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Contributions to deliverable - Internal review procedure

Deliverable produced by:	Date:
Bernardo Lino de Oliveira - Partner KCL	September 2015
Steven Niederer (KCL) - Partner KCL	September 2015
Lars Küpfer - Partner RWTH	October 2015
Deliverable internally reviewed by:	Date:
Jos Kleinjans – Partner UM	October 2015

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PUBLISHABLE SUMMARY

We have evaluated models of cardiac mitochondria and models of inflammation. Models were evaluated against their ability to simulate experimental and physiological conditions and capture the major cardiac toxicity pathways. A computational model for anthracycline induced mitochondrial toxicity was developed as this drug class made up the majority of initial drug targets identified in WP5. Initial models focused on simulating the effects of the common anthracycline doxorubicin. In a complementary approach, multiscale models were developed for various hepatotoxicants which simultaneously consider the organism level and the cellular space. Models were initially fitted and validated against literature data. The framework is now ready to exploit functional and proteomic clinical and experimental data from HeCaToS partners.

OBJECTIVES

Over the course of HeCaTos, WP3 aims to:

1. Develop a data-driven pipeline to establish and parameterise physiological toxicity pathway models (mitochondrial dysfunction and inflammation) from experimental data made available in the project;
2. Validate the predictions of the cellular modelling framework using data from toxicology assays and patient studies;
3. Quantify the different responses and underlying pathways between the experimental studies by comparing the parameter sets between models identified and validated from different preparations;
4. Integrate the cellular models of cardiac and hepatic cells into spatial models of tissue and the whole organs;
5. Simulate tissue-specific dose-response correlations.

In this report we focus on Objective 1: “Develop a data-driven pipeline to establish and parameterise physiological toxicity pathway models (mitochondrial dysfunction and inflammation) from experimental data made available in the project”.

A general model was developed to study mitochondrial cardiotoxicity and was initially applied to anthracyclines. Likewise, a generic workflow for vertical model integration has been reported (D1.4.1./T1.4.1) which was exemplarily applied to various hepatotoxicants. Both frameworks used can serve as a base for the study and prediction of cardiotoxicity and hepatotoxicity of other compounds in the future.

INTRODUCTION

The toxicity of drugs is hard to predict and is one of the main causes of preclinical and clinical drug withdrawal. This problem is aggravated when the toxicity is long-term and symptoms only show up after a considerable amount of time has passed following drug administration. Anthracyclines, which are a class of antibiotics with potent anticancer properties, are an exemplar class of drugs with long-term chronic toxicity. Patients exposed to high doses of these drugs can develop a cumulative, dose-dependent cardiomyopathy that evolves to congestive heart failure. These symptoms can show up years or even decades after the termination of the chemotherapy.

The underlying biochemical mechanisms responsible for both the anticancer properties and cardiotoxicity of anthracyclines are still not fully understood. Therefore, it is essential to investigate the exact mechanisms of cardiotoxicity and to what extent they are independent of the anticancer mechanisms, in order to find methods to reduce the toxicity of the drug without undermining its desired effects. The main anthracyclines used in clinical practice are doxorubicin (DOX), daunorubicin, epirubicin and idarubicin. These drugs have considerably similar molecules, presenting only minor structural differences that give them different spectrums of anticancer activity [1]. DOX is one of the earliest and most commonly used anthracyclines and has a significant body of data characterizing its effects, thus, it was chosen as the first case study.

DOX cardiotoxicity is strongly linked to mitochondrial dysfunction, leading to cardiac oxidative stress by increasing reactive oxygen species (ROS) production. Although ROS have important biochemical roles in cell signalling and homeostasis, its overproduction can have severe harmful effects such as DNA damage, oxidation of proteins and fatty acids in lipids, oxidative deactivation of enzymes among others.

It has been suggested that DOX can trigger a vicious cycle in the mitochondria, where increased ROS levels lead to mtDNA damage which can exacerbate the mitochondrial dysfunction and further increase ROS production [2]. This mechanism could continue to operate even after termination of the treatment.

Another frequent clinical cases for adverse drug-induced events are is hepatotoxicity. In order to detect drug-induced liver injury at an early stage, reliable predictions of toxic events are of key relevance for patient safety. In turn, this clearly requires a mechanistic understanding of the underlying cellular processes and alterations at pathway level. Changes at different biological scales in the face of extracellular toxicants can nowadays be measured by 'omics' technologies to describe cellular alterations in response to toxic drug concentrations. However, a translation of cellular *in vitro* findings to an actual *in vivo* context remains challenging.

RESULTS

The strategy adopted to represent and study these phenomena was to implement a ODE based biophysical model. The first step was to choose an appropriate mitochondrial model to serve as base for the development of the drug model. The main components necessary to represent mitochondrial processes and the effects of DOX are a tri-carboxylic acid (TCA) cycle representation that incorporates multiple substrates, transporters in the inner membrane of the mitochondria, ATP production, a detailed electron transport chain (ETC) representation that includes the production of ROS and a ROS scavenging system. As one of the future goals of this WP is to incorporate such models into spatial simulations, a compact model is also desirable to allow future scalability.

Different models found in the literature were compared. The list of models explored include the 2007 Wu et al. model [3], which is a complex and thermodynamically accurate model that is composed of multiple compartments (62 ODEs) and the 2003 Cortassa et al. model [4] which is a kinetic model that use a more phenomenological approach and is more compact (12 ODEs). However, DOX cardiotoxicity is tightly related to ROS production, which is a component not represented in the aforementioned models. For this reason, the model of choice was the 2013 Gauthier et al. model [5] which includes not only representations of the TCA cycle, and multiple transporters, but also a detailed electron transport chain model, ROS production and ROS scavenging systems. This model is also reasonably compact (37 ODEs), which is a desirable characteristic. A complete list of the proteins represented in this model can be found in the Appendix.

