, No. 1, pp. 76-85)



**Fig. S1.** Cell disruption method affects mitochondria yield. a) Mitochondria were isolated from FTC-133 cells using different cell homogenization methods followed by differential centrifugation and magnetic separation with Miltenyi Biotech Mitochondria Isolation Kit. Relative mitochondria yield was calculated as mitochondria weight ( $\mu\text{g}$ ) divided by initial cell pellet weight (g). b) Cell disruption level after sonication in sucrose buffer at 50% cycle and 90% power for 10, 15, and 20 s (mean  $\pm$  SD).



**Fig. S2.** Mitochondrial DNA quantified in whole cell and Sonication+DC+MACS fractions by real-time PCR. Relative mitochondrial DNA amount was normalized to the total protein content of each probe. Mitochondrial isolation, DNA purification, and real-time PCR were performed in triplicates. Data are represented as mean  $\pm$  SD. WCL, whole cell lysate; Sonication+DC+MACS, isolation method that includes sonication of cells in sucrose buffer, differential centrifugation, and magnetic separation of mitochondria using Miltenyi Biotech Mitochondria Isolation Kit.

**Table S1.** Results of LC–MS

Spot No	Database	Accession	Score	Mass	Num. of matches	Num. of significant matches	Num. of sequences	Num. of significant sequences	emPAI	Description
1	SwissProt	HS90B_HUMAN	6213	83212	305	247	43	42	13,23	Heat shock protein HSP 90-beta OS=Homo sapiens GN=HSP90AB1 PE=1 SV=4
2	SwissProt	ACTB_HUMAN	2071	41710	189	131	16	15	12,2	Actin, cytoplasmic 1 OS=Homo sapiens GN=ACTB PE=1 SV=1
3	SwissProt	ENO1	2978	47139	132	77	23	16	10,47	Alpha-enolase OS=Homo sapiens GN=ENO1 PE=1 SV=2
4	SwissProt	VIME_HUMAN	28398	53619	489	429	45	44	888,56	Vimentin OS=Homo sapiens GN=VIM PE=1 SV=4
5	SwissProt	VIME_HUMAN	15255	53619	368	327	43	43	264,2	Vimentin OS=Homo sapiens GN=VIM PE=1 SV=4
6	SwissProt	I433Z_HUMAN	1426	27728	159	109	19	18	45,67	14-3-3 protein zeta/delta OS=Homo sapiens GN=YWHAZ PE=1 SV=1
7	SwissProt	LMNA_HUMAN	21324	74095	322	288	54	51	34,29	Prelamin-A/C OS=Homo sapiens GN=LMNA PE=1 SV=1
8	SwissProt	LMNA_HUMAN	19089	74095	319	287	47	43	29,49	Prelamin-A/C OS=Homo sapiens GN=LMNA PE=1 SV=1
9	SwissProt	I433E_HUMAN	3725	29155	232	184	24	23	188,35	14-3-3 protein epsilon OS=Homo sapiens GN=YWHAE PE=1 SV=1
10	SwissProt	TPM4_HUMAN	1712	28504	192	132	26	23	113,32	Tropomyosin alpha-4 chain OS=Homo sapiens GN=TPM4 PE=1 SV=3
11	SwissProt	CATB_HUMAN	2216	37797	137	112	10	10	14,56	Cathepsin B OS=Homo sapiens GN=CTSB PE=1 SV=3
12	SwissProt	CATB_HUMAN	378	37797	58	40	8	7	3,14	Cathepsin B OS=Homo sapiens GN=CTSB PE=1 SV=3
13	SwissProt	ANXA2_HUMAN	12618	38580	286	216	30	29	53,02	Annexin A2 OS=Homo sapiens GN=ANXA2 PE=1 SV=2
14	SwissProt	TUFM	335	49510	18	13	15	12	NA	Elongation factor Tu, mitochondrial OS=Homo sapiens GN=TUFM PE=1 SV=2
15	SwissProt	C1QBP_HUMAN	12454	31343	177	175	11	11	6,77	Complement component 1 Q subcomponent-binding protein, mitochondrial OS=Homo sapiens GN=C1QBP PE=1 SV=1
16	SwissProt	IDH3A_HUMAN	1197	39566	25	15	7	7	0,89	Isocitrate dehydrogenase [NAD] subunit alpha, mitochondrial OS=Homo sapiens GN=IDH3A PE=1 SV=1
17	SwissProt	STML2_HUMAN	4482	38510	286	228	21	21	71,22	Stomatin-like protein 2 OS=Homo sapiens GN=STOML2 PE=1 SV=1
18	SwissProt	RM11_HUMAN	296	20670	38	20	10	10	5,52	39S ribosomal protein L11, mitochondrial OS=Homo sapiens GN=MRPL11 PE=1 SV=1
19	SwissProt	ATPB_HUMAN	6371	56525	99	91	20	19	4,61	ATP synthase subunit beta, mitochondrial OS=Homo sapiens GN=ATP5B PE=1 SV=3
20	SwissProt	P5CR1_HUMAN	1860	33340	99	78	13	12	8,51	Pyrroline-5-carboxylate reductase 1, mitochondrial OS=Homo sapiens GN=PYCR1 PE=1 SV=2
20	SwissProt	VDAC2_HUMAN	1032	31547	78	58	8	8	3,35	Voltage-dependent anion-selective channel protein 2 OS=Homo sapiens GN=VDAC2 PE=1 SV=2
20	SwissProt	VDAC1_HUMAN	985	30754	74	54	11	10	3,02	Voltage-dependent anion-selective channel protein 1 OS=Homo sapiens GN=VDAC1 PE=1 SV=2
21	SwissProt	ODO2_HUMAN	1413	48724	27	20	8	6	0,68	Dihydrolipoyllysine-residue succinyltransferase component of 2-oxoglutarate dehydrogenase complex, mitochondrial OS=Homo sapiens GN=DLST PE=1 SV=4
21	SwissProt	OAT_HUMAN	1179	48504	36	21	14	10	1,1	Ornithine aminotransferase, mitochondrial OS=Homo sapiens GN=OAT PE=1 SV=1
21	SwissProt	SCMC1_HUMAN	1139	53320	40	20	15	10	0,97	Calcium-binding mitochondrial carrier protein SCaMC-1 OS=Homo sapiens GN=SLC25A24 PE=1 SV=2