PCSK9 and Its Variants: An Unbiased Global Proteomic Study to Identify Interactors and Effects on Protein Trafficking

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Abstract

Proprotein convertase subtilisin/kexin type 9 (PCSK9) is a secreted glycoprotein that promotes degradation of low-density lipoprotein receptors. Gain- and loss-of-function variants of PCSK9 cause hypercholesterolemia and hypocholesterolemia, respectively. Although it has been a decade since the discovery of PCSK9, its effect in terms of global protein changes and interactions still require further understanding. This study provided a global outlook at the protein changes caused by PCSK9 and its variants in human hepatic HUH7 cell line. First, a proteomics-based method for protein subcellular distribution analysis has been developed. Second, through secretome analyses, six apolipoproteins and six proteins involved in the coagulation pathway were found with >2-fold changes between wild type PCSK9 and its variants. Third, through secreted interactome analyses, a list of 159 PCSK9 interactor candidates was identified. Two interacting proteins, FASN and PSMD2, were validated and demonstrated with dynamic interacting patterns between PCSK9 and its variants.

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List of Abbreviations

CAD	Coronary artery disease
LDL	Low density lipoprotein
ApoB-100	Apolipoprotein B-100
LDLR	Low-density lipoprotein receptor
ARH	Autosomal recessive hypercholesterolemia
FH	Familial Hypercholesterolemia
PCSK9	Proprotein convertase subtilisin kexin type 9
ADH	autosomal dominant hypercholesterolemia
PC	Proprotein convertase
PCSK	Proprotein convertase subtilisin/kexin
NARC-1	Neural apoptosis-regulated convertase-1
ER	Endoplasmic reticulum
CHRD	Cys/His-rich Domain
NF-Y	nuclear factor-Y
Sp-1	specificity factor-1
SRE	sterol regulatory element
HNF1α	Hepatocyte Nuclear Factor
SREBP	Sterol Regulatory Element Binding Protein
LXR	liver X receptor
PPARα	peroxisome proliferator activated receptor alpha
LDLC	Low-density lipoprotein cholesterol
E2	estradiol
FXR	Farnesoid X Receptor
BBR	berberine
EGF-A	epidermal growth factor-like repeat A
APLP2	Amyloid beta precursor-like protein 2
cIAP1	cellular inhibitor of apoptosis protein 1
ApoER2	apolipoprotein E receptor 2
BACE1	beta-site amyloid precursor protein-cleaving enzyme 1
ENaC	Na^+ channel
VLDLR	very low density lipoprotein receptor
GOF	gain-of-function
LOF	Loss-of-function
BLA	Biologics License Application
DMEM	Dulbecco's Modified Eagle Medium
FBS	Fetal bovine serum
SILAC	Stable isotope labelling of amino acids in cell culture
EGFP	Enhanced Green Fluorescent Protein
PBS	Phosphate buffered saline
G418	geneticin

MS	mass spectrometry
TLC	total cellular lysates
TBS-T	Tris Buffered Saline-Tween 20
EDTA	Ehylenediamine tetraacetic acid
ELISA	Enzyme Linked Immunosorbent Assay
ACN	Acetonitrile
DTT	dithiothritol
ABC	ammonium bicarbonate
IAA	iodoacetimide
FA	formic acid
FASP	filter-aided sample preparation
TFA	trifluoroacetic acid
MS/MS	Tandem mass spectrometry
HPLC	High performance liquid chromatography
SDS-PAGE	Sodium dodecyl sulphate polyacrylamide gel electrophoresis
LTQ	Linear trap quadrupole
FDR	false discovery rate
LFQ	label free quantified
SEM	standard error of the mean
IPA	Ingenuity Pathway Analysis
SCX	Strong Cationic Exchange
R	Arginine
K	Lysine
MK	Mock
EV	Empty Vector
QH	Q152H
AV	L10insA53V
RL	R46L
WT	Wild type
SR	S127R
DY	D374Y

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1. Introduction

1.1. Coronary Artery Disease and Hypercholesterolemia

Coronary artery disease (CAD) is one of the major causes of mortality worldwide [1]. People develop CAD because of lipid deposits of plaque building up along the inner walls of the arteries of the heart, narrowing down the lumen of arteries and resulting in insufficient blood supply to the heart. A number of risk factors of CAD have been determined including hypercholesterolemia, characterized by elevated levels of plasma low density lipoprotein (LDL) cholesterol [2]. In the human body, the LDL particle is the major carrier of cholesterol in blood and can deposit in the artery [3]. Low density lipoprotein particles contain triglycerides, cholesterol esters and other fats in its core and non-esterified cholesterol, a single apolipoprotein B-100 (ApoB-100) molecule, additional ancillary proteins and phospholipids as a shell [4].

1.1.1. Low Density Lipoprotein Receptors in Cholesterol Homeostasis

Approximately 70% of plasma LDL particles are cleared by hepatic LDL receptors (LDLRs) in the liver [5]. They are type 1 cell-surface receptors [6] whose primary function is to bind the LDL particle through interaction with ApoB-100 and mediate LDL endocytosis [7]. Cell surface LDLRs associate with clathrin-coated pits through the autosomal recessive hypercholesterolemia (ARH) protein, an adaptor protein that binds to the intracellular domain of the LDLR and to clathrin [8, 9]. It is via ARH that the LDLR:LDL complex is internalized in vesicles formed by clathrin-coated pits [10-12]. Then the vesicle will shed its clathrin coat and fuse with acidic recycling endosomes where LDLRs release the LDL particles due to a conformational change in LDLR associated with the drop of pH [13]. The

LDLRs are recycled back to the cell surface, while the LDL particles moves to late endosomes and onto lysosomes [7].

1.1.2. Familial Hypercholesterolemia

The most common and severe inherited form of hypercholesterolemia is familial hypercholesterolemia (FH). This disorder is inherited in an autosomal dominant manner and occurs more frequently in certain populations, including Afrikaners in South Africa, French Canadians, Christian Lebanese and those of Ashkenazi Jewish descent [14]. Variations in genes encoding LDLR and ApoB-100 are the two most common genetic defects causing FH, accounting for approximately 80% of all FH cases [15]. Variants of LDLR as heritable risk factors of FH were identified in 1974 [16], associating with impaired binding to LDL particles and internalization of LDL particles [17, 18]. The human *LDLR* gene was mapped by 1984 [19], and since then many *LDLR* variations causing FH have been identified (and classified as FH1). Shortly after, variations in the *APOB* gene were identified to cause defective binding with LDLR and reduced LDL clearing as a second form of FH (FH2) [20, 21]. In 2003, variations of the gene *PCSK9* (*Proprotein convertase subtilisin/kexin type 9*) were identified within two French families that result in autosomal dominant hypercholesterolemia (ADH) and *PCSK9* was classified as the third gene causing FH (*FH3*) [22].

1.2. Proprotein Convertase Family

Proprotein convertase subtilisin/kexin type 9 (PCSK9) is a member of the mammalian secretory serine proprotein convertase (PC or PCSK) family [23] that cleaves protein precursors, activating or deactivating biological molecules including prohormones, cytokines, transcription factors, cell surface receptors, and secreted proteins [24, 25]. The first discovered proprotein (prohormone) was the precursor form of insulin (proinsulin) by Donald Steiner in 1967 [26]. At the same time, another hormone, β-melanocyte-stimulating hormone was found within the β-lipotrophic hormone by Michel Chrétien and colleagues[27]. These studies evidenced that peptide hormones could exist in larger protein molecules. The enzyme responsible for processing proproteins to peptide hormones was identified decades after, when kexin (a product of gene *KEX2*) was discovered to process alpha factor mating pheromone by David Julius in 1984 [28]. Then the partial sequence of the *KEX2*-homologous *FURIN* gene was identified as the first mammalian PC in 1989 [29].

Nine members have been discovered in the mammalian PC family, namely PCSK1 (PC1/3), PCSK2 (PC2), PCSK3 (furin), PCSK4 (PC4), PCSK5 (PC5/6), PCSK6 (PACE4), PCSK7 (PC7), PCSK8 (S1P/SKI-1) and PCSK9 (NARC-1). These PCs belong to the subtilisin family of serine proteases [23]. The first seven belong to the yeast kexin subfamily of subtilases and function by cleavage following monobasic and dibasic residues within the motif $(K/R)X_n(K/R)\downarrow$, in which n=0, 2, 4, or 6 [30-32]. PCSK8 belongs to the pyrolysin subfamily of subtilases and cleaves following non-basic residues in motifs RX(hydrophobic/aliphatic)X↓ [33-35], while PCSK9 belongs to the proteinase K subfamily of subtilases [23] and cleaves following an acidic VFAQ↓ motif [36]. The PCSKs share similar structures. They consist of a signal peptide, a prodomain, a catalytic domain, a P-domain, and a C-terminal domain [37]. PCSK3, PCSK5B, PCSK6, PCSK7, and PCSK8 also possess a transmembrane domain [21, 38-41]. The signal peptide directs the newly synthesized protein to endoplasmic reticulum (ER), where the prodomain is autocatalytically cleaved and remains attached to the rest of the protein as an endogenous inhibitor and an intracellular chaperone, assisting the protein folding and transit from the ER to Golgi apparatus [37, 42]. The propeptide is released by a second cleavage en route of secretion, activating PCs enzymatic activity [43]. It is important to note that PCSK9 is the only PC that does not function as an enzyme after secretion because it lacks the second cleavage within its prodomain [23]. The catalytic domain is highly conserved throughout the PCSKs family [37]. The P-domain provides stability to the enzyme [37]. The C-terminal domain is involved in protein trafficking, protein stability, and protein-protein interactions [43].

1.3. PCSK9 Biology

1.3.1. PCSK9 Biosynthesis

The *PCSK9* gene encodes a 692 amino acid preproprotein, with a signal peptide sequence (1-30 aa), an inhibitory prodomain (31-152 aa), a catalytic domain (153-425 aa) and a His/Cys-rich C-terminal domain (CHRD; 426-692aa) [43] (Fig. 1). PCSK9 undergoes Ca^{2+}-independent autocatalytic processing early in the ER from proPCSK9 (~72kDa) to PCSK9 (~63kDa) at site $SSVFAQ_{152}$↓SIP [36]. This cleavage of the prodomain is required for PCSK9 trafficking out of ER to the Golgi apparatus [36]. However, as mentioned above, unlike the other convertases [44], the cleaved propeptide remains non-covalently attached to PCSK9, even after secretion, precluding its enzymatic activity [36]. PCSK9 is N-glycosylated at Asn-533 in the ER and further matured in the Golgi [23]. It is sulfated in the late Golgi at Tyr-38 [36, 45, 46]. It is phosphorylated at Ser-47 and Ser-688 and secreted as a PCSK9:propeptide complex [47]. In 2006, Benjannet and colleagues reported that along the late secretory pathway, PCSK9 is further cleaved within its catalytic domain by membrane-bound furin (PCSK3) and soluble PCSK5/6A at R218, and this so-called furin-cleaved PCSK9 form was also found in human plasma [45].

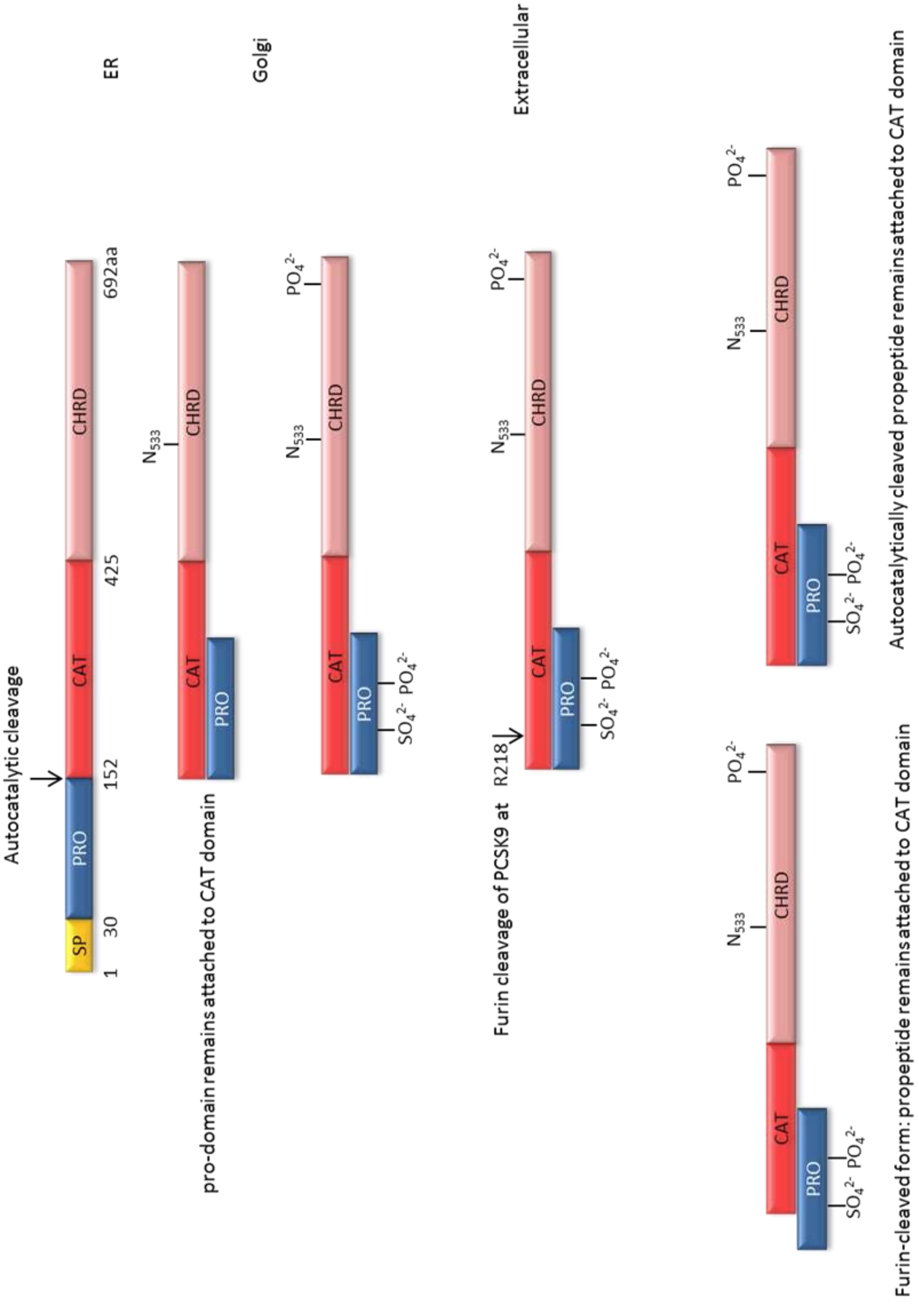

Autocatalytic cleavage
SP
PRO
CAT
CHRD
1
30
152
425
692aa
ER
pro-domain remains attached to CAT domain
N_{533}
Golgi
SO_4^{2-}
PO_4^{2-}
Extracellular
Furin cleavage of PCSK9 at R218
Furin-cleaved form: propeptide remains attached to CAT domain
Autocatalytically cleaved propeptide remains attached to CAT domain

Figure 1. PCSK9 biosynthesis. PCSK9 autocatalytically cleaves its prodomain at VFAQ$_{152}$ ↓ SIP in the ER, and the propeptide remains attached to catalytic domain after secretion. PCSK9 is glycosylated at Asn533 in the ER and further matured in the Golgi. PCSK9 is phosphorylated at Ser47 and Ser688, and sulfated at Tyr38 in the Golgi. At cell surface, PCSK9 is further cleaved by furin at Arg218, and the cleaved N-segment remains attached to catalytic domain. SP, signal peptide; PRO, prodomain; CAT, catalytic domain; CHRD, cysteine-histidine rich domain; SO_4^{2-}, sulfated; PO_4^{2-}, phosphorylated; N, Asn-linked glycosylation.

1.3.2. PCSK9 Expression and Regulation

In humans the level of plasma PCSK9 varies over a 100-fold range between 33–2988 ng/ml (median 487 ng/ml), and PCSK9 levels are significantly higher in women than in men and significantly higher in postmenopausal women compared to premenopausal women [48]. PCSK9 is highly expressed in the liver and small intestine [23], but the circulating PCSK9 is mainly contributed by the liver [49]. *PCSK9* is a 12 exon gene in chromosome 1 (1p32) with several regulatory elements in its proximal promoter, namely nuclear factor-Y (NF-Y), specificity factor-1 (Sp-1), sterol regulatory element (SRE) [50, 51], E-box, and hepatocyte nuclear factor (HNF1α) binding site [52]. The expression of *PCSK9* mRNA is up regulated by statin co-directionally with *LDLR* via the interaction of transcription factor sterol regulatory element binding protein 2 (SREBP-2) with the SRE motif in *PCSK9*'s proximal promoter [53-56]. Its expression is also induced by insulin in rodent primary hepatocytes and mice, via SREBP-1c [50]. The SREBP-2 and SREBP1-c are two isoforms of the SREBPs family who also contains another member, SREBP-1a. They activate genes involved in fatty acid synthesis, lipid and cholesterol biosynthesis [57-61]. SREBP-2 is relatively specific to cholesterol synthesis [58], while SREBP-1c is involved in fatty acid synthesis and insulin induced glucose metabolism [57]. SREBP-1a is implicated in both pathways [62]. In cell studies, both SREBP1 and SREBP2 transcriptionally activate PCSK9 via the SRE motif [54]. *In vivo*, however, SREBP-2 has been indicated as the primary mediator of PCSK9 regulation [63]. *PCSK9* expression is also moderately up-regulated by the liver X receptor (LXR) agonist T0901317 [64]. One study investigating the effect of fenofibrate (a drug used to reduce cholesterol levels as PPARα agonists) on PCSK9 levels by Kourimate and colleagues demonstrated that PCSK9 could be down regulated through the peroxisome proliferator activated receptor alpha (PPARα) pathway, repressing its promoter activity [65]. Whereas in the study conducted by Mayne and colleagues PCSK9 changes upon fenofibrate treatment in humans were inversely correlated to changes observed in LDL-Cholesterol (LDLC), indicating that fibrates affect PCSK9 expression indirectly through its modulation of cholesterol levels [66].