DOX has been reported to undergo redox cycling and inhibit the ETC by oxidative and non-oxidative mechanisms during acute administration. Furthermore, chronic administration of DOX has been reported to permanently alter the gene expression profile of the heart. To incorporate these effects into the model, data related to how DOX affects the proteins represented in the model must be incorporated. The kind of data that can be included in this model includes IC50 values for the acute case and proteomic protein profiles for the chronic case.

The inhibition of Complexes I to IV has been characterized and IC50 values have been reported in the literature [6]. Curves were adjusted to represent a dose dependent inhibition of Complexes I to IV as seen on Figure 1. Also, ROS production on Complex I was increased to represent redox cycling.

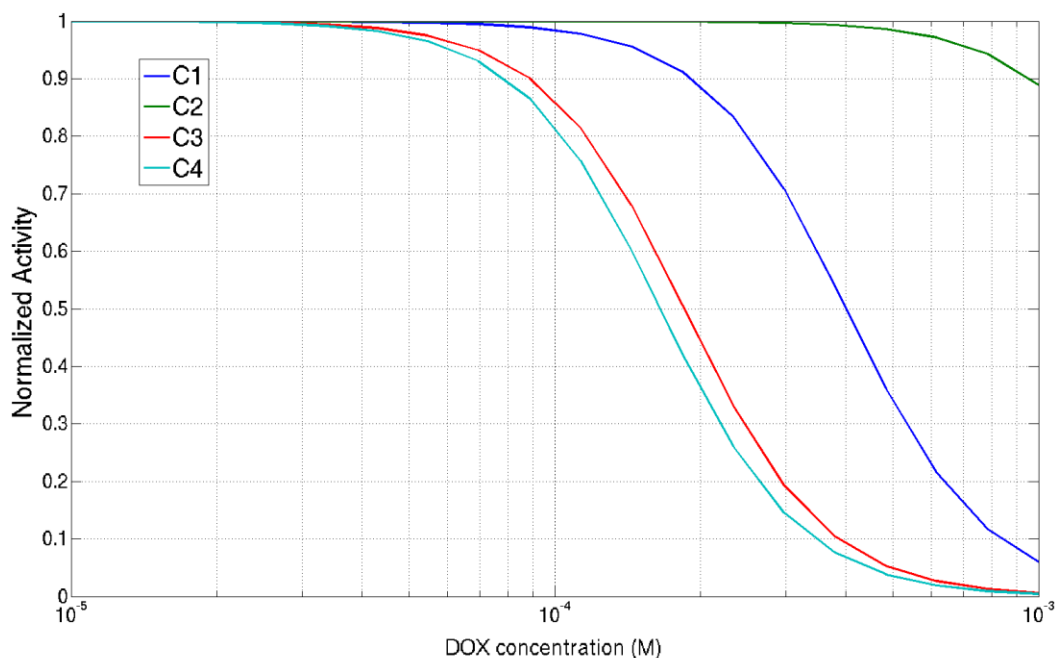


Figure 1: Dose dependent effect of DOX on isolated Complexes.

When these effects were incorporated into the mitochondrial model, a dose dependent drug response could be investigated. To quantify the mitochondrial dysfunction caused by DOX, different quantities that have been used: inner membrane potential, ATP, superoxide and glutathione levels. The predicted dose dependent response of these quantities can be observed on Figure 2. Multiple features associated with DOX toxicity were observed these simulations, including depolarization of the mitochondrial membrane, reduction in the ATP and antioxidant levels and an increase in the levels of ROS. These results are in good qualitative agreement with experimental results reported on the literature [7].

Physiologically-based pharmacokinetic (PBPK) modelling has been used before to simulate drug concentration profiles in the extracellular environment of hepatocytes thereby relating upstream drug administration to downstream pathway responses at the cellular level [9]. In order to establish a pipeline for future data available in the project we here used previously published gene expression data from the Open TG-Gates database [10]. For several of the hepatotoxicants considered in TG-Gates and in HeCaToS, human PBPK models were developed. In particular, following the development of an initial reference PBPK model for each of the compounds considered, the models were carefully validated by simulating different doses. Next, expression data from TG-Gates data were correlated to data from corresponding PBPK models for each of the hepatotoxicants considered so far, to provide insights into the temporal development of gene expression in the face of exposure to toxic compounds.

For each hepatotoxicant cellular response values were next calculated to quantify the drug response in enriched GO terms. In general, response profiles show low responses after early time points as opposed to larger changes after 24 h indicating a delayed regulatory response. Nonetheless, the initial increase of cellular response values in any pathway was found to be significant at the early time point ($p < 0.05$, Wilcoxon signed-rank test). For example, high cellular activity was identified in biological processes regulating cell replication, as well as in complexes involved in DNA replication for different immunosuppressant as such supporting the general validity of our PBPK-based approach. Further in-depth analysis are currently ongoing including in particular the translation of the current analysis to a clinically documented case of acute DILI documented by the HeCaToS partner HULAFE in Valencia.