In a rat model, PCSK9 expression is down regulated by treatment with glucagon, high-doses of ethinylestradiol and dietary cholesterol, resulting in 70%, 50% and 60-70% of reduction in PCSK9 mRNA expression, respectively [67]. Ooi and colleagues found that in women, estradiol (E2) level correlated inversely with circulating PCSK9 and directly with serum LDLC, but E2 replacement therapy did not have any effect on circulating PCSK9, suggesting that the E2-related differences in PCSK9 levels may be the result of differences in receptor-mediated PCSK9 clearance through E2-induced changes rather than production of PCSK9 [68]. Starr and colleagues found that β-estradiol decreased PCSK9 phosphorylation level without changing its protein level and induced up-regulation of LDLR in a PCSK9-dependent manner in HUH7 cells, suggesting the existence of PCSK9 interactors that switch PCSK9's functional states towards or away from LDLR degradation [69]. Another study conducted by Langhi and colleagues has demonstrated that *PCSK9* expression is repressed through the bile acids–activated activation of Farnesoid X Receptor (FXR) by its synthetic specific agonist GW4064, and that the statin-induced PCSK9 up-regulation could be prevented by co-administration with the bile acid Chenodeoxycholic acid [70]. The natural cholesterol-lowering compound berberine (BBR) has been reported to have a reducing effect on PCSK9 mRNA and protein level [71] while it up-regulates LDLR expression [72, 73]. In the investigation of the mechanism of the regulation of PCSK9 expression by BBR, Li and colleagues have identified the HNF1α binding site upstream of SRE on PCSK9, whose variation causes over 90% reduction on *PCSK9* promoter activity [52, 71]. They also concluded that HNF1α site works cooperatively with SRE, and BBR suppresses *PCSK9* expression through coordinate reduction of HNF1α and SREBP2 [52, 71].

1.3.3. PCSK9's Effects on Cholesterol Homeostasis and Mechanism

Because the propeptide of PCSK9 remains attached to PCSK9 and inactivates its enzymatic activity, there are no substrates known other than itself for cleavage [23, 36]. Instead, PCSK9 plays an important role in the regulation of plasma cholesterol homeostasis as an escort protein of LDLRs [23]. This is illustrated in Figure 2. In fact, parallel and reciprocal regulation between LDLR and PCSK9 was proposed by Tavori and colleagues, that is, beyond PCSK9 promoting LDLR degradation, PCSK9 is cleared largely from circulation through LDLR [74].

As mentioned earlier, *PCSK9* variations were first found to cause ADH in humans in 2003 [22]. In 2004, Maxwell and colleagues demonstrated that adenovirus-mediated overexpression of PCSK9 in mice caused 72% decrease in liver protein LDLR but not the mRNA, and increased plasma LDLC concentrations to levels similar of those of LDLR-knockout mice [75]. In 2005, Rashid and colleagues carried out studies in PCSK9 knockout mice and showed that protein LDLR was up-regulated, but not the mRNA [55]. These studies demonstrated that PCSK9 promotes post-transcriptional degradation of LDLR. In 2007, PCSK9's catalytic domain was found to directly bind the epidermal growth factor-like repeat A (EGF-A) domain of the LDLR at cell surface and binds to LDLR with a ~150-fold higher affinity at an acidic endosomal pH [76-78]. This interaction resulted in the reroute of LDLR from recycling endosomes to lysosome and degradation [77]. Several groups predict

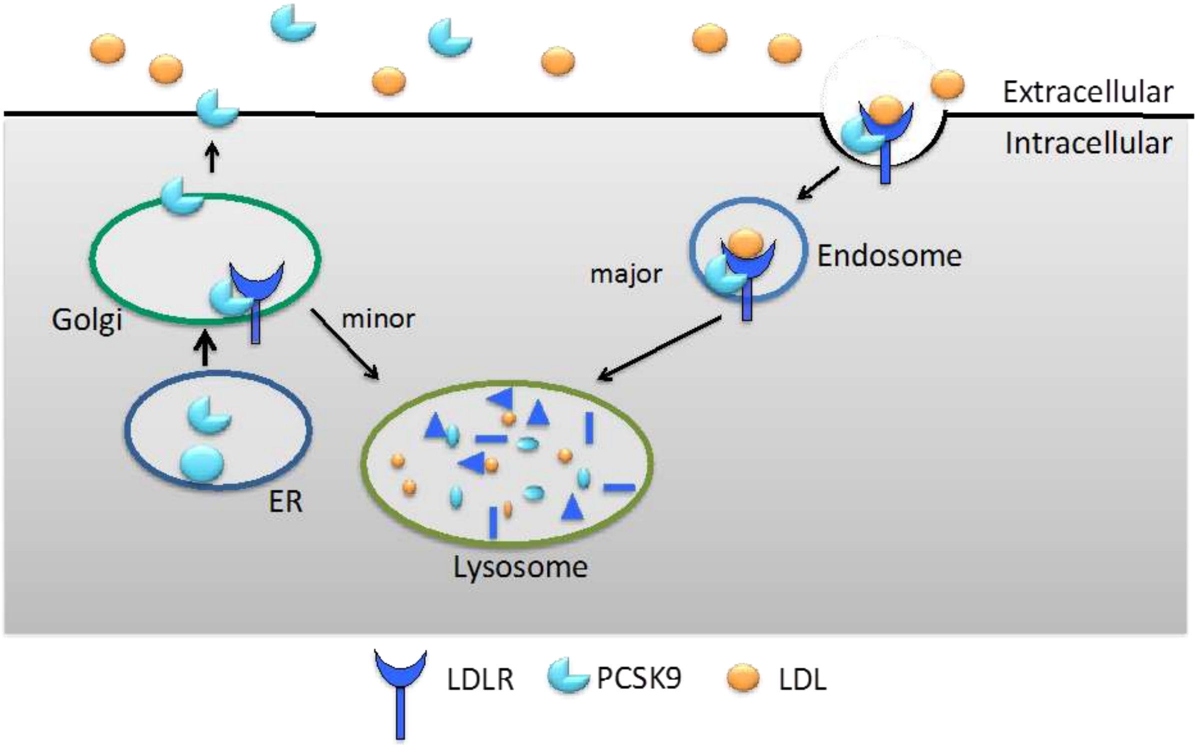

Extracellular
Intracellular
Endosome
major
Golgi
minor
ER
Lysosome
LDLR
PCSK9
LDL

Figure 2. Schematic of PCSK9 mediated LDLR degradation. PCSK9 directly binds to LDLR at cell surface, preventing the receptor from being recycled and inducing its degradation in lysosomes (major pathway). Intracellularly, PCSK9 also promotes LDLR degradation (minor pathway) through a Golgi-lysosomal pathway.

that the binding of PCSK9 may keep LDLR in an open conformation that is suggested to promote lysosomal sorting [79-81]. The c-terminal CHRD of PCSK9 has been reported to be important in the lysosomal targeting but is not required for PCSK9 interaction with LDLR [80, 82, 83]. Lack of glycosylation or sulfation does not affect overexpressed PCSK9 enhanced LDLR degradation in cells [45]. One group proposed that an intracellular Golgi-lysosomal pathway also contributes to PCSK9-mediated LDLR degradation [84]. However studies using PCSK9 antibodies that do not pass into the cell but remain in circulation have shown that they are very effective at inhibiting PCSK9-mediated LDLR degradation, pointing to cell surface to lysosome (via endosomes) as PCSK9's primary route of regulating LDLR levels [85-87]. Furin cleavage was found to inactivate PCSK9's activity in promoting LDLR degradation [88, 89].

1.3.4. PCSK9 Interactors and Effects on LDLR Degradation

As discussed the exact mechanism of how PCSK9 promotes LDLR degradation and regulates cholesterol homeostasis have not been fully understood. Several proteins have been identified or implied to interact with PCSK9 and to affect or be involved in PCSK9 mediated LDLR degradation.

Identified Interactors: Annexin A2 and ApoB-100 have been identified to interact with PCSK9 and inhibit its action towards LDLR (Fig. 3). At the cell surface, the N-terminal of Annexin A2 interacts with the C-terminal of PCSK9 and, as a competitive binder, prevents PCSK9 from interacting with LDLR [90]. PCSK9 was also found to interact with ApoB-100 both intracellularly and in conditioned media, preventing ApoB-100's degradation regardless of LDLR expression [91]. An *in vivo* study showed that 42±3% of total circulating PCSK9 isolated from normolipidemic human plasma was associated with LDL particles through

ApoB-100 [92]. In FH homozygotes plasma PCSK9 was significantly decreased by 55–56% after a single LDL- apheresis treatment, and by 46–56% in FH heterozygotes, indicating PCSK9's association to LDL in FH population [93]. The LDL-ApoB-100:PCSK9 binding inhibited PCSK9 mediated LDLR degradation in HUH7 cells [92]. During secretion, COPII-coated vesicle component Sec24A has been identified as an indirect interactor of PCSK9 in the process of vesicular packaging and PCSK9 subcellular trafficking from ER to Golgi, with SEC24A-deficient mice exhibiting lower plasma PCSK9 and hence increased LDLR levels [94]. Amyloid beta precursor-like protein 2 (APLP2), a ubiquitously expressed cell surface protein known to be involved in lysosomal trafficking [95], has been shown to interact with PCSK9's CHRD domain and mediate its endosome-lysosome trafficking, suggesting a bridging role of PCSK9 for LDLR and lysosomally targeted transmembrane proteins [96]. The cellular inhibitor of apoptosis protein 1 (cIAP1) was found to bind and promote pro-PCSK9 processing and also act as an E3 ligase for ubiquitination of PCSK9, indicating its role on both secretion of PCSK9 and its lysosomal localization [97].

Implied Interactors: Studies showed that an LDLR variant lacking its cytoplasmic domain [98] or a chimeric LDLR with a transferrin receptor cytoplasmic domain [99] were degraded in the presence of PCSK9, suggesting that LDLR's cytoplasmic domain is not involved in the endosomal sorting machinery in PCSK9-mediated degradation of the LDLR and implying a role for extracellular and/or other transmembrane accessory proteins in PCSK9-mediated LDLR rerouting [98, 99]. In addition, an antibody that binds to the CHRD domain of secreted PCSK9 was shown to inhibit PCSK9: LDLR complex internalization without blocking their binding activity, suggesting the PCSK9 CHRD domain is involved in the complex's internalization likely through interacting with other secreted or cell surface

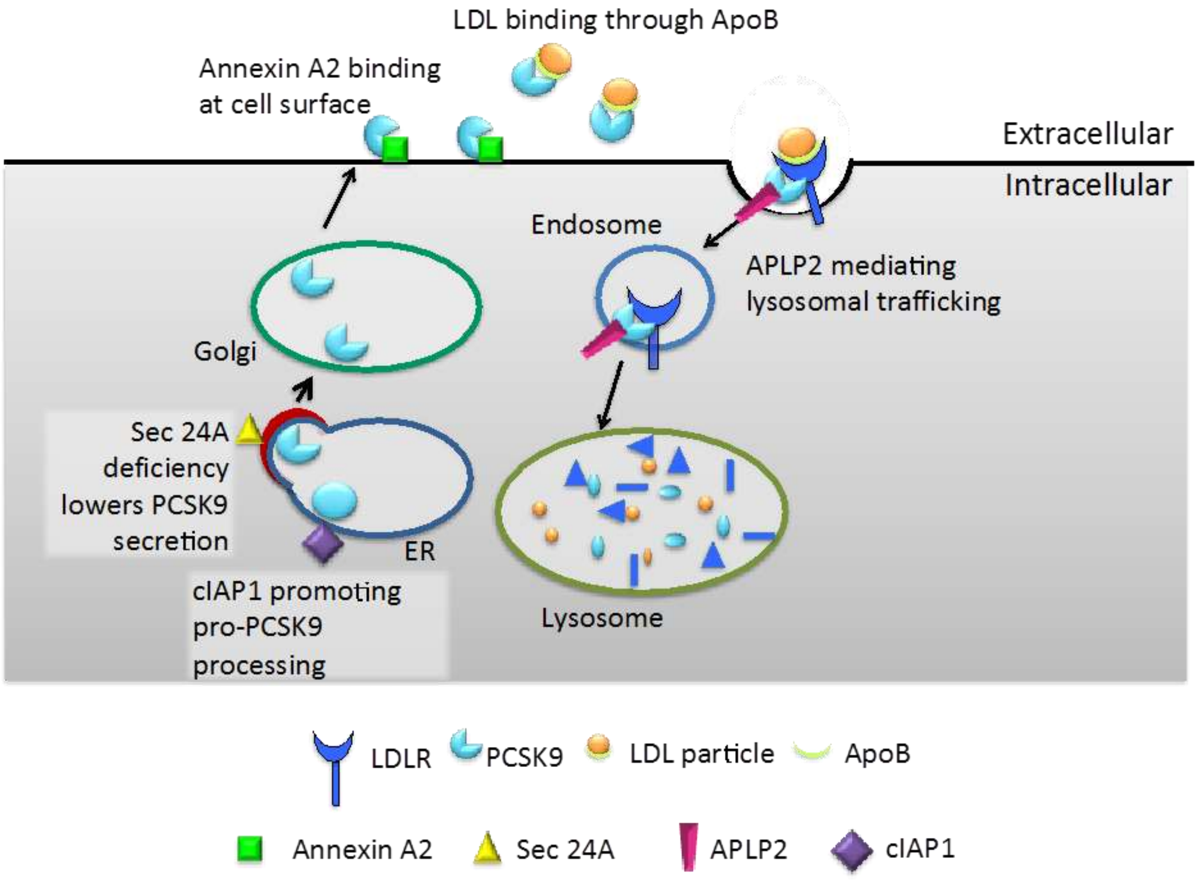

LDL binding through ApoB
Annexin A2 binding at cell surface
Extracellular
Intracellular
Endosome
APLP2 mediating lysosomal trafficking
Golgi
Sec 24A deficiency lowers PCSK9 secretion
ER
cIAP1 promoting pro-PCSK9 processing
Lysosome
LDLR
PCSK9
LDL particle
ApoB
Annexin A2
Sec 24A
APLP2
cIAP1

Figure 3. Schematic of direct and indirect interactors of PCSK9. Annexin A2 and LDL both interact with PCSK9 and inhibit PCSK9's action towards LDLR. CIAP1 promotes proPCSK9 processing. Sec24A was found to regulate PCSK9 trafficking on the secretory pathway while APLP2 mediates PCSK9 lysosomal trafficking. PCSK9 self-association has also been found.

proteins [100]. Furthermore, in PCSK9-resistent fibroblast cells, the binding affinity of PCSK9 and LDLR was reduced in endosomes and LDLR was recycled back to cell surface, with or without bound PCSK9, suggesting the existence of cell-specific intracellular interacting proteins that are involved in PCSK9 binding LDLRs and PCSK9 lysosomal trafficking [101]. Finally, Wooten and colleagues demonstrated that excessive levels of secreted PCSK9 were not sufficient to induce LDLR: PCSK9 complex formation in cell culture, indicating the existence of secreted effector(s) or inhibitor(s) of PCSK9 [102].

1.3.5. PCSK9's Role beyond Cholesterol Regulation

Besides regulating cholesterol homeostasis through inducing LDLR degradation, PCSK9 has been implied to play a role in other pathways and activities such as neuronal apoptosis, neuron differentiation, Na^+ reabsorption and blood pressure regulation, viral infection, glucose metabolism and visceral fat accumulation. PCSK9 has been suggested to have a dual regulatory effect on neuronal apoptosis by Wu and colleagues [103]. On the one hand, PCSK9 plays a role in promoting neuronal apoptosis determined by the observations that PCSK9 potentiates neuronal apoptosis via enhancing degradation of apolipoprotein E receptor 2 (ApoER2) which is required for protecting neuron cells from apoptosis [104, 105] and PCSK9 siRNA inhibits human umbilical vein endothelial cells apoptosis induced by oxidized LDL, one of the key apoptosis inducers in the brain in neurodegenerative diseases [106]. On the other hand, PCSK9 inhibits neuronal apoptosis indicated by the study showing that PCSK9 is required for the disposal of non-acetylated beta-site amyloid precursor protein-cleaving enzyme 1 (BACE1), a rate-limiting enzyme in the development of Alzheimer's disease [107]. However, other studies have shown no association between PCSK9 and neuronal apoptosis [108-110]. Seidah and colleagues showed that PCSK9 overexpression in mice primary embryonic telencephalon cells resulted in higher percentage of differentiated neurons, implying its role in neuron differentiation [23]. PCSK9 has been found to be involved in liver regeneration as its mRNA level increased during induction of liver regeneration after partial hepatectomy [23, 49]. ProPCSK9 was found to bind and enhance proteasomal degradation of full-length Na^+ channel (ENaC) in cell culture, implying a role for PCSK9 in reducing Na^+ reabsorption and down regulating blood pressure [111]. While in mouse models of hypertension PCSK9-deficiency does not alter blood pressure and sodium balance [112], a recent study implied the association of multiple *PCSK9* variants with blood pressure in an African-American population at high risk for cardiovascular disease [113], suggesting further validation of PCSK9's effect on blood pressure. PCSK9 is also involved in impeding viral infection by down-regulating the expression level of mouse liver CD81, a major HCV receptor in cell culture and *in vivo* [114]. Mbikay and colleagues showed that PCSK9-null mice exhibit glucose-intolerance and malfunction in their islets, suggesting PCSK9 plays an important role in normal pancreatic functioning [115]. In the contrast, Langhi and colleagues demonstrated no detectable alteration of glucose homeostasis in PCSK9-null mice, including glucose-induced insulin secretion *in vitro* on primary cultured pancreatic islets and *in vivo* on whole-body animals [116]. The reason for the conflicting results are unclear but may be due to the genetic background of the mice used [117]. To date, the effect of PCSK9-deficiency on glucose metabolism in mice remains controversial. PCSK9 overexpression in cell culture resulted in enhanced degradation of very low density lipoprotein receptor (VLDLR) [105], and PCSK9-null mice exhibit increased level of VLDLR accumulation at the surface of adipocytes, leading to increased visceral fat accumulation [118], suggesting that PCSK9 regulates fat accumulation in adipocytes.