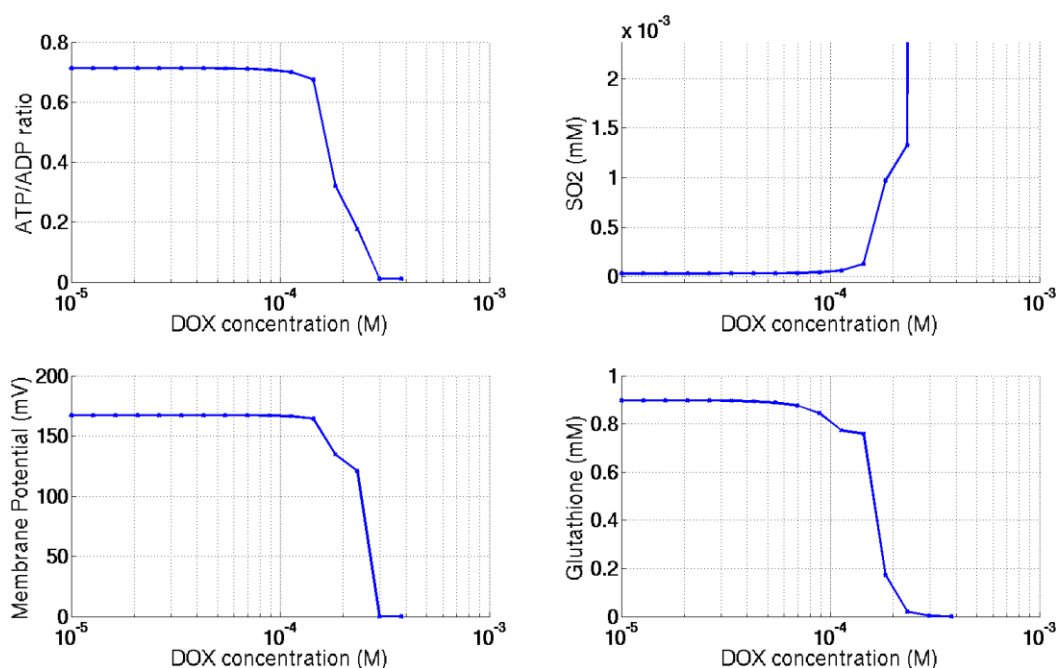


Figure 2: Dose dependent effect of DOX on the mitochondria

DIFFICULTIES AND DISCUSSION

A detailed model of the mitochondria that includes hundreds of different proteins (see Appendix) was implemented. By including the dominant targets and toxic pathways of DOX, it was possible to reproduce key experimental data associated with mitochondrial dysfunction caused by the drug. However, this only included data about a small fraction of the proteins represented in the model as the main difficulty in the development of this model was the scarcity of data available.

Ultimately, when more data is made available for the proteins present in the model, either in the form of IC₅₀ values or proteomic profile measurements of the proteins, they can be directly incorporated into the model by adjusting activities and densities of such proteins. This will allow a more complete representation of the drugs' effects and allow a more thorough investigation of both acute and chronic mitochondrial dysfunction and cardiotoxicity pathways. The framework developed here can serve as a base for the development of a pipeline of prediction of cardiotoxicity of drugs and investigation of toxicity pathways of other compounds. By estimating how much the mitochondrial function is affected

by the effect of drugs on different proteins activities and expressions, it is possible to perform a sensitivity analysis and identify and quantify the main pathways of toxicity.

The established PBPK-based pipeline outlines the analysis of experimental omics data generated within HeCaToS in the future. Developing a specific PBPK model for each of the toxicants considered within HeCaToS provides a platform of calculating tissue drug exposure both in the liver and in the heart. Moreover, PBPK presents a generic way of translating in vitro results to an in vivo situation.

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ANNEX: LIST OF PROTEINS

Table 1. List of proteins represented in the mitochondrial model.

Number	Entry name	Protein names	Gene names	Length
1	CISY_HUMAN	Citrate synthase	CS	466
2	ACON_HUMAN	Aconitate hydratase	ACO2	780
3	IDH3B_HUMAN	Isocitrate dehydrogenase [NAD] subunit beta	IDH3B	385
4	IDHP_HUMAN	Isocitrate dehydrogenase [NADP] Isocitrate dehydrogenase [NAD]	IDH2	452
5	IDH3A_HUMAN	subunit alpha Isocitrate dehydrogenase [NAD]	IDH3A	366
6	IDH3G_HUMAN	subunit gamma	IDH3G	393
7	ODO1_HUMAN	2-oxoglutarate dehydrogenase	OGDH	1023
8	SUCB1_HUMAN	Succinyl-CoA ligase [ADP-forming] subunit beta	SUCLA2	463
9	SUCB2_HUMAN	Succinyl-CoA ligase [GDP-forming] subunit beta	SUCLG2	432
10	SUCA_HUMAN	Succinyl-CoA ligase [ADP/GDP-forming] subunit alpha	SUCLG1	346
11	FUMH_HUMAN	Fumarate hydratase	FH	510
12	MDHM_HUMAN	Malate dehydrogenase	MDH2	338
13	NDUV1_HUMAN	NADH dehydrogenase [ubiquinone] flavoprotein 1	NDUFV1 UQOR1	464
14	NDUS7_HUMAN	NADH dehydrogenase [ubiquinone] iron-sulfur protein 7	NDUFS7	213
15	NDUS8_HUMAN	NADH dehydrogenase [ubiquinone] iron-sulfur protein 8	NDUFS8	210
16	NDUV2_HUMAN	NADH dehydrogenase [ubiquinone] flavoprotein 2	NDUFV2	249
17	NDUS3_HUMAN	NADH dehydrogenase [ubiquinone] iron-sulfur protein 3	NDUFS3	264
18	NDUS2_HUMAN	NADH dehydrogenase [ubiquinone] iron-sulfur protein 2	NDUFS2	463
19	NDUS1_HUMAN	NADH-ubiquinone oxidoreductase 75 kDa subunit	NDUFS1	727
20	NU1M_HUMAN	NADH-ubiquinone oxidoreductase chain 1 (EC 1.6.5.3) (NADH dehydrogenase subunit 1)	MT-ND1 MTND1 NADH1 ND1	318
21	NU2M_HUMAN	NADH-ubiquinone oxidoreductase chain 2 (EC 1.6.5.3) (NADH dehydrogenase subunit 2)	MT-ND2 MTND2 NADH2 ND2	347
22	NU3M_HUMAN	NADH-ubiquinone oxidoreductase chain 3 (EC 1.6.5.3) (NADH dehydrogenase subunit 3)	MT-ND3 MTND3 NADH3 ND3	115
23	NU4M_HUMAN	NADH-ubiquinone oxidoreductase chain 4 (EC 1.6.5.3) (NADH dehydrogenase subunit 4)	MT-ND4 MTND4 NADH4 ND4	459

24	NU4LM_HUMAN	NADH-ubiquinone oxidoreductase chain 4L (EC 1.6.5.3) (NADH dehydrogenase subunit 4L)	MT-ND4L MTND4L NADH4L ND4L	98
25	NU5M_HUMAN	NADH-ubiquinone oxidoreductase chain 5 (EC 1.6.5.3) (NADH dehydrogenase subunit 5)	MT-ND5 MTND5 NADH5 ND5	603
26	NU6M_HUMAN	NADH-ubiquinone oxidoreductase chain 6 (EC 1.6.5.3) (NADH dehydrogenase subunit 6)	MT-ND6 MTND6 NADH6 ND6	174
27	NDUS6_HUMAN	NADH dehydrogenase [ubiquinone] iron-sulfur protein 6	NDUFS6	124
28	NDUAC_HUMAN	NADH dehydrogenase [ubiquinone] 1 alpha subcomplex subunit 12 (13 kDa differentiation-associated protein) (Complex I-B17.2) (CI-B17.2) (CIB17.2) (NADH-ubiquinone oxidoreductase subunit B17.2)	NDUFA12 DAP13	145
29	NDUA9_HUMAN	NADH dehydrogenase [ubiquinone] 1 alpha subcomplex subunit 9	NDUFA9 NDUF52L	377
30	NDUS4_HUMAN	NADH dehydrogenase [ubiquinone] iron-sulfur protein 4	NDUFS4	175
31	ACPM_HUMAN	Acyl carrier protein	NDUFAB1	156
32	NDUA2_HUMAN	NADH dehydrogenase [ubiquinone] 1 alpha subcomplex subunit 2 (Complex I-B8) (CI-B8) (NADH-ubiquinone oxidoreductase B8 subunit)	NDUFA2	99
33	NDUA1_HUMAN	NADH dehydrogenase [ubiquinone] 1 alpha subcomplex subunit 1 (Complex I-MWFE) (CI-MWFE) (NADH-ubiquinone oxidoreductase MWFE subunit)	NDUFA1	70
34	NDUB3_HUMAN	NADH dehydrogenase [ubiquinone] 1 beta subcomplex subunit 3 (Complex I-B12) (CI-B12) (NADH-ubiquinone oxidoreductase B12 subunit)	NDUFB3	98
35	NDUA5_HUMAN	NADH dehydrogenase [ubiquinone] 1 alpha subcomplex subunit 5 (Complex I subunit B13) (Complex I-13kD-B) (CI-13kD-B) (NADH-ubiquinone oxidoreductase 13 kDa-B subunit)	NDUFA5	116
36	NDUA6_HUMAN	NADH dehydrogenase [ubiquinone] 1 alpha subcomplex subunit 6 (Complex I-B14) (CI-B14) (LYR motif-containing protein 6) (NADH-ubiquinone oxidoreductase B14 subunit)	NDUFA6 LYRM6 NADHB14	154

		NADH dehydrogenase [ubiquinone] 1 alpha subcomplex subunit 11 (Complex I-B14.7) (CI-B14.7) (NADH- ubiquinone oxidoreductase subunit B14.7)	NDUFA11	141
37	NDUAB_HUMAN		NDUFB11	
38	NDUBB_HUMAN	NADH dehydrogenase [ubiquinone] 1 beta subcomplex subunit 11 NADH dehydrogenase [ubiquinone] iron-sulfur protein 5 (Complex I-15 kDa) (CI-15 kDa) (NADH-ubiquinone oxidoreductase 15 kDa subunit)	UNQ111/PRO1064	153
39	NDUS5_HUMAN		NDUFS5	106
40	NDUB4_HUMAN	NADH dehydrogenase [ubiquinone] 1 beta subcomplex subunit 4 (Complex I-B15) (CI-B15) (NADH-ubiquinone oxidoreductase B15 subunit)	NDUFB4	129
41	NDUAD_HUMAN	NADH dehydrogenase [ubiquinone] 1 alpha subcomplex subunit 13 (Cell death regulatory protein GRIM-19) (Complex I-B16.6) (CI-B16.6) (Gene associated with retinoic and interferon-induced mortality 19 protein) (GRIM-19) (Gene associated with retinoic and IFN-induced mortality 19 protein) (NADH- ubiquinone oxidoreductase B16.6 subunit)	NDUFA13 GRIM19 CDA016 CGI-39	144
42	NDUB7_HUMAN	NADH dehydrogenase [ubiquinone] 1 beta subcomplex subunit 7 (Cell adhesion protein SQM1) (Complex I- B18) (CI-B18) (NADH-ubiquinone oxidoreductase B18 subunit)	NDUFB7	137
43	NDUA8_HUMAN	NADH dehydrogenase [ubiquinone] 1 alpha subcomplex subunit 8 (Complex I-19kD) (CI-19kD) (Complex I-PGIV) (CI-PGIV) (NADH-ubiquinone oxidoreductase 19 kDa subunit)	NDUFA8	172
44	NDUB9_HUMAN	NADH dehydrogenase [ubiquinone] 1 beta subcomplex subunit 9 (Complex I-B22) (CI-B22) (LYR motif-containing protein 3) (NADH-ubiquinone oxidoreductase B22 subunit)	NDUFB9 LYRM3 UQOR22	179
45	NDUBA_HUMAN	NADH dehydrogenase [ubiquinone] 1 beta subcomplex subunit 10 (Complex I-PDSW) (CI-PDSW) (NADH- ubiquinone oxidoreductase PDSW subunit)	NDUFB10	172
46	NDUB8_HUMAN	NADH dehydrogenase [ubiquinone] 1 beta subcomplex subunit 8	NDUFB8	186