Proteome- and genome-wide studies have also revealed that PCSK9 affects protein level or gene expression in activities and pathways outside of LDLR metabolism [119, 120]. In the proteomic subcellular study conducted by Denis and colleagues, several proteins were found to be up- or down-regulated by a PCSK9 membrane-bound chimera with known enhanced capacity to promote LDLR degradation [119]. These proteins are involved in actin cytoskeleton distribution/transport of endosomes and lysosomes, receptor-mediated endocytosis, SREBP-2 activity, protein folding activity, cell signaling events and so on [119]. In the microarray analyses to identify genome-wide expression changes and pathways affected by overexpression of the gain-of-function (GOF) PCSK9 mutant D374Y in the HepG2 cells, several gene expression pathways were identified beyond cholesterol metabolism including "protein ubiquitination," "xenobiotic metabolism," "cell cycle," and "inflammation and stress response" [120].

Collectively, these studies have suggested a broader role for PCSK9 beyond cholesterol metabolism that should be investigated fully as we move toward PCSK9 antagonism in dyslipidemic therapies.

1.3.6. PCSK9 Variants

PCSK9 is a highly polymorphic gene. Loss-of-function (LOF) variants associate with different levels of hypocholesterolemia and protection from developing CAD (Fig. 4). In the Dallas Heart study conducted by Cohen and colleagues, African-Americans harboring nonsense variants (Y142X and C679X) of PCSK9 exhibited a 28% decrease in LDLC levels and a 88% lower risk of CAD, whereas Caucasians with milder variants (R46L) exhibited a 15% decrease in LDL levels and a 47% lower risk of CAD [121]. Furthermore, people have been identified without any immunodetectable circulating PCSK9 that are healthy, elucidating the safety of drugs targeting PCSK9 [122, 123].In the contrast, GOF PCSK9 variants associate with hypercholesterolemia and carriers are at greater risk to develop CAD (Fig. 4) [22]. In addition, in Caucasian Canadians, people carrying combinations of LOF PCSK9 variants have been identified with combined effects of the variations on LDLC levels [124]. So far, over 150 variants of PCSK9 have been identified in humans as recorded in the list of naturally occurring PCSK9 variants in the website:

http://www.ucl.ac.uk/ldlr/LOVDv.1.1.0/index.php?select_db=PCSK9

So far, most of the variant studies have been population studies in terms of their effects on LDLC but not their mechanisms. Their multiple mechanisms of action or inaction remain to be further studied.

1.3.7. PCSK9 as Pharmaceutical Target

Although statin has been the most commonly used drug for controlling plasma cholesterol level and reducing CAD risk, it is not always sufficiently effective to achieve cholesterol lowering goals especially in patients with inherited forms of dyslipidemia such as familial hypercholesterolemia [125]. As well, statin treatments have caused side-effects with potentially severe consequences, most prominently myopathy or rhabdomyolysis and polyneuropathy [126]. The regulatory role of PCSK9 in plasma cholesterol homeostasis through mediating LDLR degradation, the various effects of GOF and loss of function (LOF) PCSK9 variants on promoting or preventing cardiovascular disease, and the fact that people without any immune-detectable circulating PCSK9 are healthy have led to the exploration of controlling PCSK9 levels and its interaction with LDLR as pharmaceutical goals in blood cholesterol lowering medications beyond statins [23, 121-123]. Three drug development approaches have been carried out by the pharmaceutical industry to test their inhibiting

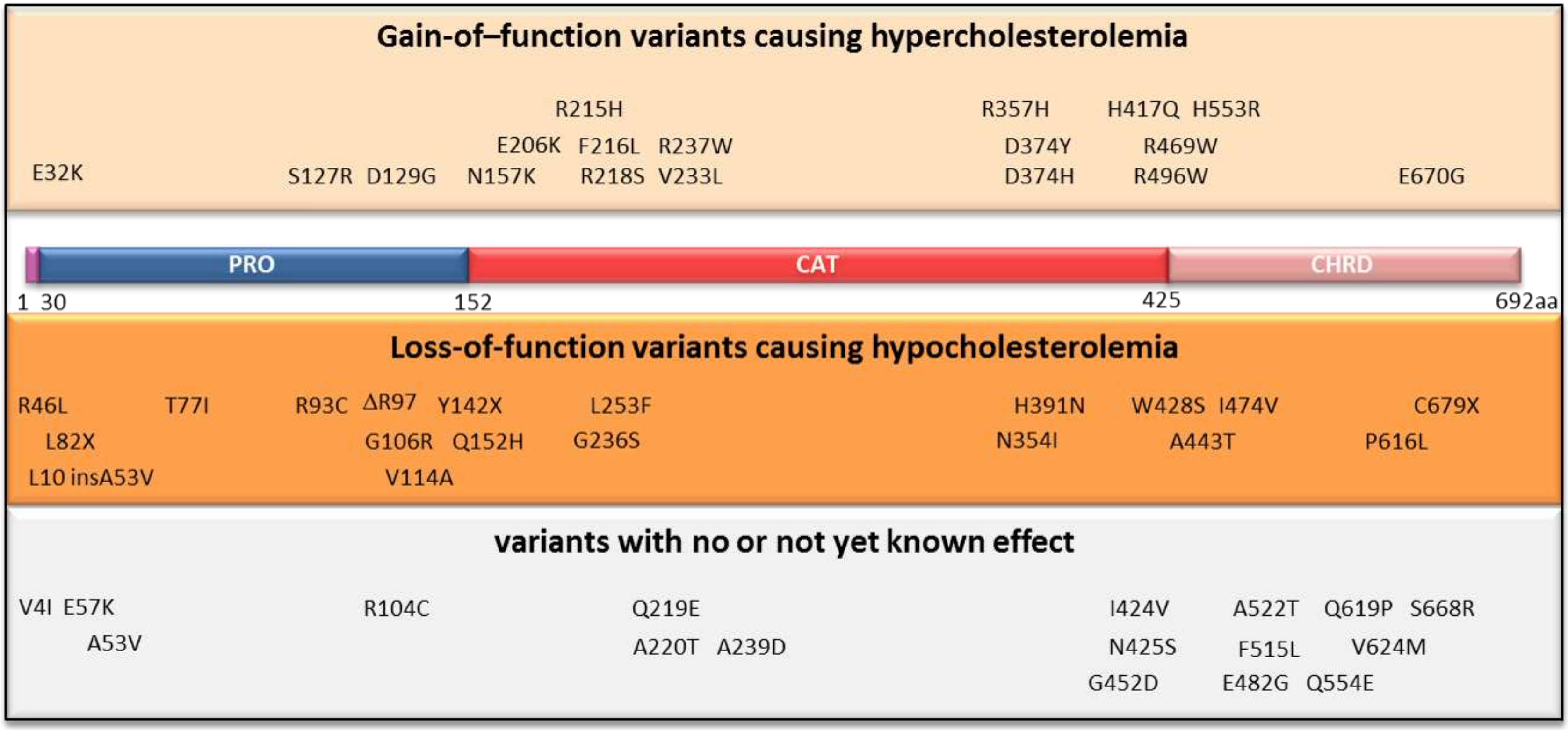

Gain-of–function variants causing hypercholesterolemia
R215H
R357H
H417Q
H553R
E206K
F216L
R237W
D374Y
R469W
E32K
S127R
D129G
N157K
R218S
V233L
D374H
R496W
E670G
PRO
CAT
CHRD
1
30
152
425
692aa
Loss-of-function variants causing hypocholesterolemia
R46L
T77I
R93C
ΔR97
Y142X
L253F
H391N
W428S
I474V
C679X
L82X
G106R
Q152H
G236S
N354I
A443T
P616L
L10 insA53V
V114A
variants with no or not yet known effect
V4I
E57K
R104C
Q219E
I424V
A522T
Q619P
S668R
A53V
A220T
A239D
N425S
F515L
V624M
G452D
E482G
Q554E

Figure 4. Schematic of PCSK9 domains and the relative position of variants on the protein. PCSK9 Loss-of-function (LOF) variants associating with hypocholesterolemia, gain-of-function (GOF) PCSK9 variants associating with hypercholesterolemia. Many variants have also been identified with no or not yet known effects on LDLR metabolism. Pro, prodomain; CAT, catalytic domain; CHRD, cysteine-histidine rich domain.

capacity of PCSK9’s action on LDLR, namely monoclonal antibodies targeting circulating PCSK9, gene silencing targeting both PCSK9 intra- and extra-cellular functions, and mimetic peptides targeting PCSK9 extracellular functions [127, 128]. So far the monoclonal antibodies have become the most promising inhibitor compared to the other two approaches [129]. Some of the monoclonal antibodies have gone into phase 3 clinical trials [130]. The evolocumab from Amgen and alirocumab from Sanofi and Regeneron have both gone under priority review by FDA for its Biologics License Application (BLA) [131, 132]. They are being evaluated in people with high risk of cardiovascular events that cannot be controlled with statin treatment.

Although PCSK9 has become the most promising target for dyslipidemia drug development since statins, there is still a lot to be learned about its mechanism in cholesterol regulation and global effects on other activities.

1.4. Rationale

Despite just over a decade of PCSK9 research in cell, animal models and human studies, the exact mechanism of PCSK9-mediated LDLR degradation and the identification of other proteins involved in PCSK9’s action have not been fully elucidated, nor do we know about the molecular mechanisms of many GOF and LOF PCSK9 variants. Furthermore, PCSK9’s effect on global protein changes and pathways besides cholesterol homeostasis still requires further understanding. Given the expanding biological roles of PCSK9, and as we move toward PCSK9’s commercialization as a dyslipidemia drug target, further research with PCSK9 and its variants is warranted to gain a more detailed understanding of the molecular mechanism and global effects of PCSK9 and its variants.

1.5. Hypothesis and Objectives

1.5.1. Hypothesis

I propose that a global proteomic study of PCSK9 and its variants will help identify more novel proteins and pathways that are affected by them, as well as understand the influence of PCSK9 interactors on these pathways, and may further define the mechanism of PCSK9's action towards LDLR.

1.5.2. Objectives

1) Develop quantitative proteomics-based method to identify changes in subcellular distribution of proteins affected by gain- and loss- of function PCSK9 variants
2) Identify changes in secretomes of cells overexpressing PCSK9 and its variants by quantitative proteomics to compile biological pathways affected by PCSK9 and its variants using bioinformatics tools.
3) Identify secreted interactors of PCSK9 and its variants using co-immunoprecipitation in combination with mass spectrometry analyses.

1.5.3. Experimental Tools

1.5.3.1. PCSK9 Variants for Study

In this thesis, several PCSK9 variants were chosen for study, namely GOF variants S127R and D374Y and LOF variants R46L, L10InsA53V, and Q152H (Table 1).

The variant S127R was one of the first GOF PCSK9 variants identified [22] yet little about its molecular mechanism is known. Although for GOF variants the secretion of PCSK9 is expected to be higher, the S127R on the contrast has reduced autocatalytic cleavage of prodomain by 50-60% and lower secretion in cell culture [36]. The PCSK9 S127R variant also exhibits lower plasma concentrations in humans [133]. The binding affinity of S127R to LDLR does not seem to be the reason for its gain of function, with some studies demonstrating higher binding affinity at neutral pH than wild type [5], while other studies showing similar binding affinity [26].

The most severe GOF variant, in terms of increasing cholesterol levels and causing ADH, D374Y was first screened out in a Utah pedigree in 2004 [134]. Similar to S127R, its prodomain cleavage in the ER is decreased [36], and it lacks the inactivating cleavage event by furin [45]. However, distinguished from S127R, this variant has been found to bind LDLR with 25-fold greater affinity and is internalized more efficiently than wild type PCSK9, accounting for most of the over 10-fold enhanced LDLR degradation [76, 135, 136]. It is used here as an ultimate representation of changes caused by strong gain of function effects.

The LOF variant R46L was identified in Caucasians with 21% reduction of plasma LDLC levels in the Dallas Heart study among populations with the lowest plasma LDLC levels (<5th percentile) [121, 137, 138]. The R46L also has a positive effect on reducing risk of cardiovascular disease for carriers in FH populations [139]. In cell culture, it is secreted at same level as wild type PCSK9 but with significantly reduced phosphorylation in the prodomain [47, 76], which might be explained by the theory that the phosphorylation state of PCSK9 determines in part of its functional state [69]. In humans, the level of R46L is significantly reduced [111]. This study might further illustrate its mechanism of LOF and reveal other effectors.

In the serum study of Caucasian Canadians with LDLC levels in the <25th percentile, L10Ins and A53V were identified as linked variants with significantly lower LDLC without PCSK9 reduction in plasma [140]. Similar to R46L, it is found to have decreased phosphorylation level by 17% in cell culture study [47]. Unlike R46L, the L10InsA53V in human plasma is not reduced, implying different mechanism for these two LOF variants [124].

The French Canadian LOF variant Q152H does not undergo autocatalytic processing of proPCSK9 and is not secreted [141]. People carrying this variant were found to have decreased plasma PCSK9 and LDLC concentrations by 79% and 48% [141]. In cell culture, co-transfection of Q152H and wild type PCSK9 resulted in 78% reduction of wild type PCSK9 secretion [141]. Thus the variant Q152H could represent the condition with very little secreted PCSK9 and can be used as a strong loss of function contrast to PCSK9 WT and other variants.

For global proteomics study I chose these variants because I wanted to (1) contrast strong loss of function (Q152H) with strong gain of function (D374Y), (2) understand the varying mechanisms of loss of function. For example, as LOF variants, human plasma PCSK9 level is reduced in R46L but not in L10insA53V, indicating different mechanisms for the loss of function that may involve the role of interacting proteins after secretion and (3) understand the varying mechanisms of gain of function. For example the variant S127R is a GOF variant despite lower secretion and not great increase in binding affinity with LDLR, suggesting other mechanism for its effect.

Table 1. List of PCSK9 variants studied

	PCSK9 Variants	PCSK9 cellular modification	PCSK9 secretion in cell culture	Cellular LDLR concentration	PCSK9:LDLR binding affinity	Plasma PCSK9 concentration in humans	Plasma LDLC changes in humans
LOF	Q152H (QH)	ProPCSK9 not autocatalytically cleaved to PCSK9 [141]	no secretion; WT PCSK9 secretion by 78% in co-transfection[141]	increased[141]	NR	79% lower[141]	48% reduction[113]
	L10insA53V (AV)	decreased phosphorylation by 17% (A53V) [47]	normal[114]	NR	NR	normal [112]	<25th %ile in Caucasian Canadians [112]
	R46L (RL)	decreased phosphorylation by 34% [47]	normal [47]	NR	NR	15% lower [111]	11% to 16% reduction [98, 99]
GOF	S127R (SR)	~50–60% decreased proPCSK9 processing[36]; reduces cleavage by furin at cell surface [45, 48]	lower[36]	decreased [115]	maybe higher, inconclusive [105, 106]	lower[102]	>95th%ile [109]
	D374Y (DY)	reduces cleavage by furin at cellsurface[45, 48]	lower[111]	decreased[115]	10-25-fold higher [105, 106]	lower [102]	>95th%ile [109]

Abbreviation： Loss of function, LOF; Gain of function, GOF;
Not reported, NR; percentile, %ile.

1.5.3.2. SILAC ‘Spike-in’ Quantification

For the first two objectives, stable isotope labeling by amino acids in cell culture (SILAC) as a ‘Spike-in’ standard [142-145] was utilized to quantify protein abundance changes. The basic workflow of a SILAC ‘Spike-in’ experimental procedure is as depicted in Figure 5 and in Materials and Methods section 2.12.3.

SILAC is an *in vivo* metabolic incorporation of a label into proteins [142-145]. To generate SILAC samples, two or three cell populations are grown in media with different isotope labeled amino acids for > 6 passages till all the proteins incorporate the labeled amino acids [142]. In the classical SILAC approach, all samples are digested together and doublets or triplets of chemically identical peptides with different masses can be differentiated in MS spectra. The ratios between the samples are calculated directly by comparing the differences in the intensities of the peaks [142, 145].

However, in the SILAC ‘Spike-in’ quantification, the ‘heavy’ amino acid (for this study, [13C6, 15N2]-L-lysine (K8) and [13C6, 15N4]-L-Arginine (R10)) labeled sample is used as a ‘spike-in’ standard [145], while the control and experimental samples are grown under normal cell culture conditions [145]. Each non-labeled sample is combined with the SILAC standard and analyzed individually by MS/MS. The protein abundance for the ‘light’ samples are represented as the Light/Heavy (L/H) ratio relative to the ‘Spike-in’ standard [145]. Thus the relative differences in protein abundance between the ‘light’ samples can be compared as ‘ratio of ratios’ by dividing the L/H ratios of two samples resulting in L(sample 1)/ L(sample 2) [145]. This approach has been further utilized as a Super-SILAC Spike-in for human tissue proteome studies wherein several different heavy cell lines are combined to produce a ‘super SILAC’ mixed-reference standard [146].

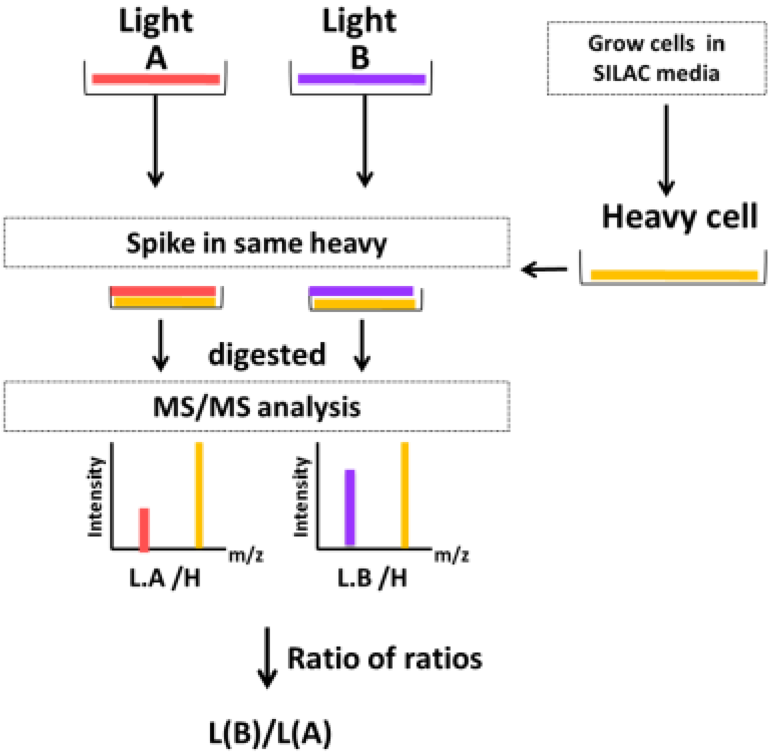

Light
A
Light
B
Grow cells in
SILAC media
Heavy cell
Spike in same heavy
digested
MS/MS analysis
Intensity
m/z
L.A /H
Intensity
m/z
L.B /H
Ratio of ratios
L(B)/L(A)

Figure 5. Schematic of a SILAC ‘Spike-in’ experiment workflow. Cells were grown in SILAC media containing ‘heavy’ isotope labeled amino acids to generate SILAC ‘heavy’ cells, combined with ‘light’ cells and individually analyzed by MS/MS. The protein abundances for ‘light’ samples were quantified as L/H ratios relative to ‘heavy’ standards. Because the ‘heavy’ standards were identical, protein abundance differences between experimental controls and samples were calculated as ‘ratio of ratios’ L (B)/L (A) by dividing their L/H ratios.

2. Materials and Methods

2.1. Cell Culture

Human hepatic HUH7 cells (Japanese Collection of Research Bioresources Cell Bank, Osaka, Japan) were grown at 37°C and 5% CO_2 in Dulbecco's Modified Eagle Medium (DMEM; Gibco, Invitrogen, Burlington, ON, CAN) supplemented with 10% (v/v) qualified fetal bovine serum (FBS; Invitrogen), 1mM sodium pyruvate (Invitrogen), 28µg/mL gentamycin (Invitrogen), and 5µg/ml Plasmocin (InvivoGen, San Diego, California, USA); complete DMEM media. These are referred to as 'light' HUH7 cells. HUH7 SILAC (stable isotope labeled amino acids in cell culture) cells were generated by growth in SILAC 'heavy' media at 37°C and 5% CO_2. SILAC 'heavy' media is customized DMEM (AthenaES, Baltimore, MD, USA) lacking lysine, arginine and methionine. It is supplemented with Methionine (30 mg/L) and isotopically labeled amino acids [$^{13}C_6$, $^{15}N_2$]-L-lysine (Lys-8, 146 mg/L) and [$^{13}C_6$, $^{15}N_4$]-L-Arginine (Arg-10, 42 mg/L) purchased from Sigma-Aldrich (Oakville, ON, CAN). It is further supplemented with 10% (v/v) dialyzed FBS (Invitrogen), 1mM sodium pyruvate, 28µg/mL gentamycin and 5 µg/ml Plasmocin; complete SILAC media. Cells were grown for at least 10 doubling times to allow for complete incorporation of the isotope-labeled amino acids. Incorporation of heavy HUH7 cells was over 96% for both lysine and arginine incorporation. These are referred to as 'heavy' or SILAC HUH7 cells and are used as a 'spiked-in' reference for MS quantification as described in section 2.12.3.

2.2. Transfection

Transient transfections were performed using HUH7 cells with a bicistronic plasmid pIRES2-EGFP vector encoding C-terminal V5-tagged PCSK9 wild type (obtained from Dr.

Nabil G. Seidah, IRCM, Montreal) and C-terminal V5-tagged PCSK9 variants Q152H, L10InsA53V, R46L, S127R and D374Y. The plasmid encodes for EGFP (Enhanced Green Fluorescent Protein) that is co-expressed with the protein of interest PCSK9-V5. Thus the transfection efficiency can be visualized and estimated as the number of cells expressing EGFP/ the total number of cells. The PCSK9 variants were generated from the pIRES2 plasmid encoding wild type PCSK9-V5 using QuikChange II XL Site-Directed Mutagenesis Kit (Agilent, Santa Clara, CA, US) by Dr. Janice Mayne and sequenced for verification through BioBasic's sequencing services (Markham, ON, CAN). Mock transfection and empty vector transfection were used as controls. Cells were seeded in 6-well plates at 1 million cells/well in complete DMEM media. The following day cells at ~80% confluent were washed 1× with phosphate-buffered saline (PBS; Invitrogen) and media replaced with 500μl antibiotic-free media DMEM + 10% (v/v) qualified FBS and 1mM sodium pyruvate. Cells were transfected as per Lipofectamine 3000 manufacturer's protocol (Invitrogen). Briefly 2μg of plasmid DNA, 4μl P3000 reagent and 8 μl Lipofectamine 3000 were used per well with a ratio of DNA to P3000 to Lipofectamine 3000 at 1:2:4 (w/v/v). Lipofectamine 3000 was first diluted in Opti-MEM medium (Invitrogen) to final volume of 250 μl and vortexed 3 seconds. Plasmid DNA was diluted in Opti-MEM Medium followed by addition of P3000 to final volume of 250 μl and vortexed 3 seconds. Diluted plasmid DNA/P3000 was added to diluted Lipofectamine 3000 at 1:1 ratio with final volume of 500 μl, vortexed 3 seconds and incubated at room temperature for 5 minutes. The plasmid DNA-lipid complex was then added onto cells. 3 hours following transfection, transfection media was removed. The cells were washed with Opti-MEM media twice, and maintained at 37°C and 5% CO_2 for 48 hours in 2ml Opti-MEM medium supplemented with 1mM sodium pyruvate, 28μg/mL gentamycin, and 5 μg/ml Plasmocin. Transfection efficiency was determined as ~80% using a Zeiss AxioObserver.Z1 fluorescence microscope (Carl Zeiss Iberia, S.L., Spain). Cells and media were collected as per section 2.4.

For protein subcellular distribution changes study, cells stably transfected with wild type PCSK9-V5, variants D374Y-V5 and Q152H-V5 and empty vector were generated by Dr. Alexandre Noah. Cells were seeded in 6-well plates and transfected as above except that 24 hours following transfection, the cells were trypsinized and transferred into 10-well plates and changed to complete DMEM media with 1000 μg/ml geneticin (Invitrogen) and cultured at 37°C and 5% CO_2 for selection. The plasmid pIRES2 encodes geneticin (G418) resistance for selection. The cells were changed to fresh complete DMEM media with 1000 μg/ml geneticin every day. A mock transfection served as the control for monitoring cell death under geneticin treatment. Following geneticin selection of the pooled stable PCSK9 and variant expressers (~1 month), cells were maintained in 500 μg/ml geneticin. Cells and media were collected as per section 2.3. PCSK9 expression was monitored by immunoblotting and ELISA as per sections 2.5 and 2.6, respectively.

As a ‘heavy’ spike-in reference for subsequent mass spectrometry (MS) quantification for the protein subcellular distribution study, stably transfected cells were cultured in SILAC complete media with 500 μg/ml geneticin for 10 passages to generate ‘heavy’ HUH7 cells stably expressing PCSK9 wild type, Q152H, D374Y and empty vector and then maintained in that media. Incorporation of ‘heavy’ stably transfected HUH7 cells was over 96% for both arginine and lysine. Cells were collected and the ‘heavy’ microsomal pellet prepared as reference material as per section 2.7.

2.3. Cell and Secretome Sample Harvest

To generate my secretome reference material, heavy HUH7 cells were plated in 6-well plates at the same number as light HUH7 cells (1million cells/well) in complete SILAC media. Following overnight incubation, the cells were washed with PBS three times and changed to serum-free SILAC media and cultured for 48 hours.

For secretome and secreted interactome studies, the transiently transfected HUH7 cells (section 2.2) and heavy HUH7 cells (my reference material) were collected in ice-cold 1× RIPA buffer (50 mM Tris pH 7.6, 150 mM NaCl, 1% (v/v) NP40, 0.5% (w/v) Na deoxycholate, 0.1% (w/v) SDS) containing 1× complete mini protease inhibitor cocktail tablets (Roche, Indianapolis, IN, USA), vortexed, incubated at 4^{o}C with rotation for 10 min and then spun at 17000×g for 10 min to remove cellular debris. Supernatants were collected as total cellular lysates (TLC) and stored at -20 °C. Secretome was collected from 48 hours spent media, spun at 17,000×g for 20min to remove cellular debris, added 1× complete mini protease inhibitor cocktail tablets and stored at –20°C.

2.4. SDS-PAGE Fractionation and Staining

For secretomes, 100 µl samples were concentrated in a refrigerated, centrifugal vacuum evaporator (ThermoFisher Scientific). Samples were reconstituted in 30µl of 2×NuPAGE sample running buffer (Invitrogen.) For total cell lysates (TCL), protein concentrations were determined by Bio-Rad protein assay (Bio-Rad, Mississauga, ON, CAN) following standard protocols and 25 µg of protein samples were reconstituted in an equal volume of 2×NuPAGE sample running buffer. All samples were heated at 70°C for 10 minutes and electrophoresed through a 4-12% NuPAGE Bis-Tris gel (Invitrogen) or a 7% NuPAGE Tris Acetate gel (Invitrogen) at 150V as per standard protocols. A SeeBlue Plus2 pre-stained protein standard

(Invitrogen) was also loaded onto gels to estimate molecular weight. Gels were stained using a Colloidal Blue Staining kit (Invitrogen) and destained according to manufacturer's protocol or used for immunoblotting as described below.

2.5. Immunoblotting and Analyses

Following gel electrophoresis, proteins were transferred onto nitrocellulose membranes (pore size 0.45 um, Bio-Rad) using Invitrogen's semi-wet transfer apparatus for 1.5hour at 45V. The transfer buffer consisted of 1.25 mM Bicine, 1.25 mM Bis-tris, 0.05 mM EDTA and 20% (v/v) methanol. Protein transfer was confirmed using Ponceau staining (3 min room temperature) and ddH_2O destaining (3x rinses). Membranes were blocked for 1 hour at room temperature in 5% (w/v) skim milk (United States Biological, Salem, MA, US) in Tris Buffered Saline (25mM Tris, 2.7mM KCl, 137mM NaCl, pH 7.6) with 0.1% (v/v) Tween 20 (TBS-T). Blots were probed with primary antibodies as listed in Table 2 diluted in 5% skim milk in 1×TBS-T for 1 hour at room temperature (except anti-LDLR blots were incubated at 4°C overnight). Blots were then washed three times for five minutes each with 1×TBS-T buffer then incubated with secondary antibodies (Table 2) at dilutions of 1:5000 with 5% skim milk in 1×TBS-T for 1 hour at room temperature. Blots were washed with 1×TBS-T three times for five minutes each. Immunoblots were revealed by chemiluminescence with Western Lightning Plus (Perkin Elmer) on Progene film (Ultident, St. Laurent, QC, CAN). Densitometry was performed with ImageJ and signals from TCL were normalized to the densitometry value for the corresponding actin. Values shown are relative to the normalized density of signals for control cells.

Table 2. Antibody list

Primary Antibodies	**Species**	**Source**	**Product #**	**Dilution**
Anti-V5-HRP	Mouse	Invitrogen	R961-25	1:5000
Anti-V5	Mouse	Invitrogen	46-0705	1:5000
Anti-Human PCSK9	Rabbit	CircuLex	CY-P1037	1:5000
Anti-Human LDLR	Goat	R&D Systems	AF2148	1:500
Anti-FASN	Rabbit	Bethyl	A301-324A	1:1000
Anti-PSMD2	Rabbit	Abcam	ab140675	1:1000
Anti-Actin	Mouse	Cedarlane	CLT9001	1:5000
Anti-EEA1	Rabbit	Abcam	ab109110	1:500
021Anti-Calnexin	Rabbit	Sigma	C4731	1:1000
Secondary Antibodies	**Species**	**Source**	**Product #**	**Dilution**
ECL Anti-mouse IgG-HRP	sheep	GE Healthcare UK Limited	NA931V	1:5000
ECL Anti-rabbit IgG-HRP	donkey	GE Healthcare UK Limited	NA934V	1:5000
Anti-goat IgG-HRP	donkey	Santa Cruz Biotechnology	D2514	1:5000

2.6. ELISA Analysis

The PCSK9 concentration from secretomes was quantified using a human PCSK9 ELISA (Enzyme Linked Immunosorbent Assay) kit from CircuLex (Japan). The samples were diluted 1/200 in provided dilution buffer and measured as per the standard protocol. Absorbance was measured at 450/550 nm with an Infinite F200 Pro Reader (Tecan, San Jose, CA, USA). This assay has an intra-assay CV of 1.5-2.6% and an inter-assay CV of 2.9-7.1%.

2.7. Subcellular Fractionation by Density Gradient Centrifugation

‘Light’ HUH7 cells stably transfected with plasmids encoding for D374Y-V5, Q152H-V5, wild type PCSK9-V5 and empty vector as described in section 2.2 were used for the subcellular fractionation study. Cells were seeded in 150×25mm dishes at 5 million cells/dish and cultured in complete DMEM media with 500 μg/ml geneticin for 48 hours. Four dishes each of D374Y-V5, Q152H-V5, wild type PCSK9-V5 and empty vector stable expressing cells were used for each subcellular fractionation. As well, 'heavy' HUH7 cells stably transfected with PCSK9 Q152H-V5 and D374Y-V5 were seeded in 150×25mm dishes at 5 million cells/dish and cultured in complete SILAC media with 500 μg/ml geneticin for 48 hours. Four dishes each of these 'heavy' HUH7 cells were used for each subcellular fractionation. For subcellular fraction cells were washed with ice-cold PBS twice and harvested with 8ml ice-cold homogenization buffer (250 mM sucrose, 20 mM Tris-HCl, 1 mM EDTA, 0.02% NaN_3, pH 7.4) containing 1× Complete Mini Protease Inhibitor Cocktail, and transferred into a Dounce tissue grinder (Wheaton, USA). Cells were homogenized 40 times on ice and centrifuged at 700×g, 4°C for 10 min to pellet nuclei and cell debris. The supernatant was collected and spun at 700×g, 4°C for 10 min a second time to pellet any remaining nuclei and cell debris. Supernatant was centrifuged at 17000×g, 4°C for 20 min to pellet mitochondria and plasma membrane. Supernatant was then ultracentrifuged at 100,000×g, 4°C for 1hour in a SW41 Ti swinging bucket ultracentrifuge rotor (Beckman) to pellet microsomes. 'Heavy' microsomes were dissolved in 150 μl 4% (w/v) SDS and incubated at 70°C for 10 min to solubilize proteins and stored at -20°C. The 'light' microsomal pellet was resuspended in 100 μl homogenization buffer and layered on top of a density gradient solution. A pre-test of four established density gradient solutions for subcellular fractionation by ultracentrifugation was conducted to select the most optimal for this study. Compositions of the four density gradients are: (1) 20% and (2) 30% (w/v) Nycodenz freezing-thawing gradient solutions prepared as per reference [147], (3) Nycodenz step gradient solution prepared as per reference [148] and (4) sucrose step gradient solution prepared as per reference [149]. To generate the 20% and 30% Nycodenz freezing-thawing gradient solutions, 10ml of the solutions containing 20% or 30% (w/v) Nycodenz in 20mM Tris-HCl (pH 7.4), 1mM EDTA and 50mM sucrose as an osmotic balancer was frozen in Beckman SW41 centrifuge tubes (Ultra-Clear, thinwall, 14 × 89 mm, 13.2 mL) at -20°C overnight. They were then thawed at room temperature for a few hours before use. The Nycodenz step gradient was generated in Beckman SW41 centrifuge tubes by layering 2.5 ml of 24%, 19.33%, 14.66% and 10% of Nycodenz in 20 mM Tris-HCl (pH 7.4), 1 mM EDTA and 0.02% NaN3 (bottom to top, respectively). The tube was placed horizontally for 45 min at room temperature to form a linear gradient. The non-linear sucrose step gradient was generated with 2 ml of 65%, 40.6%, 35%, 25% and 8% (bottom to top, respectively) of sucrose solution (20 mM Tris-HCl (pH 7.4), 1 mM EDTA and 0.02% NaN3). The microsomal pellets were resuspended in 100 µl homogenization buffer, loaded on top of the gradients and then centrifuged at 100,000×g, 4°C overnight in the SW41 Ti swinging bucket ultracentrifuge rotor. The centrifuged solution was then fractionated from the top of the gradient into 15× 0.75 ml fractions. Each fraction was diluted with an equal volume of 4% (w/v) SDS and incubated at 70°C for 10 min to break membrane structure of microsomes and solubilize proteins. Fractions were stored at -20°C. Equal volumes of fractionated samples were analyzed by immunoblotting as described above and quantitative mass spectrometry as described in 2.9.