		NADH dehydrogenase [ubiquinone] 1 subunit C2 (Complex I-B14.5b) (CI-B14.5b) (Human lung cancer oncogene 1 protein) (HLC-1) (NADH-ubiquinone oxidoreductase subunit B14.5b)	NDUFC2 HLC1	119
47	NDUC2_HUMAN			
48	NDUB2_HUMAN	NADH dehydrogenase [ubiquinone] 1 beta subcomplex subunit 2	NDUFB2	105
		NADH dehydrogenase [ubiquinone] 1 alpha subcomplex subunit 7 (Complex I-B14.5a) (CI-B14.5a) (NADH-ubiquinone oxidoreductase subunit B14.5a)	NDUFA7	113
49	NDUA7_HUMAN			
		NADH dehydrogenase [ubiquinone] 1 alpha subcomplex subunit 3 (Complex I-B9) (CI-B9) (NADH-ubiquinone oxidoreductase B9 subunit)	NDUFA3	84
50	NDUA3_HUMAN			
		Cytochrome c oxidase subunit NDUF44 (Complex I-MLRQ) (CI-MLRQ) (NADH-ubiquinone oxidoreductase MLRQ subunit)	NDUFA4	81
51	NDUA4_HUMAN			
52	NDUB5_HUMAN	NADH dehydrogenase [ubiquinone] 1 beta subcomplex subunit 5	NDUFB5	189
		NADH dehydrogenase [ubiquinone] 1 beta subcomplex subunit 1 (Complex I-MNLL) (CI-MNLL) (NADH-ubiquinone oxidoreductase MNLL subunit)	NDUFB1	58
53	NDUB1_HUMAN			
54	NDUC1_HUMAN	NADH dehydrogenase [ubiquinone] 1 subunit C1	NDUFC1	76
		NADH dehydrogenase [ubiquinone] 1 alpha subcomplex subunit 10	NDUFA10	355
55	NDUAA_HUMAN			
		NADH dehydrogenase [ubiquinone] 1 alpha subcomplex subunit 4-like 2 (NADH-ubiquinone oxidoreductase MLRQ subunit homolog) (NUOMS)	NDUFA4L2	87
56	NUA4L_HUMAN			
57	NDUV3_HUMAN	NADH dehydrogenase [ubiquinone] flavoprotein 3	NDUFV3	108
		NADH dehydrogenase [ubiquinone] 1 beta subcomplex subunit 6 (Complex I-B17) (CI-B17) (NADH-ubiquinone oxidoreductase B17 subunit)	NDUFB6	128
58	NDUB6_HUMAN			
		Complex I intermediate-associated protein 30	NDUFAF1 CIA30 CGI-65	327
59	CIA30_HUMAN			
60	MIMIT_HUMAN	Mimitin	NDUFAF2 NDUF12L	169
		NADH dehydrogenase [ubiquinone] 1 alpha subcomplex assembly factor 3	NDUFAF3 C3orf60	184
61	NDUF3_HUMAN			

		NADH dehydrogenase [ubiquinone] 1 alpha subcomplex assembly factor 4 (Hormone-regulated proliferation-associated protein of 20 kDa)	NDUFAF4 C6orf66 HRPAP20 HSPC125 My013	
62	NDUF4_HUMAN	Succinate dehydrogenase [ubiquinone] cytochrome b small subunit		175
63	DHSD_HUMAN	Succinate dehydrogenase cytochrome b560 subunit	SDHD SDH4	159
64	C560_HUMAN	Succinate dehydrogenase [ubiquinone] iron-sulfur subunit	SDHC CYB560 SDH3	169
65	SDHB_HUMAN	Succinate dehydrogenase [ubiquinone] flavoprotein subunit	SDHB SDH SDH1	280
66	SDHA_HUMAN	Succinate dehydrogenase assembly factor 1	SDHA SDH2 SDHF	664
67	SDHF1_HUMAN	Succinate dehydrogenase assembly factor 2	SDHAF1 LYRM8	115
68	SDHF2_HUMAN	Succinate dehydrogenase assembly factor 4	SDHAF2 C11orf79 PGL2 SDH5	166
69	SDHF4_HUMAN	Succinate dehydrogenase assembly factor 3	SDHAF4 C6orf57	108
70	SDHF3_HUMAN	Cytochrome b (Complex III subunit 3) (Complex III subunit III) (Cytochrome b-c1 complex subunit 3) (Ubiquinol-cytochrome-c reductase complex cytochrome b subunit)	SDHAF3 ACN9 DC11	125
71	CYB_HUMAN	Cytochrome c1	MT-CYB COB CYTB MTCYB	380
72	CY1_HUMAN	Cytochrome b-c1 complex subunit Rieske	CYC1	325
73	UCRI_HUMAN	Cytochrome b-c1 complex subunit 1	UQCRC1	274
74	QCR1_HUMAN	Cytochrome b-c1 complex subunit 2	UQCRC2	480
75	QCR2_HUMAN	Cytochrome b-c1 complex subunit 6	UQCRH	453
76	QCR6_HUMAN	Cytochrome b-c1 complex subunit 7 (Complex III subunit 7) (Complex III subunit VII) (QP-C) (Ubiquinol-cytochrome c reductase complex 14 kDa protein)		91
77	QCR7_HUMAN	Cytochrome b-c1 complex subunit 8 (Complex III subunit 8) (Complex III subunit VIII) (Ubiquinol-cytochrome c reductase complex 9.5 kDa protein) (Ubiquinol-cytochrome c reductase complex ubiquinone-binding protein QP-C)	UQCRB UQBP	111
78	QCR8_HUMAN	Cytochrome b-c1 complex subunit 9 (Complex III subunit 9) (Complex III subunit X) (Cytochrome c1 non-heme 7 kDa protein) (Ubiquinol-cytochrome c reductase complex 7.2	UQCRQ	82
79	QCR9_HUMAN		UQCR10 UCRC HSPC119	63