2.8. Co-Immunoprecipitation of Transfected PCSK9 and Its Variants

Secretomes collected as described above from transiently transfected mock, empty vector, PCSK9 WT-V5 and its variants were subjected to co-immunoprecipitation using Anti-V5 Agarose Affinity Beads (Sigma-Aldrich). 50µl of anti-V5 agarose affinity beads were added to 1.75 ml secretomes and incubated at 4°C overnight with rotation. Samples were spun at 1500×g for 5 minutes and supernatants removed. Immunoprecipitates were washed 3 times by adding 1ml fresh Opti-MEM Medium and incubating at 4°C for 10 minutes with rotation followed by centrifugation at 1500×g for 5 minutes. Supernatants were discarded. 50 µl of 2× NuPAGE gel loading buffer was added to immunoprecipitates and heated at 70°C for 10 minutes with agitation to keep beads in suspension. Following elution, samples were spun at 15000×g for 2 min and supernatants loaded onto gels or stored at -20°C for further MS/MS and immunoblotting analyses.

2.9. In-Gel Digestion of Protein

Proteins were electrophoresed through a 7% NuPAGE Tris Acetate gel and stained using Colloidal Blue and destained as described in section 2.4. After rinsing the gel with ddH_2O, the gels were cut into 25 slices per lane. Each slice was further cut into cubes of ~1×1 mm size. The gel pieces were transferred into 96-well PCR plates (VWR International, PA, USA) and spun at 100×g for 20 seconds in a M-20 rotor (Thermo) using ST 16R centrifuge (Thermo). Gel pieces were dehydrated by adding 300 µl 100% Acetonitrile (ACN, J.T Baker, PA, U.S.A.) and incubating for 10 minutes at room temperature. Acetonitrile was removed and the gel pieces allowed to air dry. The plate was covered with a kimwipe (KIMTECH Science, GA, USA) to prevent keratin contamination. Protein disulfide bridges were reduced by adding 50 µl 20mM dithiothritol (DTT, Sigma) in 50mM ammonium bicarbonate (ABC, pH 8.0; MultiPharm) and incubating at 37°C for 20min followed by ACN dehydration of the gel pieces as described above. Thiols were alkylated by adding 50µl 20mM iodoacetimide (IAA; Sigma) in 50mM ABC, pH 8.0 and incubating at room temperature in darkness for 40 min followed by ACN dehydration of gel pieces as described above. The proteins were digested by adding 30µl of 20ng/µl trypsin (Invitrogen) in 50mM ABC, pH 8.0 to the gel pieces and incubating at 37°C overnight with agitation. The tryptic-peptides in the overnight supernatant were collected by centrifugation. Twice 150 µl of 80% ACN and 0.5% formic acid (FA; Sigma) were added to the gel pieces and centrifuged to further extract the tryptic-peptides. Supernatants containing the tryptic-peptides were combined and dehydrated in the refrigerated vapor trap (ThermoFisher Scientific). Samples were stored at -20°C. Immediately prior to loading onto mass spectrometer samples were suspended in 20 µl 0.5% FA. Mass spectrometry measurement was carried out as described in section 2.12.2.

2.10. Filter-aided sample preparation (FASP)-trypsin Digestion of Protein

The filter-aided sample preparation (FASP) method was used for in-solution tryptic digestion of samples from secretome and subcellular protein distribution studies. For secretome studies, 500µL each of light and heavy secretomes collected as described in section 2.3 were combined. For the protein subcellular distribution studies, the 'heavy' microsomes were pooled and an equal amount (20 µl) of the pooled 'heavy' microsomes were spiked in every 500 µl of 'light' fraction as collected in section 2.7. Amicon Ultra-0.5 mL centrifugal filters (Millipore; MW cutoff 3000 Da) were pre-rinsed with ddH_2O and centrifuged at 16000 ×g for 20min at room temperature. To the samples above was added 400 l 8M urea in 50mM Tris-HCl (pH 8.0), and transferred into the filter and centrifuged at 16000 ×g for 20min at room temperature. They were resuspended in 200 µl 8M urea in 50mM Tris-HCl (pH 8.0) and concentrated by centrifugation for three times. Protein disulfide bridges were reduced by adding 200 µl 8M urea in 50mM Tris-HCl (pH 8.0) and 4 µl 1M DTT (final concentration 20mM), and incubating in Multitron shaker at 37°C, 250 rpm for 30 min and centrifugation. Thiols were alkylated by adding 200 µl 8M urea in 50mM Tris-HCl (pH 8.0) and 4 µl 1M

IAA (final concentration 20mM), and incubating in Multitron shaker at room temperature, 250 rpm for 30 min, covered by aluminum foil. Then the samples were washed three times with 8M urea in 50mM Tris-HCl (pH 8.0). The samples on the filter were washed 3 times with 200µl of 50 mM ABC (pH 8.0) with centrifugation as above to remove urea that interferes with subsequent trypsin digestion. Finally the filter was transferred to a new tube and added 1µg trypsin (Invitrogen) in 200µl 50mM ABC (pH 8.0), and digested overnight at 37°C with agitation of 250 rpm. The resulting peptides were collected by centrifugation and combined with peptides collected from a second centrifugation with 200µl 1% (v/v) formic acid (FA). The peptides were stored at -20°C for SCX fractionation and desalting as described below.

2.11. SCX Fractionation and Desalting of Peptides

For secretome studies, following the FASP-trypsin digestions, Strong Cationic Exchange (SCX) fractionation was performed on the tryptically-generated peptides to reduce the complexity of sample prior to MS loading. First, columns with SCX beads (1000Å, Agilent) were pre-washed with 100 µl 0.5% FA and centrifuged at 200 ×g for 1min for three times. Acidified peptides in FA (pH<2.0) were added on SCX beads (Agilent) and fractionated into 5 fractions by eluting with 100 µl each of Britton–Robinson buffer containing 20mM boric acid, 20mM phosphoric acid and 20mM acetic acid at pH 4, 6, 8, 10 and 12 by centrifugation at 200 ×g for 1min.

Desalting: Prior to loading onto the mass spectrometer all samples were desalted using Reprosil-Pur C18-AQ (10µm, Dr. Maisch GmbH, Ammerbuch, Germany). Columns filled with C18 beads were washed three times with 200µl 100% ACN and centrifuged at 100 ×g for 1min, equilibrated twice with 200µl 0.1% FA by centrifuged at 100×g for 1min. The trypically-generated peptide samples and fractionated by SCX above were acidified to reach pH 2~3 with 10% TFA (trifluoroacetic acid) and loaded on the C18 beads. Three washes were carried out with 200µl 0.1% FA and then peptides were eluted with 200µl of 80% ACN and 0.1% FA. The eluent was dehydrated in the refrigerated centrifugal vacuum evaporator and suspended in 25 µl 0.5% FA prior to loading on the mass spectrometer.

2.12. LC-MS/MS Measurement and Quantification

2.12.1. LC-MS/MS Measurement by LTQ-Orbitrap XL for Secretome and Protein Subcellular Distribution Studies

The LC-MS/MS used for secretome and protein subcellular distribution studies consisted of an Agilent 1100 micro-HPLC system (Agilent Technologies, Santa Clara, CA, USA) and an LTQ -Orbitrap XL mass spectrometer (ThermoFisher Scientific, San Jose, CA). The mobile phases consisted of 0.1% (v/v) fomic acid (FA) in water as buffer A and 0.1% (v/v) FA in acetonitrile as buffer B. Peptide separation was performed on a 75µm × 100 mm analytical column packed in-house with reverse phase Magic C18AQ resins (1.9µm; 120-Å pore size; Dr. Maisch GmbH, Ammerbuch, Germany). Briefly, the sample was loaded on the column using 98% buffer A at a flow rate of 1.5µL/min for 15min. Then, a gradient from 5% to 45% of buffer B was performed in 120 min at a flow rate of ~300nL/min. The MS/MS method consisted of one full MS scan from 300 to 1700 m/z followed by data-dependent MS/MS scan of the 5 most intense ions, with dynamic exclusion repeat count of 2, and repeat duration of 90 s. The full MS was performed in the Orbitrap analyzer with R = 60,000 defined at m/z 400, while the MS/MS analysis were performed in the LTQ. To improve the mass accuracy, all the measurements in Orbitrap mass analyzer were performed with internal recalibration (“Lock Mass”). On the Orbitrap, the charge state rejection function was enabled, with single and “unassigned” charge states rejected. All data were recorded with Xcalibur software (ThermoFisher Scientific).

2.12.2. LC-MS/MS Measurement by Q-Exactive-Obitrap for Secreted Interactome Study

The LC-MS/MS used for secreted interactome study consisted of an Eksigent nanoLC Ultra system (Eksigent, Sigapore) and a Q-Exactive Hybrid Quadrupole-Orbitrap mass spectrometer (ThermoFisher Scientific). The mobile phases consisted of 0.1% (v/v) FA in water as buffer A and 0.1% (v/v) FA in acetonitrile as buffer B. Peptide separation was performed on a 75µm × 100 mm analytical column packed in-house with reverse phase Magic C18AQ resins (1.9µm; 120-Å pore size; Dr. Maisch GmbH, Ammerbuch). Briefly, the sample was loaded on the column using 96% buffer A at a flow rate of 1.5µL/min for 15min. Then, a gradient from 4% to 30% buffer B was performed in 60 min at a flow rate of ~300nL/min obtained from splitting a 20 µL/min through a restrictor. The MS/MS method consisted of one full MS scan from 300 to 1800 m/z followed by data-dependent MS/MS scan of the 12 most intense ions, with dynamic exclusion repeat count of 2, and repeat duration of 90 s. The full MS was performed in the Orbitrap analyzer with R = 70,000 defined at m/z 400, and the MS/MS analyses were also performed in the Orbitrap. To improve the mass accuracy, all the measurements in Orbitrap mass analyzer were performed with Lock Mass. On the Orbitrap, the charge state rejection function was enabled, with single and “unassigned” charge states rejected. All data were recorded with Xcalibur software.

2.12.3. SILAC Spiked-in and Label Free Quantitative Analyses by MaxQuant

All the raw files were processed and analyzed by MaxQuant Version 1.2.2.5 against Uniprot protein fasta database (2012, July version), including commonly observed contaminants. The following parameters were used: cysteine carbamidomethylation was selected as a fixed modification; the methionine oxidation, protein N-terminal acetylation was selected as variable modification. Enzyme specificity was set to trypsin. Up to two missing cleavages of trypsin were allowed. The precursor ion mass tolerances were 6 ppm, and fragment ion mass tolerance for MS/MS spectra was 0.8 Da for XL and 20 ppm for Q-Exactive. For protein identification, if the identified peptide sequences of one protein was equal to or contained by another protein's peptide sequences, these two proteins were grouped together by MaxQuant and reported as one protein group. The false discovery rate (FDR) for peptide and protein was set at 1% and a minimum length of six amino acids was used for peptides identification. SILAC double labeling (light: K0R0; heavy: K8R10) was set for search and quantitation for secretome and protein subcellular distribution studies. The normalized H/L ratio between heavy and light was used for quantification for secretome study and the raw H/L ratio was used for protein subcellular distribution study. The label free quantified (LFQ) intensity was used for quantification of secreted interactome study.

2.13. Statistical Analysis and Bioinformatics Processes

All data for ELISA and densitometry are presented as means ± standard error of the mean (SEM). Significant differences were assessed by ANOVA followed Tukey's *post hoc* comparisons using GraphPad Prism 6.0 Software. P-values <0.05 were considered statistically significant.

The proteingroup files were imported into Perseus (Provision Enterprise Resources & Clusters Enabling Uniform Systems Cluster, version 1.3.0.4) for statistical analysis of the data. All the identifications from contaminants, only identified by site and reversed databases were removed. For secretome study, SILAC ratios were inverted from H/L to L/H, and then logarithmized using the base of 2 ($\log_2$ L/H). The dataset was filtered based on numbers of quantified values, that is, only proteins with quantification values in over half of the MS/MS measurements (12 out of 24) were used for further analysis. For secreted interactome study, the LFQ intensity was logarithmized using the base of 10 ($\log_{10}$ LFQ intensity). The dataset was group-based filtered meaning only proteins with quantification values in at least one of the three replicates were used for further analysis. Two-way ANOVA was used to analyze protein level difference, with p-values <0.05 indicating statistical significance. For the hierarchical clustering analysis, median value of $\log_2$ L/H (secretome study) and $\log_{10}$ LFQ intensity (secreted interactome study) of each protein was processed following z-score normalization within Euclidean distances. Protein subcellular localization and pathway analyses were carried out using Ingenuity Pathway Analysis (IPA).

2.14. SILAC incorporation Calculation

To assess SILAC incorporation, a confluent flask of 'heavy' cells were collected, FASP-trypsin digested, desalted, dehydrated and measured on LTQ-Orbitrap XL as described above. Following MaxQuant database searching and removal of contamination from the peptide file, all "0"s in the column of "Intensity light" were replaced by "'1'. Calculation of 1-1/(H/L) using the values in columns 'Intensity H" and 'Intensity L' were performed and displayed in a new column. In the 'sequence' column, peptides not ending with R (Arginine) and K (Lysine) were filtered out. The numbers of peptides that end with R and K were counted as Total R and Total K. The numbers of peptides that end with R and K with the calculation results of 95% or more were counted as Labeled R and Labeled K. The incorporation percentages were then calculated as the ratios of Labeled R/Total R and Labeled K/Total K.

3. Results

3.1. Protein Subcellular Distribution Analysis on a Global Proteomic Scale

As previously mentioned, PCSK9 undergoes prodomain processing and glycosylation in the ER, is further matured in the Golgi and is secreted into circulation [36, 45, 46]. Secreted PCSK9 binds cell surface LDLR and the complex is internalized into cells through endosomes and degraded in lysosomes [76-78]. Thus, PCSK9 travels to several subcellular compartments during its biosynthesis and maturation, as well as during its 'escort' functions. It has the opportunity to interact with other proteins, affect their expression levels and subcellular localization, along these pathways. For example, in a mouse model, overexpressed PCSK9 was found to interact with ApoB100 and decrease its mobilization toward autophagosomes and degradation via the autophagosome/lysosome pathway [91]. This caused increased secretion levels of ApoB100 [91]. Herein, I developed the methodology and conducted a pilot comparison study of changes in protein subcellular distribution on a global scale upon expression of a strong LOF PCSK9 variant, namely Q152H and a strong GOF PCSK9 variant, namely D374Y to identify PCSK9's effect on other proteins during its trafficking.

3.1.1. Density Gradient Optimization for Subcellular Fractionation

A pre-test of four density gradient solutions for subcellular fractionation by ultracentrifugation [147-149] was first conducted using HUH7 total cell lysates to select the optimal gradient solution for the study. This assessment was based on the relative enrichment and distribution of subcellular compartment marker proteins in density gradient fractions visualized by immunoblotting using antibodies such as anti-EEA1 (early endosome marker) and anti-calnexin (ER marker). Figure 6 illustrates representative immunoblots of anti-EEA1 and anti-Calnexin for each tested density gradient solution. The 20% Nicodenz freeze-thaw generated density gradient failed to separate endosomes from ER, as evidenced by the overlap of immuno-signal for EEA1 in later fractions with calnexin (Fig. 6A). The 30% Nicodenz freeze-thaw generated density gradient resulted in misdistribution of early endosome expected to be in earlier (lighter) fractions than the ER and overall minimal enrichment of the immune-signal for EEA1 in any fractions [150] (Fig 6B). Likewise, Figure 6C shows that the Nycodenz step gradient provided minimial enrichment of the early endosomes (anti-EEA1) in the intermediate fractions from the ER (anti-calnexin) contents found in the later fractions. Only the sucrose step gradient resulted in reproducible fractionation of subcellular compartments; the early endosomes (anti-EEA1) were enriched in intermediate fractions (5-9) and the ER (anti-calnexin) in the later fractions from 10-12 (Fig. 6D). This pattern is expected since endosomes are less dense than ER [150]. To further access the utility of this gradient, three receptors that partition between the ER and endosome were visualized by immunoblotting including LDLR (anti-LDLR), transferrin receptor (anti-transferrin receptor; both of which are largely cell surface and endosome localized [9, 151]) and integrin-β1 receptor (anti-integrin-β1 receptor; detectable as an immature ER form and a larger, mature cell surface and endosomal form; Fig. 6D). LDLR and transferrin receptor were enriched in the endosomal fractions (fractions 7-9 and 6-9, respectively) as was the mature form of integrin-β1 receptor (fractions 6-9). In contrast the precursor, immature form of integrin-β1 receptor was enriched in ER containing fractions 11-13.