kDa protein)				
80	QCR10_HUMAN	Cytochrome b-c1 complex subunit 10 (Complex III subunit 10) (Complex III subunit XI) (Ubiquinol-cytochrome c reductase complex 6.4 kDa protein) Putative cytochrome b-c1 complex subunit Rieske-like protein 1 (Ubiquinol-cytochrome c reductase Rieske iron-sulfur subunit	UQCR11 UQCR	56
81	UCRIL_HUMAN	pseudogene 1) Ubiquinol-cytochrome-c reductase complex assembly factor 1 (Basic FGF-repressed Zic-binding protein) (bFGF-repressed Zic-binding protein) (bFZb) (Ubiquinol-cytochrome c reductase complex chaperone CBP3	UQCRFS1P1 UQCRFSL1	283
82	UQCC1_HUMAN	homolog) Ubiquinol-cytochrome-c reductase complex assembly factor 3 (Assembly factor CBP4 homolog)	UQCC1 BZFB C20orf44 UQCC	299
83	UQCC3_HUMAN	Ubiquinol-cytochrome-c reductase complex assembly factor 2 (Breast cancer-associated protein SGA-81M) (Mitochondrial nucleoid factor 1) (Mitochondrial protein M19)	UQCC3 C11orf83 UNQ655/PRO1286	93
84	UQCC2_HUMAN	Complex III assembly factor LYRM7 (LYR motif-containing protein 7)	UQCC2 C6orf125 MNF1	126
85	LYRM7_HUMAN	Cytochrome c oxidase subunit 1 (EC 1.9.3.1) (Cytochrome c oxidase polypeptide I)	LYRM7 C5orf31 MZM1L	104
86	COX1_HUMAN	Cytochrome c oxidase subunit 2 (Cytochrome c oxidase polypeptide II)	MT-CO1 COI COXI MTCO1	513
87	COX2_HUMAN	Cytochrome c oxidase subunit 3 (Cytochrome c oxidase polypeptide III)	MT-CO2 COII COXII MTCO2	227
88	COX3_HUMAN	Cytochrome c oxidase subunit 4 isoform 1	MT-CO3 COIII COXIII MTCO3	261
89	COX41_HUMAN	Cytochrome c oxidase subunit 4 isoform 2	COX4I1 COX4	169
90	COX42_HUMAN	Cytochrome c oxidase subunit 5A	COX4I2 COX4L2	171
91	COX5A_HUMAN	Cytochrome c oxidase subunit 5B	COX5A	150
92	COX5B_HUMAN	Cytochrome c oxidase subunit 6A1	COX5B	129
93	CX6A1_HUMAN	Cytochrome c oxidase subunit 6A2	COX6A1 COX6AL COX6A2 COX6A	109
94	CX6A2_HUMAN		COX6AH	97

95	CX6B1_HUMAN	Cytochrome c oxidase subunit 6B1 (Cytochrome c oxidase subunit VIb isoform 1) (COX VIb-1)	COX6B1 COX6B	86
96	CX6B2_HUMAN	Cytochrome c oxidase subunit 6B2 (Cancer/testis antigen 59) (CT59) (Cytochrome c oxidase subunit VIb isoform 2) (COX VIb-2) (Cytochrome c oxidase subunit VIb	COX6B2	88
97	COX6C_HUMAN	Cytochrome c oxidase subunit 6C (Cytochrome c oxidase polypeptide VIc)	COX6C	75
98	CX7A1_HUMAN	Cytochrome c oxidase subunit 7A1	COX7A1 COX7AH	79
99	CX7A2_HUMAN	Cytochrome c oxidase subunit 7A2	COX7A2 COX7AL	83
100	COX7S_HUMAN	Putative cytochrome c oxidase subunit 7A3	COX7A2P2 COX7A3 COX7AL2 COX7AP2	106
101	COX7B_HUMAN	Cytochrome c oxidase subunit 7B	COX7B	80
102	COX7C_HUMAN	Cytochrome c oxidase subunit 7C	COX7C	63
103	COX7R_HUMAN	Cytochrome c oxidase subunit 7A- related protein	COX7A2L COX7AR COX7RP	114
104	COX8A_HUMAN	Cytochrome c oxidase subunit 8A	COX8A COX8 COX8L	69
105	COX8C_HUMAN	Cytochrome c oxidase subunit 8C	COX8C	72
106	COA1_HUMAN	Cytochrome c oxidase assembly factor 1 homolog (Mitochondrial translation regulation assembly intermediate of cytochrome c oxidase protein of 15 kDa)	COA1 C7orf44 MITRAC15	146
107	COA3_HUMAN	Cytochrome c oxidase assembly factor 3 homolog	COA3 CCDC56 MITRAC12 HSPC009	106
108	COA4_HUMAN	Cytochrome c oxidase assembly factor 4 homolog	COA4 CHCHD8 E2IG2	87
109	COA5_HUMAN	Cytochrome c oxidase assembly factor 5	COA5 C2orf64	74
110	COA6_HUMAN	Cytochrome c oxidase assembly factor 6 homolog	COA6 C1orf31	125
111	COA7_HUMAN	Cytochrome c oxidase assembly factor 7 (Beta-lactamase hcp-like protein) (Respiratory chain assembly factor 1) (Sel1 repeat-containing protein 1)	COA7 C1orf163 RESA1 SELRC1	231
112	COX11_HUMAN	Cytochrome c oxidase assembly protein COX11	COX11	276
113	COX14_HUMAN	Cytochrome c oxidase assembly protein COX14	COX14 C12orf62	57
114	COX15_HUMAN	Cytochrome c oxidase assembly protein COX15 homolog	COX15	410
115	COX16_HUMAN	Cytochrome c oxidase assembly protein COX16 homolog	COX16 C14orf112 HSPC203 PTD019	106
116	COX17_HUMAN	Cytochrome c oxidase copper chaperone	COX17	63