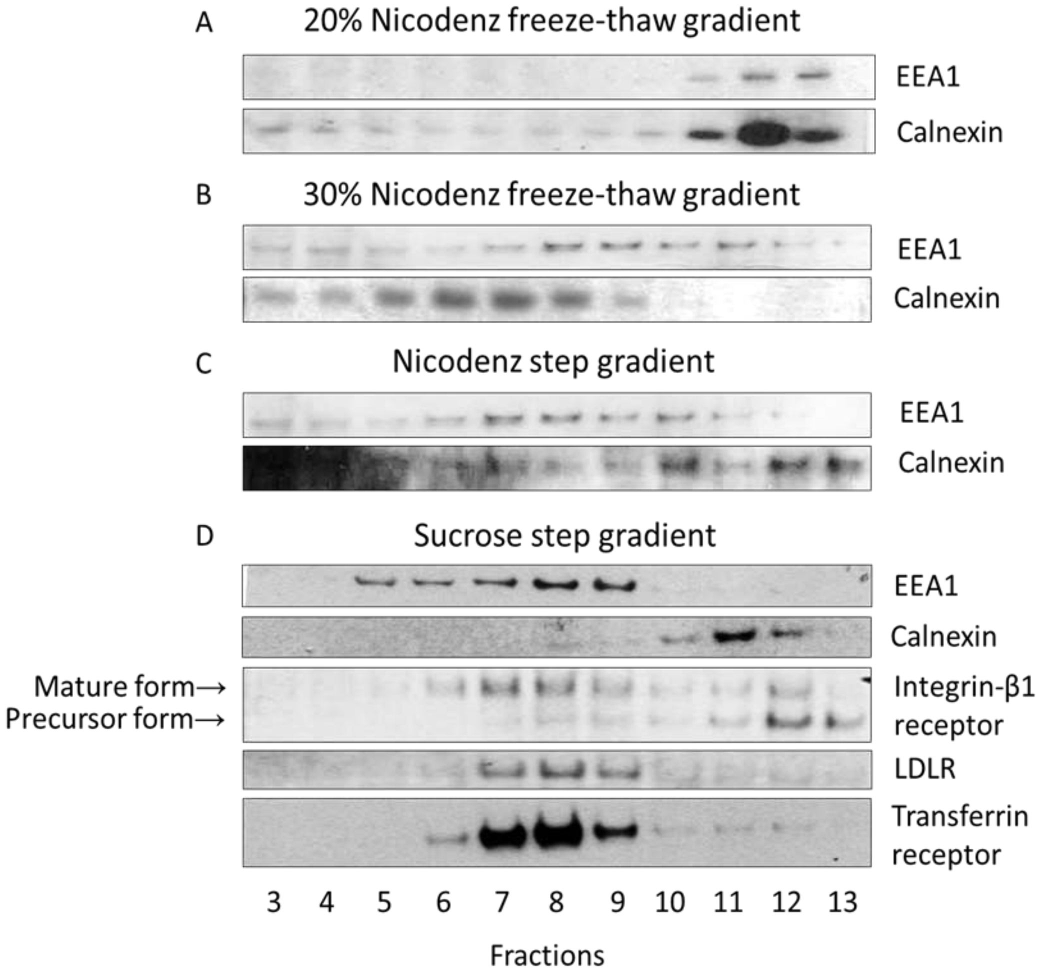

A
20% Nicodenz freeze-thaw gradient
EEA1
Calnexin
B
30% Nicodenz freeze-thaw gradient
EEA1
Calnexin
C
Nicodenz step gradient
EEA1
Calnexin
D
Sucrose step gradient
EEA1
Calnexin
Mature form→
Precursor form→
Integrin-β1 receptor
LDLR
Transferrin receptor
3 4 5 6 7 8 9 10 11 12 13
Fractions

Figure 6. Density gradient selection for optimal subcellular fractionation. Representative immunoblots of subcellular compartment protein marker antibodies anti-EEA1 (early endosome marker) and anti-calnexin (ER marker) reveals the subcellular fractionation results using (A) 20% Nicodenz freezing-thawing gradient solution, (B) 30% Nicodenz freezing-thawing gradient solution, (C) Nicodenz step gradient solution, and (D) sucrose step gradient solution which was further analyzed using antibodies against LDLR, integrin-β1 receptor and transferrin receptors.

3.1.2. Method Development for Studying Global Protein Subcellular Distribution Using Subcellular Fractionation in Combination with MS/MS SILAC ‘Spike-in’ Analyses

Following density gradient solution determination for subcellular fractionation and enrichment, I designed a SILAC ‘spike-in’ strategy and MS/MS workflow that in combination would allow for the identification, quantification and comparison of global subcellular protein distributions between experimental situations.

The workflow is shown in Figure 7. In this study I compared the subcellular distributions of proteins from HUH7 cells stably transfected with the LOF PCSK9 Q152H and GOF PCSK9 D374Y variants. Briefly ‘heavy’ and ‘light’ microsomes were prepared as per materials and methods, and the ‘light’ microsomes were fractionated independently using a sucrose step density gradient and ultracentrifugation as described above. The ‘heavy’ microsomal pellets were combined for use as the SILAC ‘spike-in’ reference material as outlined below. The subcellular fractionation procedure produced 14 fractions in total for each sample (in this case Q152H and D374Y PCSK9 expressing cells). It is important to note that the protein composition as well as abundance in each fraction will differ from each other [147]. The organelle enriched fractions would contain more abundant proteins and the protein composition of lysosome-enriched versus endosome-enriched versus ER-enriched would also be expectedly different. To compensate appropriately for different sample amounts both in terms of amount and composition, an identical ‘spike-in’ of pooled SILAC ‘heavy’ microsomes were added to every ‘light’ fraction as a quantification reference, and then each fraction was processed using FASP-trypsin digestion followed by desalting and identification by LC-MS/MS as described in materials and methods (Fig. 7).

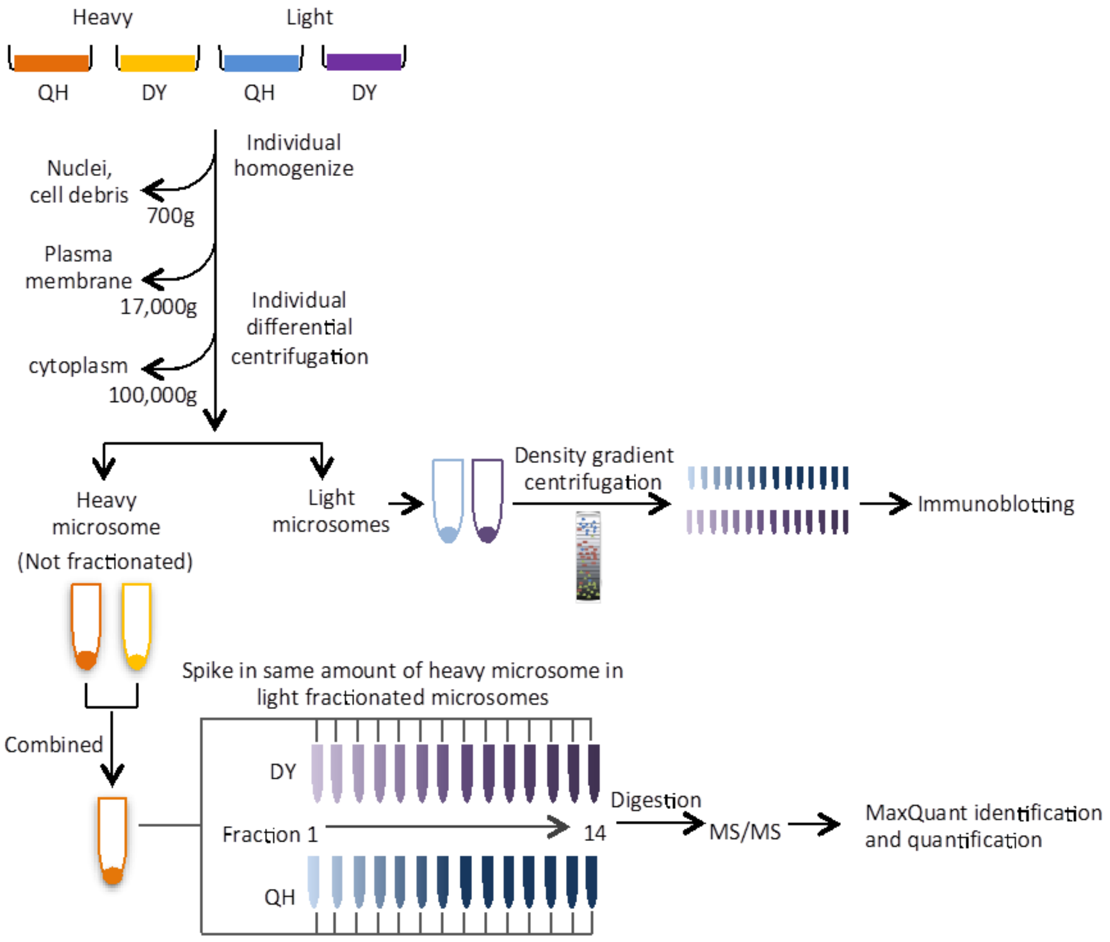

Heavy
Light
QH
DY
QH
DY
Individual homogenize
Nuclei, cell debris
700g
Plasma membrane
17,000g
Individual differential centrifugation
cytoplasm
100,000g
Heavy microsome
(Not fractionated)
Light microsomes
Density gradient centrifugation
Immunoblotting
Spike in same amount of heavy microsome in light fractionated microsomes
Combined
DY
Fraction 1
14
QH
Digestion
MS/MS
MaxQuant identification and quantification

Figure 7. Subcellular protein distribution study workflow. SILAC ‘heavy’ labeled and ‘light’ HUH7 cells stably transfected with PCSK9 variants Q152H and D374Y were used to extract microsomes. ‘Light’ microsomes were fractionated by density gradient centrifugation. The ‘heavy’ microsomes were pooled and ‘spiked-in’ each of the ‘light’ fractions. The ‘spiked-in’ samples were digested and measured by MS/MS. Proteins were identified and quantified by MaxQuant.

Figure 8A shows the fractionation results of the ‘light’ microsomal pellets from HUH7 cells stably expressing PCSK9 Q152H and PCSK9 D374Y and relative enrichment by immunoblotting for EEA1, calnexin, and MAN2A1 (Golgi marker): EEA1 was most abundant in fractions 6-8 and 7-8, calnexin was enriched in fractions 9-12 and 10-12 and MAN2A1 was most enriched in fractions 7-8 and 8-10 for the Q152H and D274Y microsomal fractionations, respectively. There were some overlaps of subcellular markers as expected and as described previously by others[152].

MaxQuant analyses identified in total 23112 peptides representing 2240 proteins from the mass spectral output of the Q152H and D374Y analyses and the SILAC ‘spike in’ enabled quantification of 14666 of these peptides representing 1501 proteins (Appendix 1). Quantitative data are presented visually in Figure 8B using a heat map of the protein distribution in the fractions of Q152H (left panel) and D374Y (right panel) relative to the ‘spiked-in heavy’ generated using the z-score normalized $\log_2$L/H ratios by Perseus as described in materials and methods. The color gradient represents z-score normalized $\log_2$L/H ratios range from -4 (green) to 0 (black) to 4 (red). 0 represents the mean of all the proteins. Values smaller than 0 represent protein down regulation and values greater than 0 represent protein up regulation relative to the ‘heavy’ reference. The corresponding subcellular compartment marker protein enriched fractions determined by immunoblotting in Fig. 8A were indicated at the bottom (Fig. 8B). In general, the heat map showed that in lighter fractions 1-5 and heavier fractions 13-14, the total protein abundance is much lower than intermediate fractions 6-12. This is consistent with above mentioned expectation since the intermediate fractions contained most of ER, Golgi and endosome. It provided visualization of how proteins were distributed between and within subcellular compartments

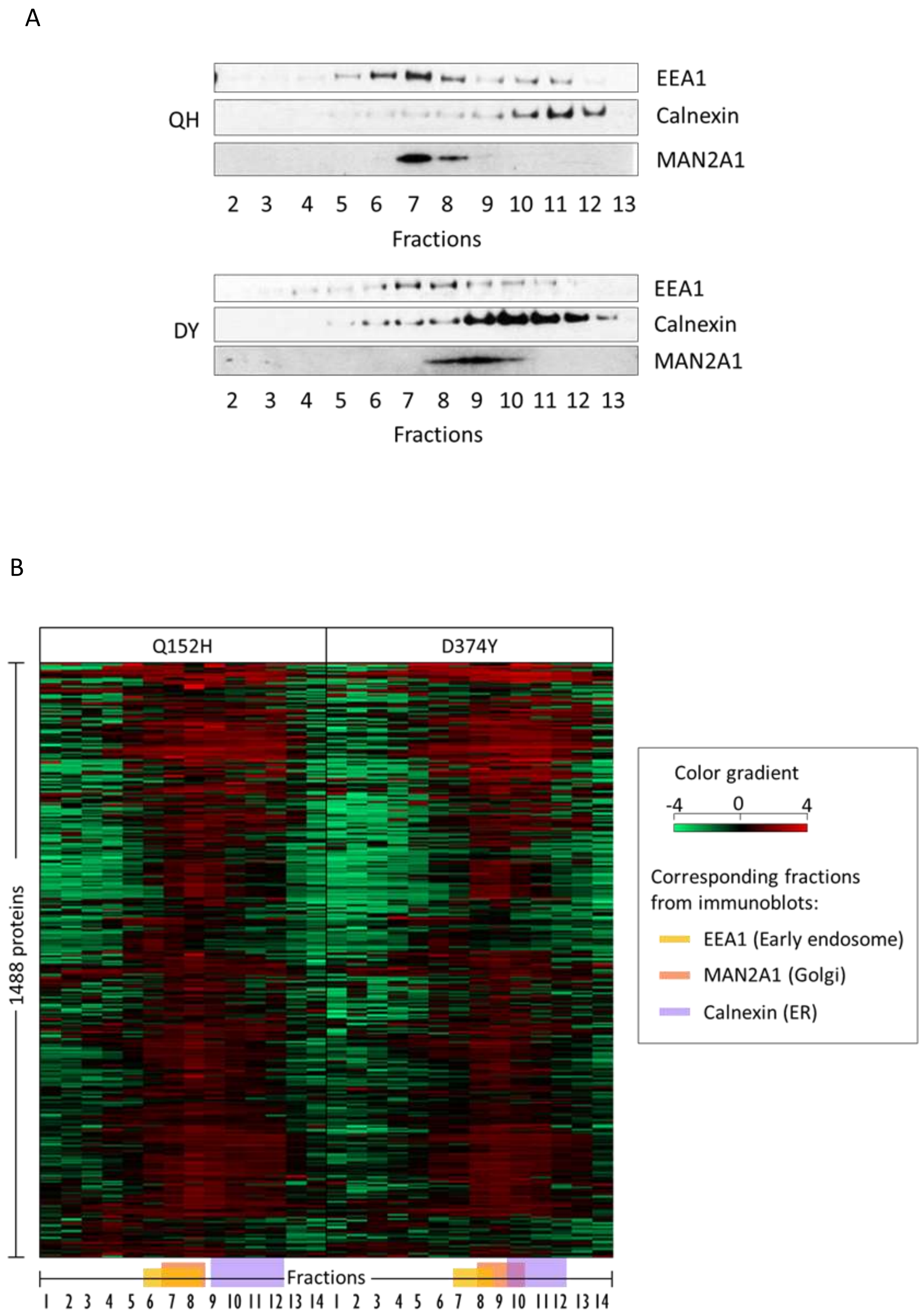

A
QH
EEA1
Calnexin
MAN2A1
2 3 4 5 6 7 8 9 10 11 12 13
Fractions
DY
EEA1
Calnexin
MAN2A1
2 3 4 5 6 7 8 9 10 11 12 13
Fractions
B
Q152H
D374Y
1488 proteins
Color gradient
-4 0 4
Corresponding fractions from immunoblots:
EEA1 (Early endosome)
MAN2A1 (Golgi)
Calnexin (ER)
Fractions
1 2 3 4 5 6 7 8 9 10 11 12 13 14 1 2 3 4 5 6 7 8 9 10 11 12 13 14

Figure 8. Subcellular fractionation results and protein distribution heatmap. (A) Immunoblots against anti-EEA1 (early endosome marker), anti-calnexin (ER marker) and anti-MAN2A1 (Golgi marker) for the fractionation of the 'light' microsomal pellets from HUH7 cells stably expressing PCSK9 Q152H and PCSK9 D374Y. (B) Heatmap of the protein expression level of 1501 proteins following subcellular fractionation. The color gradient represents z-score normalized $\log_2$L/H ratio range from -4 (green) to 4 (red). The corresponding subcellular compartment marker protein enriched fractions determined by immunoblotting in (A) are indicated at the bottom. Yellow block represents EEA1 enriched, orange represents MAN2A1 enriched and purple represents ER enriched fractions.

for Q152H and D374Y. It demonstrated that the fractionation profiles of Q152H and D374Y were highly consistent between each other. Stable expression of the LOF Q152H in comparison to the GOF D374Y did not grossly affect the overall profile of the majority of proteins and their subcellular distributions (Fig. 8B). These are illustrated with specific examples in Section 3.1.3.

3.1.3. Comparison of Subcellular Trafficking of Microsomal Proteins from HUH7 Cells Stably Expressing Q152H and D374Y PCSK9 Variants

To overcome subtle differences in the subcellular fractionations between samples and to improve the resulting comparisons, I developed and employed two normalization steps based upon the ‘spiked in’ reference material. The algorithms for these normalization steps were written and performed by Dr. Zhibin Ning. First, a longitudinal normalization was conducted to equalize the total ‘heavy’ intensities in every fraction as was initially ‘spiked in’ eliminating variations caused during sample processing and measuring on the mass spectrometer. Secondly, the total ‘light’ intensities in each fraction were adjusted and normalized according to the ‘heavy’ intensities. Algorithms were written to generate continuous protein L/H curves across the fractions calculated by the SILAC quantified L/H ratios of the 14 fractions for D374Y and Q152H. Representative protein profiles were extracted to illustrate the comparison of protein subcellular trafficking between LOF Q152H and GOF D374Y (green and red, respectively; Fig. 9). Consistent with the immunoblot data for anti-calnexin (Fig. 8A), the MS/MS detected calnexin (CANX) in fractions 6-14 and showed it as enriched in fractions 9-12 (Fig. 9A). Calnexin, as expected, was not detected in the earlier fractions that would contain lysosomes. This supports the accuracy of this approach.

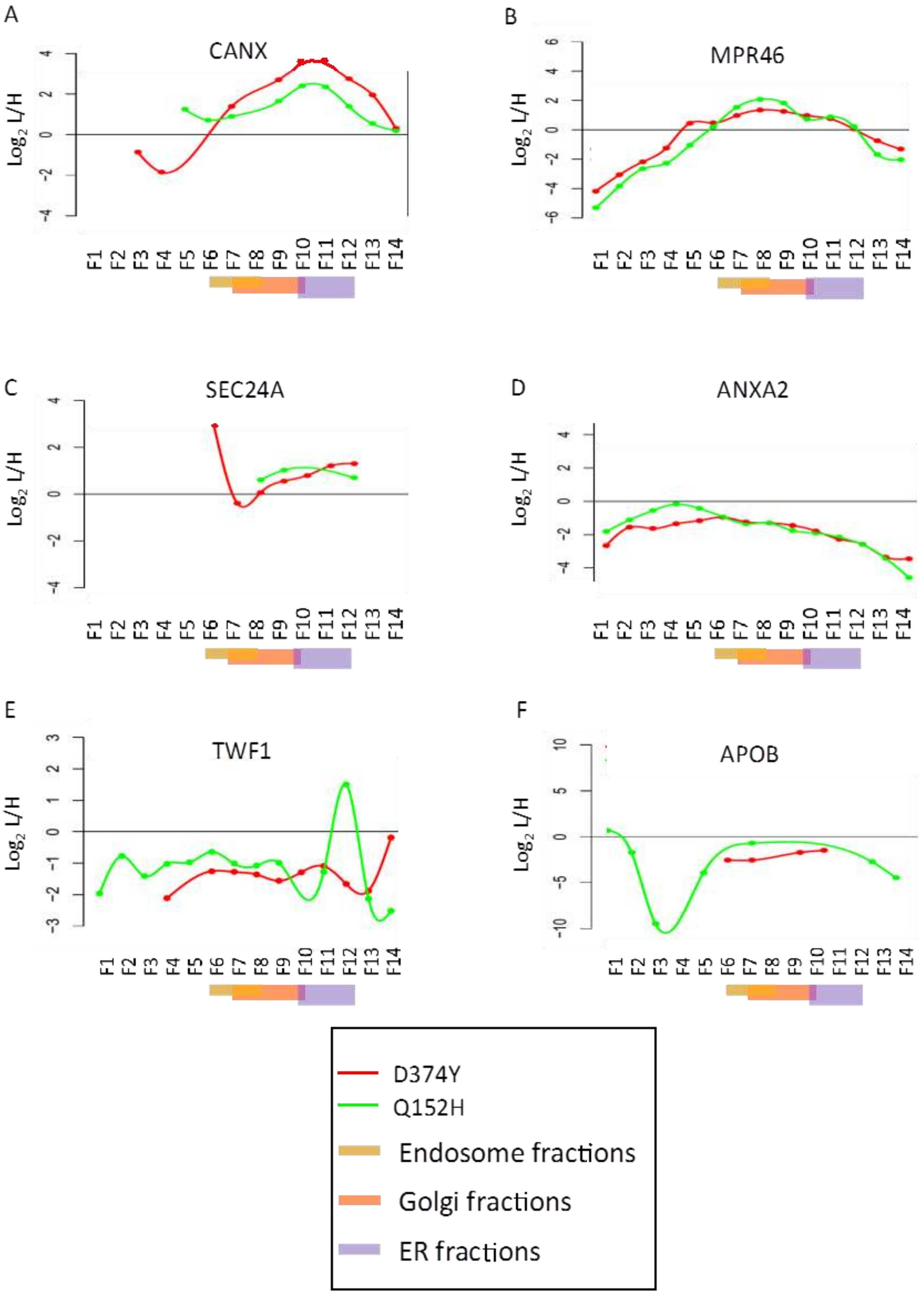

A
CANX
B
MPR46
C
SEC24A
D
ANXA2
E
TWF1
F
APOB
Log2 L/H
F1
F2
F3
F4
F5
F6
F7
F8
F9
F10
F11
F12
F13
F14
D374Y
Q152H
Endosome fractions
Golgi fractions
ER fractions

Figure 9. Representative profiles of $\log_2$L/H ratios of proteins following subcellular fractionation of microsomes from HUH7 cells stably expressing the PCSK9 Q152H and D374Y variants. (A). Calnexin (CANX, ER marker). (B). 46 kDa mannose 6-phosphate receptor (MPR46, late-endosomes/lysosomes marker). (C). SEC24 family, member A (SEC24A). (D). Annexin A2 (AnxA2). (E). Twinfilin actin binding protein 1 (TWF1). (F). Apolipoprotein B-100 (ApoB100). D374Y (DY), red; Q152H (QH), green. Corresponding subcellular compartment enriched fractions determined by immunoblotting in Figure 8A were indicated at the bottom. Yellow block represents endosome enriched, orange represents Golgi enriched and purple represents ER enriched fractions. Abbreviate: fraction, F.

The 46 kDa mannose 6-phosphate receptor (MPR46) is a transmembrane receptor that binds mannose-6-phosphate tags on acid hydrolase precursors in the Golgi that are destined for transport to the lysosome [153]. Thus it is largely localized in the late endosomes and Golgi [154]. From my data, the MPR46 was highly enriched in fractions 7-9, corresponding to the endosome and Golgi enriched fractions as expected (Fig. 9B). As mentioned previously, Sec24A is a component of coat protein II (COPII)-coated vesicles that mediate protein transport from ER to Golgi [155], and it assists PCSK9's exit from ER [94]. Thus Sec24A is largely localized on the membrane of ER and Golgi. My MS/MS data demonstrated that it was detected in the Golgi enriched fractions 7-10 and ER enriched fractions 10-12 as expected (Fig. 9C). The detection of Sec24A peptides was exclusive since there were no peptides detected in the other fractions. Annexin A2 (AnxA2) is secreted by a 'non-classical' mechanism [156]. As mentioned previously, it interacts with PCSK9 at the cell surface as an endogenous inhibitor for PCSK9 binding LDLR [90]. My subcellular fractionation data showed that the AnxA2 detected by MS/MS was mostly distributed in the early fractions 3-5 for both Q152H and D374Y (Fig. 9D). The subcellular trafficking data indicates a decrease in AnxA2 in the presence of D374Y in fractions 3-5 relative to Q152H however this is an n=1 and will need to be repeated before concluded that AnxA2 is affected by the PCSK9 D374Y. Twinfilin actin binding protein 1 (TWF1) was a protein identified related to skeleton organization and endocytosis [157]. It is shown in Figure 9E as a representative protein with difference in protein abundance in the same fraction between Q152H and D374Y. It had higher protein abundance in the ER enriched fraction 12 in Q152H than D374Y (Fig. 9E). ApoB100 was also identified in my data set (Fig. 9F). It was distributed in the endosome and golgi-ER fractions of both Q152H and D374Y (fractions 7-10). However, the Q152H was present in other subcellular fractions (1-5 and 11-14).

These preliminary differences in the protein distribution of ApoB100 might reflect the retention of ApoB100 in the ER with the LOF Q152H and/or reflect increased uptake of ApoB100 through the LDLR pathway.

3.1.4. Discussion

Density gradient centrifugation is a classic technique for monitoring protein distribution. Normally several proteins of interest are monitored and studied by subsequent ELISA or immunoblot analyses of each fraction. These studies can tell us how a protein is distributed or moves between fractions upon stimulation. In this section I wanted to combine the proteomic technologies with the classical density gradient centrifugation to develop a methodology that allows me to study on a global level how protein distribution and expression are affected by experimental stimuli. By ‘spiking in’ a heavy reference I was able to simultaneously study the profile of over 1500 proteins and how they changed in a liver cell line expressing PCSK9 LOF and GOF variants. The advantage of this method over traditional methods such as immunoblotting or immunofluorescence is that in the latter two methods only one or few proteins can be studied whereas my MS based analyses of protein changes in subcellular distribution allows me to monitor thousands of proteins in one experiment. For instance, in the experiment shown 1501 proteins were quantified followed by normalization to analyze the distribution pattern in the 14 fractions. This section has succeeded to the point of carrying out n=1 proof of the principle experiment followed by MS analyses comparing changes in the subcellular distribution of the strong GOF PCSK9 variant D374Y with the strong LOF Q152H variant. Overall the bulk of the proteins identified and quantified were not affected by PCSK9 loss- versus gain-of-function expression. There were changes observed. These will be pursued if they are reproduced n=3.

3.2. Comparative Global Proteomic Analyses of Secretomes from Human Hepatoma HUH7 Cells Following Overexpression of PCSK9 and Its Loss and Gain of Function Variants

As previously mentioned PCSK9 largely functions from the ‘outside-in’: binding to LDLR, mediating its degradation and regulating plasma cholesterol levels [76-78]. As well extracellular proteins have been implied in this mechanism [82, 98, 99, 158]. Yet, the global effect of PCSK9 and its variants on the milieu of the secretome has not been thoroughly studied. This is despite the fact that the most promising PCSK9 drugs are largely targeting the secreted form of PCSK9 and its extracellular function. My goal in this section was to study whether overexpression of PCSK9 and its variants affect other proteins in addition to LDLR.

3.2.1. Secretome Analyses Workflow

The workflow I developed for the secretome study is as illustrated in Figure 10. Briefly, and as described in materials and methods, ‘light’ HUH7 cells transiently transfected with V5 tagged PCSK9 wild type (WT), LOF variants (Q152H (QH), L10InsA53V (AV) and R46L (RL)), GOF variants (S127R (SR) and D374Y (DY)), and, as controls, empty vector (EV) and mock (MK) transfected, and, as the reference sample for quantification, SILAC ‘heavy’ HUH7 cells were cultured. 48-hour serum-reduced secretomes were collected and cleared of cellular debris before ‘spiking in’ of the ‘heavy’ reference material and subsequent processing for MS/MS analyses as described materials and methods. In total, 120 samples (MK, EV, WT and 5 PCSK9 variants × 5 SCX fractions × 3 replicates) were processed by MS/MS and analyzed by MaxQuant.

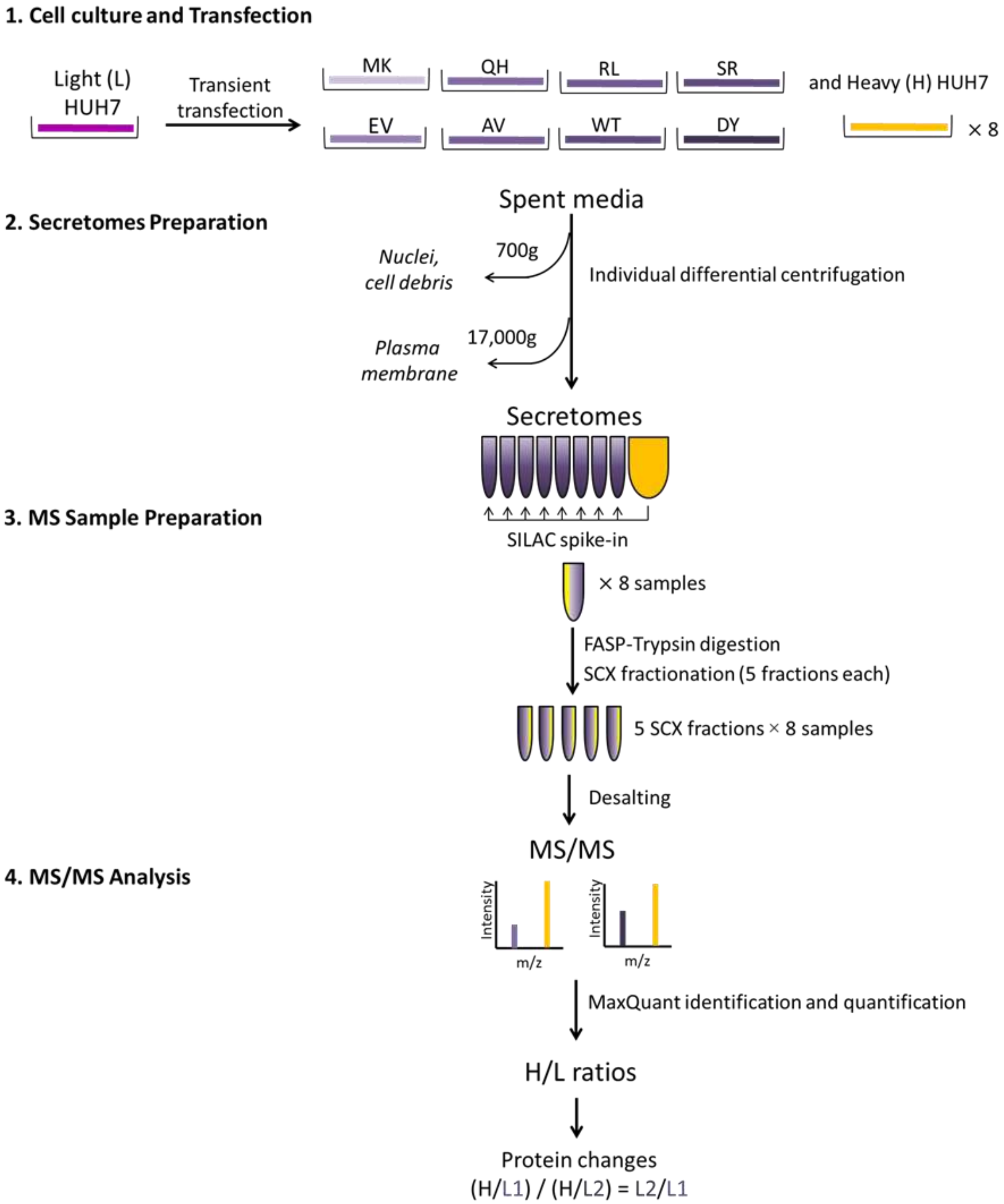

1. Cell culture and Transfection
Light (L) HUH7
Transient transfection
MK
QH
RL
SR
EV
AV
WT
DY
and Heavy (H) HUH7
× 8
2. Secretomes Preparation
Spent media
Nuclei, cell debris
700g
Individual differential centrifugation
Plasma membrane
17,000g
Secretomes
3. MS Sample Preparation
SILAC spike-in
× 8 samples
FASP-Trypsin digestion
SCX fractionation (5 fractions each)
5 SCX fractions × 8 samples
Desalting
4. MS/MS Analysis
MS/MS
Intensity
m/z
Intensity
m/z
MaxQuant identification and quantification
H/L ratios
Protein changes
(H/L1) / (H/L2) = L2/L1

Figure 10. Workflow for proteomic analyses of the secretomes from human hepatoma HUH7 Cells following overexpression of PCSK9 and its variants. Each of the secretomes from ‘light’ HUH7 cells transiently transfected with V5 tagged PCSK9 wild type (WT), Q152H (QH), L10InsA53V (AV), R46L (RL), S127R (SR), D374Y (DY) variants, empty vector (EV), mock (MK) transfected and SILAC ‘heavy’ HUH7 cells were collected. ‘Heavy’ secretomes were spiked into each of the light secretomes. The ‘spiked-in’ secretome samples were digested, fractionated, desalted and measured on MS/MS and analyzed by MaxQuant.