		Mitochondrial inner membrane protein COX18 (COX18Hs)		
117	COX18_HUMAN	(Cytochrome c oxidase assembly protein 18)	COX18 OXA1L2	333
118	COX19_HUMAN	Cytochrome c oxidase assembly protein COX19 (hCOX19)	COX19	90
119	COX20_HUMAN	Cytochrome c oxidase protein 20 homolog	COX20 FAM36A	118
120	AT5EL_HUMAN	ATP synthase subunit epsilon-like protein	ATP5EP2	51
121	AT5F1_HUMAN	ATP synthase F(0) complex subunit B1	ATP5F1	256
122	AT5G1_HUMAN	ATP synthase F(0) complex subunit C1	ATP5G1	136
123	AT5G2_HUMAN	ATP synthase F(0) complex subunit C2	ATP5G2 PSEC0033	141
124	AT5G3_HUMAN	ATP synthase F(0) complex subunit C3	ATP5G3	142
125	ATP5E_HUMAN	ATP synthase subunit epsilon	ATP5E	51
126	ATP5H_HUMAN	ATP synthase subunit d	ATP5H My032	161
127	ATP5J_HUMAN	ATP synthase-coupling factor 6	ATP5J ATP5A ATPM	108
128	ATP6_HUMAN	ATP synthase subunit a (F-ATPase protein 6)	MT-ATP6 ATP6 ATPASE6 MTATP6	226
129	ATP8_HUMAN	ATP synthase protein 8 (A6L) (F-ATPase subunit 8)	MT-ATP8 ATP8 ATPASE8 MTATP8	68
130	ATPA_HUMAN	ATP synthase subunit alpha	ATP5A1 ATP5A ATP5AL2 ATPM	553
131	ATPB_HUMAN	ATP synthase subunit beta	ATP5B ATPMB ATPSB	529
132	ATPD_HUMAN	ATP synthase subunit delta	ATP5D ATP5C1 ATP5C	168
133	ATPG_HUMAN	ATP synthase subunit gamma	ATP5CL1	298
134	ATPK_HUMAN	ATP synthase subunit f	ATP5J2 ATP5JL	94
135	ATPO_HUMAN	ATP synthase subunit O	ATP5O ATPO	213
136	ATPF2_HUMAN	ATP synthase mitochondrial F1 complex assembly factor 2 (ATP12 homolog)	ATPAF2 ATP12 LP3663	289
137	ATPF1_HUMAN	ATP synthase mitochondrial F1 complex assembly factor 1 (ATP11 homolog)	ATPAF1 ATP11	328
138	ATP5S_HUMAN	ATP synthase subunit s	ATP5S ATPW	215
139	ATP5L_HUMAN	ATP synthase subunit g	ATP5L	103
140	AT5L2_HUMAN	ATP synthase subunit g 2	ATP5L2 ATP5K2	100
141	ATP5I_HUMAN	ATP synthase subunit e	ATP5I ATP5K	69
142	AT5SL_HUMAN	ATP synthase subunit s-like protein	ATP5SL	257
143	ADT1_HUMAN	ADP/ATP translocase 1 (ADP,ATP carrier protein 1) (ADP,ATP carrier protein, heart/skeletal muscle isoform T1) (Adenine nucleotide	SLC25A4 ANT1	298

		translocator 1) (ANT 1) (Solute carrier family 25 member 4)		
144	ADT2_HUMAN	ADP/ATP translocase 2 (ADP,ATP carrier protein 2) (ADP,ATP carrier protein, fibroblast isoform) (Adenine nucleotide translocator 2) (ANT 2) (Solute carrier family 25 member 5) [Cleaved into: ADP/ATP translocase 2, N-terminally processed]	SLC25A5 ANT2	298
145	ADT3_HUMAN	ADP/ATP translocase 3 (ADP,ATP carrier protein 3) (ADP,ATP carrier protein, isoform T2) (ANT 2) (Adenine nucleotide translocator 3) (ANT 3) (Solute carrier family 25 member 6) [Cleaved into: ADP/ATP translocase 3, N-terminally processed]	SLC25A6 ANT3 CDABP0051	298
146	ADT4_HUMAN	ADP/ATP translocase 4 (ADP,ATP carrier protein 4) (Adenine nucleotide translocator 4) (ANT 4) (Solute carrier family 25 member 31) (Sperm flagellar energy carrier protein) [Cleaved into: ADP/ATP translocase 4, N-terminally processed]	SLC25A31 AAC4 ANT4 SFEC	315
147	NAC1_HUMAN	Sodium/calcium exchanger 1 (Na(+)/Ca(2+)-exchange protein 1) (Solute carrier family 8 member 1)	SLC8A1 CNC NCX1	973
148	NAC2_HUMAN	Sodium/calcium exchanger 2 (Na(+)/Ca(2+)-exchange protein 2) (Solute carrier family 8 member 2)	SLC8A2 KIAA1087 NCX2	921
149	NAC3_HUMAN	Sodium/calcium exchanger 3 (Na(+)/Ca(2+)-exchange protein 3) (Solute carrier family 8 member 3)	SLC8A3 NCX3	927
150	SL9A1_HUMAN	Sodium/hydrogen exchanger 1 (APNH) (Na(+)/H(+) antiporter, amiloride-sensitive) (Na(+)/H(+) exchanger 1) (NHE-1) (Solute carrier family 9 member 1)	SLC9A1 APNH1 NHE1	815
151	SL9A3_HUMAN	Sodium/hydrogen exchanger 3 (Na(+)/H(+) exchanger 3) (NHE-3) (Solute carrier family 9 member 3)	SLC9A3 NHE3	834
152	SL9A4_HUMAN	Sodium/hydrogen exchanger 4 (Na(+)/H(+) exchanger 4) (NHE-4) (Solute carrier family 9 member 4)	SLC9A4 NHE4	798

153	SL9A2_HUMAN	Sodium/hydrogen exchanger 2 (Na ⁺)/H ⁺ exchanger 2) (NHE-2) (Solute carrier family 9 member 2)	SLC9A2 NHE2	812
154	SL9A5_HUMAN	Sodium/hydrogen exchanger 5 (Na ⁺)/H ⁺ exchanger 5) (NHE-5) (Solute carrier family 9 member 5)	SLC9A5 NHE5	896
155	SL9A7_HUMAN	Sodium/hydrogen exchanger 7 (Na ⁺)/H ⁺ exchanger 7) (NHE-7) (Solute carrier family 9 member 7)	SLC9A7 NHE7	725
156	SL9A8_HUMAN	Sodium/hydrogen exchanger 8 (Na ⁺)/H ⁺ exchanger 8) (NHE-8) (Solute carrier family 9 member 8)	SLC9A8 KIAA0939 NHE8	581
157	SL9A9_HUMAN	Sodium/hydrogen exchanger 9 (Na ⁺)/H ⁺ exchanger 9) (NHE-9) (Solute carrier family 9 member 9)	SLC9A9 NHE9 Nb1a00118	645
158	SL9A6_HUMAN	Sodium/hydrogen exchanger 6 (Na ⁺)/H ⁺ exchanger 6) (NHE-6) (Solute carrier family 9 member 6)	SLC9A6 KIAA0267 NHE6	669
159	EMRE_HUMAN	Essential MCU regulator, mitochondrial (Single-pass membrane protein with aspartate- rich tail 1, mitochondrial)	SMDT1 C22orf32 EMRE	107
160	MCU_HUMAN	Calcium uniporter protein, mitochondrial (Coiled-coil domain- containing protein 109A)	MCU C10orf42 CCDC109A	351
161	MCUB_HUMAN	Calcium uniporter regulatory subunit MCUb, mitochondrial (MCUb) (Coiled-coil domain-containing protein 109B)	CCDC109B MCUB	336
162	MCUR1_HUMAN	Mitochondrial calcium uniporter regulator 1 (Coiled-coil domain- containing protein 90A, mitochondrial)	MCUR1 C6orf79 CCDC90A	359
163	MICU1_HUMAN	Calcium uptake protein 1, mitochondrial (Atopy-related autoantigen CALC) (ara CALC) (Calcium-binding atopy-related autoantigen 1) (allergen Hom s 4)	MICU1 CALC CBARA1	476
164	MICU2_HUMAN	Calcium uptake protein 2, mitochondrial (EF-hand domain- containing family member A1)	MICU2 EFHA1	434
165	NCKX2_HUMAN	Sodium/potassium/calcium exchanger 2 (Na ⁺)/K ⁺ /Ca ²⁺ - exchange protein 2) (Retinal cone Na-Ca+K exchanger) (Solute carrier family 24 member 2)	SLC24A2 NCKX2	661
166	NCKX3_HUMAN	Sodium/potassium/calcium exchanger 3 (Na ⁺)/K ⁺ /Ca ²⁺ - exchange protein 3) (Solute carrier	SLC24A3 NCKX3	644

family 24 member 3)				
167	NCKX4_HUMAN	Sodium/potassium/calcium exchanger 4 (Na(+)/K(+)/Ca(2+)-exchange protein 4) (Solute carrier family 24 member 4)	SLC24A4 NCKX4	622
168	NCKX1_HUMAN	Sodium/potassium/calcium exchanger 1 (Na(+)/K(+)/Ca(2+)-exchange protein 1) (Retinal rod Na-Ca+K exchanger) (Solute carrier family 24 member 1)	SLC24A1 KIAA0702 NCKX1	1099
169	NCKX5_HUMAN	Sodium/potassium/calcium exchanger 5 (Na(+)/K(+)/Ca(2+)-exchange protein 5) (Solute carrier family 24 member 5)	SLC24A5 JSX NCKX5	500
170	NCKX6_HUMAN	Sodium/potassium/calcium exchanger 6, mitochondrial (Na(+)/K(+)/Ca(2+)-exchange protein 6) (Sodium/calcium exchanger protein, mitochondrial) (Solute carrier family 24 member 6) (Solute carrier family 8 member B1)	SLC8B1 NCKX6 NCLX SLC24A6	584
171	MPCP_HUMAN	Phosphate carrier protein, mitochondrial (Phosphate transport protein) (PTP) (Solute carrier family 25 member 3)	SLC25A3 PHC OK/SW-cl.48	362
172	NNTM_HUMAN	NAD(P) transhydrogenase, mitochondrial (EC 1.6.1.2) (Nicotinamide nucleotide transhydrogenase) (Pyridine nucleotide transhydrogenase)	NNT	1086
173	UCP1_HUMAN	Mitochondrial brown fat uncoupling protein 1 (UCP 1) (Solute carrier family 25 member 7) (Thermogenin)	UCP1 SLC25A7 UCP	307
174	UCP2_HUMAN	Mitochondrial uncoupling protein 2 (UCP 2) (Solute carrier family 25 member 8) (UCPH)	UCP2 SLC25A8	309
175	UCP3_HUMAN	Mitochondrial uncoupling protein 3 (UCP 3) (Solute carrier family 25 member 9)	UCP3 SLC25A9	312
176	UCP4_HUMAN	Mitochondrial uncoupling protein 4 (UCP 4) (Solute carrier family 25 member 27)	SLC25A27 UCP4 UNQ772/PRO1566	323
177	UCP5_HUMAN	Brain mitochondrial carrier protein 1 (BMCP-1) (Mitochondrial uncoupling protein 5) (UCP 5) (Solute carrier family 25 member 14)	SLC25A14 BMCP1 UCP5 UNQ791/PRO1682	325
178	SODM_HUMAN	Superoxide dismutase [Mn], mitochondrial (EC 1.15.1.1)	SOD2	222

		Glutathione peroxidase 3 (GPx-3) (GSHPx-3) (EC 1.11.1.9) (Extracellular glutathione peroxidase) (Plasma glutathione peroxidase) (GPx-P)		
179	GPX3_HUMAN	(GSHPx-P)	GPX3 GPXP	226
		Glutathione reductase, mitochondrial (GR) (GRase) (EC 1.8.1.7)		
180	GSHR_HUMAN		GSR GLUR GRD1	522
		Glutathione synthetase (GSH synthetase) (GSH-S) (EC 6.3.2.3)		
181	GSHB_HUMAN	(Glutathione synthase)	GSS	474
182	GLRX2_HUMAN	Glutaredoxin-2, mitochondrial Thioredoxin, mitochondrial (MTRX)	GLRX2 GRX2 CGI-133	164
183	THIOM_HUMAN	(Mt-Trx) (Thioredoxin-2)	TXN2 TRX2	166
		Extracellular superoxide dismutase [Cu-Zn] (EC-SOD) (EC 1.15.1.1)		
184	SODE_HUMAN		SOD3	240
		Superoxide dismutase [Cu-Zn] (EC 1.15.1.1) (Superoxide dismutase 1)		
185	SODC_HUMAN	(hSod1)	SOD1	154