3.2.2. Characterization of PCSK9 Overexpression and SILAC Quantification

3.2.2.1. Transfection and PCSK9 Overexpression

PCSK9 overexpression was confirmed by ELISA for secretomes and by immunoblotting for total cell lysates (Fig 11A and B, respectively) following secretome collection (Step 2, Fig. 10) and prior to sample processing for MS/MS analyses (Step 3, Fig 10). Secreted PCSK9 was significantly lower in MK (25.8 ng/ml), EV (8.7 ng/ml) and Q152H (26.4 ng/ml) than PCSK9 WT (634.0 ng/ml) and the other 4 variants (A53V; 569.8 ng/ml, R46L; 590.1 ng/ml, S127R; 184.1 ng/ml and D374Y; 443.3 ng/ml) as assessed by ANOVA and Tukey's *post hoc* test ($p<0.05$). These levels were as expected since MK and EV did not have transfected PCSK9 and the secreted PCSK9 detected by ELISA reflected endogenous levels. Similarly, because transfected Q152H does not process the pro-PCSK9 nor secrete PCSK9, the ELISA results again represented the endogenous PCSK9 level. Cellular transfected PCSK9 levels were determined by immunoblotting against anti-V5 antibody (Fig. 11B). Intracellular PCSK9 forms included proPCSK9 (~74 kDa) and the auto-catalytically cleaved form (PCSK9, ~63 kDa). In MK and EV, neither form was detected since there were no transfected PCSK9 in these two samples. In the V5-PCSK9 transfected samples the two forms were detected and the ratios of the cleaved PCSK9/total PCSK9 were quantified by densitometry (Fig.11 C). The densitometry of the immunoblots was performed using ImageJ as described in material and methods. The average ratios of each sample were Q152H: 0.24, A53V: 0.69, R46L: 0.69, WT: 0.72, S127R: 0.49 and D374Y: 0.65. The cleaved PCSK9/total PCSK9 was significantly lower in Q152H, consistent with the fact that Q152H had greatly reduced proPCSK9 processing [141]. In addition, the cleaved PCSK9/total PCSK9 was significantly lower in S127R than A53V, R46L, D374Y and WT, consistent

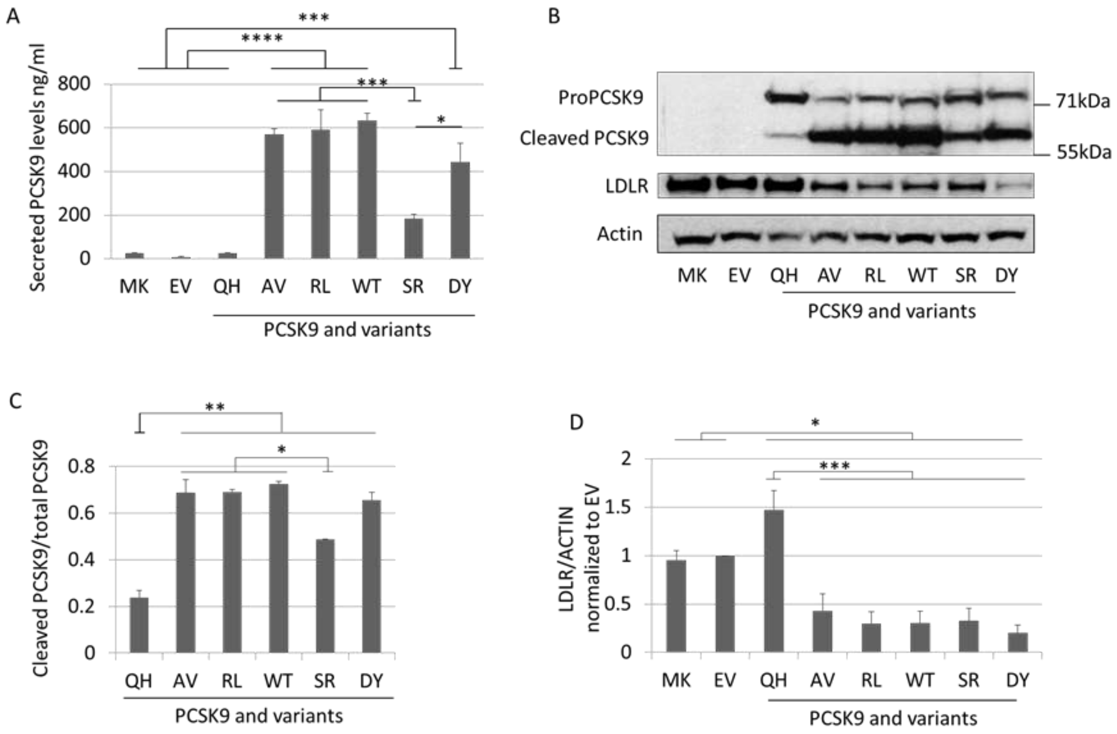

A
Secreted PCSK9 levels ng/ml
0
200
400
600
800
***
****
***
*
MK
EV
QH
AV
RL
WT
SR
DY
PCSK9 and variants
B
ProPCSK9
Cleaved PCSK9
LDLR
Actin
71kDa
55kDa
MK
EV
QH
AV
RL
WT
SR
DY
PCSK9 and variants
C
Cleaved PCSK9/total PCSK9
0
0.2
0.4
0.6
0.8
**
*
QH
AV
RL
WT
SR
DY
PCSK9 and variants
D
LDLR/ACTIN normalized to EV
0
0.5
1
1.5
2
*
***
MK
EV
QH
AV
RL
WT
SR
DY
PCSK9 and variants

Figure 11. Secreted and cellular PCSK9 and cellular LDLR levels from HUH7 cells transiently transfected with PCSK9 WT, its variants and controls. (A). Secreted PCSK9 level (ng/ml) assessed by PCSK9 ELISA (B). Representative immunoblots of the total cell lysates against anti-V5, human LDLR and actin antibodies. V5-tagged PCSK9 presented with proPCSK9 form (~74kDa) and cleaved PCSK9 form (~63kDa). (C). Cleaved PCSK9/total PCSK9 densitometry of the anti-V5 immunoblot. (D). Relative LDLR levels in cells by densitometric analysis, normalized to actin with EV samples set as 1. Data are presented as the mean ± SEM (n=3). Differences were assessed by ANOVA with Tukey's *post hoc* comparison test. Significance is indicated as $*p<0.05$, $**p<0.001$ and $***p<0.0001$. PCSK9 wild type (WT), Q152H (QH), L10InsA53V (AV), R46L (RL), S127R (SR), D374Y (DY), empty vector (EV) and mock (MK) transfection.

with previous finding of decreased prodomain cleavage of S127R [36].

Cellular LDLR level was determined by immunoblotting against human LDLR and against actin (housekeeping protein) as a reference (Fig. 11B). LDLR was normalized to actin for each sample and the ratio of LDLR/Actin for EV then set as 1. LDLR level is significantly higher in MK (0.96), EV (1) and Q152H (1.47) than in the PCSK9 WT (0.43) and the other 4 variants (A53V; 0.30, R46L; 0.30, S127R; 0.32 and D374Y; 0.20) as assessed by ANOVA and *post hoc* test ($p<0.05$, Fig. 11D). The higher LDLR level in Q152H versus controls is consistent with the previously reported dominant-negative effect of this variant on WT PCSK9 which up-regulates LDLR relative to un-transfected or transfected cells. [141]. Although not statistically significant, the LDLR level in D374Y was lowest. This is as expected for D374Y as the strongest GOF variant.

3.2.2.2. Characterization of the 'Heavy' Spike-in Reference for SILAC Quantification

Following these preliminary analyses, and again before mass spectrometry preparation I ensured that the total protein abundance in each of the secretome samples to be analyzed was similar and therefore comparable for the 'spike in' workflow. Figure 12A (upper panel) shows a representative colloidal blue stained gel of secretomes fractionated by SDS-PAGE. The total protein abundances between each of the 'light' secretomes from transfected HUH7 cells were similar as assessed by densitometry analysis (Fig. 12A lower panel). Figure 12B (upper panel) shows a representative colloidal blue stained gel of n=3 'light' WT PCSK9 secretomes and the 'heavy' secretome to be 'spiked in' secretome fractionated by SDS-PAGE. Densitometry illustrates that the WT PCSK9 biological replicates are similar in protein abundance (Fig. 12B lower panel). The 'heavy' secretome to be used as the MS/MS

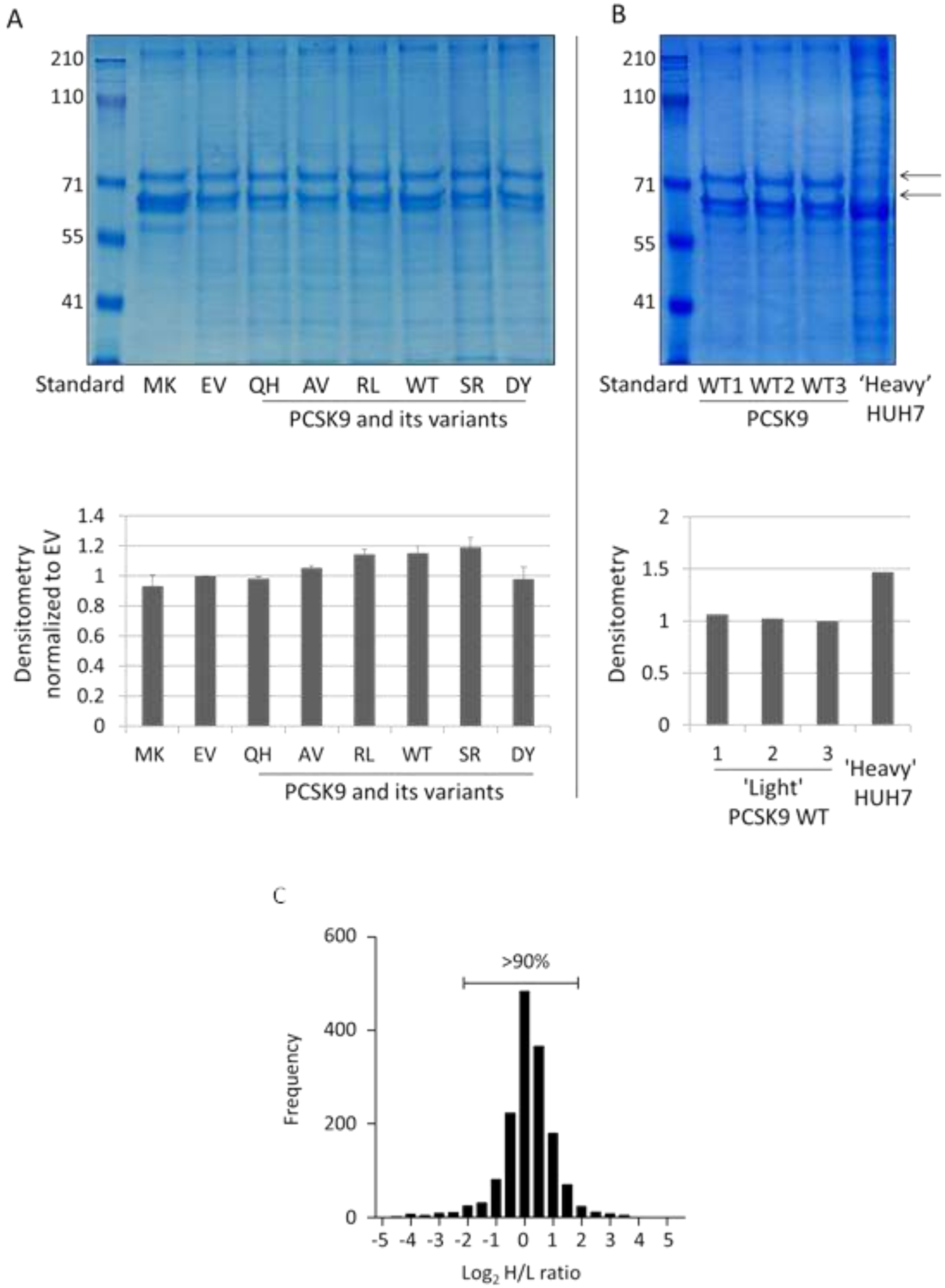

A
210
110
71
55
41
Standard
MK
EV
QH
AV
RL
WT
SR
DY
PCSK9 and its variants
Densitometry normalized to EV
1.4
1.2
1
0.8
0.6
0.4
0.2
0
B
Standard
WT1
WT2
WT3
PCSK9
'Heavy' HUH7
Densitometry
2
1.5
1
0.5
0
1
2
3
'Light' PCSK9 WT
C
>90%
Frequency
600
400
200
0
-5
-4
-3
-2
-1
0
1
2
3
4
5
Log2 H/L ratio

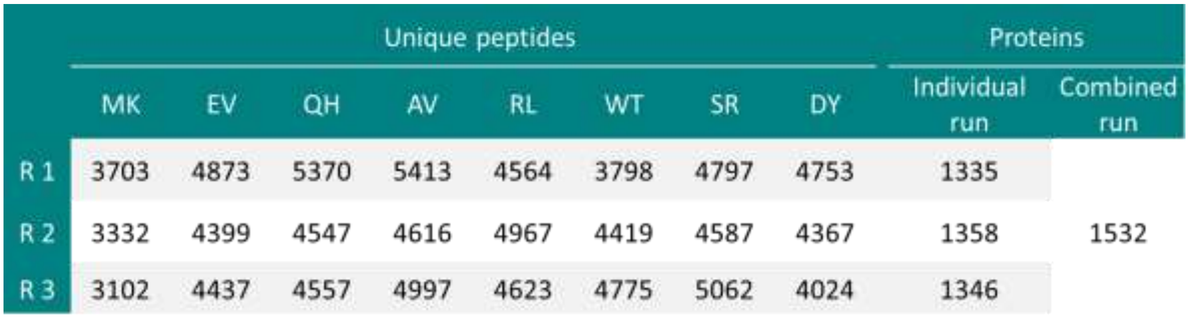
Figure 12. Protein abundance of equal volume of ‘light’ secretomes and evaluation of the quality of the SILAC ‘heavy spike-in’ standard. (A). Protein abundance of equal volume of ‘light’ secretomes assessed by SDS-PAGE fractionation and colloidal blue staining. Densitometry of each lane was normalized to EV =1 (means ± SEM, n=3). (B). Protein abundance of equal volume of ‘light’ WT (n=3) and ‘heavy’ secretomes assessed by SDS-PAGE fractionation and colloidal blue staining. Densitometry was normalized to WT replicate 3 (WT3) =1. (C). Frequency distribution of $\log_2$ H/L of proteins. More than 90% of the proteins demonstrate a $\log_2$ H/L ratio within range of -2 to 2. PCSK9 wild type (WT), Q152H (QH), L10InsA53V (AV), R46L (RL), S127R (SR), D374Y (DY), empty vector (EV) and mock (MK) transfection.

‘spike in’ reference material was similar, but not identical, in gross composition (that is comparing the pattern and intensity of colloidal blue stained proteins). For example, for certain proteins, the ‘heavy’ secretome contained less than the ‘light’ secretome samples (indicated by arrows in Fig. 12B upper panel). This is not unexpected since the source and base media for the ‘heavy’ reference cell line is different from the ‘light’ (DMEM for ‘heavy’ cells and OPTI-MEM for ‘light’ cells, respectively). This can change the expression and secretion pattern of cells, however, for my purpose the ‘heavy’ reference is valid if it contains comparable levels of the majority of the proteins (90% proteins within 10-fold difference) represented in the experimental ‘light’ samples [145]. To ensure maximum coverage of the ‘light’ secreome by the ‘heavy’ one, I spiked in ‘heavy’ secretome samples as a reference for the MS/MS quantification at 80% of the volume of the ‘light’ secretome samples. Following MaxQuant analyses, the quality of the ‘heavy’ reference was assessed by frequency distribution histogram of the $\log_2$ ratio H/L as instructed in the standard MaxQuant procedure [145]. The histogram shows that the ‘heavy’ reference efficiently represented the sample (>90% of $\log_2$ ratio H/L within the range of -2 to 2; Fig. 12C) [145].

3.2.2.3. Determination of the Experimental Control and Confirmation of the SILAC Quantification for Identifying Protein Changes

In each 1 of the 3 MaxQuant experimental runs (containing 8 samples: MK, EV, PCSK9 WT and variants), ~1350 proteins were identified (Table 3). When analyzed together by MaxQaunt in one experimental run, 1532 proteins were identified. This showed over 87.5% reproducibility between MaxQuant runs as defined by the overlap of identified proteins (Appendix 2). The number of unique peptides identified in each MK, EV, PCSK9 WT and variants in 3 biological replicates are as shown in Table 3. The average number of unique peptides in MK (3379) was significantly lower than the other secretome samples (4330-5008) assessed by ANOVA and Tukey's *post hoc* comparisons ($p<0.05$, Fig. 13A). This indicates that transfection of a plasmid versus a mock transfection has significant impact on the secreted proteome. Thus, for future analyses, MK was not considered as an appropriate control.

Table 3. Number of unique peptides and proteins identified by MS/MS in comparative global proteomic analyses of secretomes from HUH7 cells overexpressing PCSK9 and its variants



	Unique peptides								Proteins	
	MK	EV	QH	AV	RL	WT	SR	DY	Individual run	Combined run
R 1	3703	4873	5370	5413	4564	3798	4797	4753	1335	1532
R 2	3332	4399	4547	4616	4967	4419	4587	4367	1358	
R 3	3102	4437	4557	4997	4623	4775	5062	4024	1346	

Abbreviation: mock, MK; empty vector, EV; Q152H, QH; L10insA53V, AV; R46L, RL; wild type, WT; S127R, SR; D374Y, DY.

The quality of secretome from EV as an appropriate control was further evaluated by histograms comparing EV's H/L ratio to the others' in Figure 13. This comparison was conducted using 'ratio of ratios', where the H/L ratio of sample 1 is divided by the H/L ratio of sample 2 (resulting in a ratio of sample 2/sample 1) [145]. In this case, 'ratio of ratios' of each PCSK9 (WT and variants) to EV was obtained and generated histograms in base 2 logarithmic scale ($\log_2$ PCSK9/EV, Fig. 13). The histograms showed that the frequency distributions of $\log_2$ PCSK9/EV ratios were centered at 0 and over 90% were within range of -1 to 1. This indicates that the majority of proteins were not changing when comparing secretomes of EV to PCSK9 WT and variants. Therefore, EV serves as an appropriate control for quantitative analysis in terms of total secretome levels. To confirm the SILAC

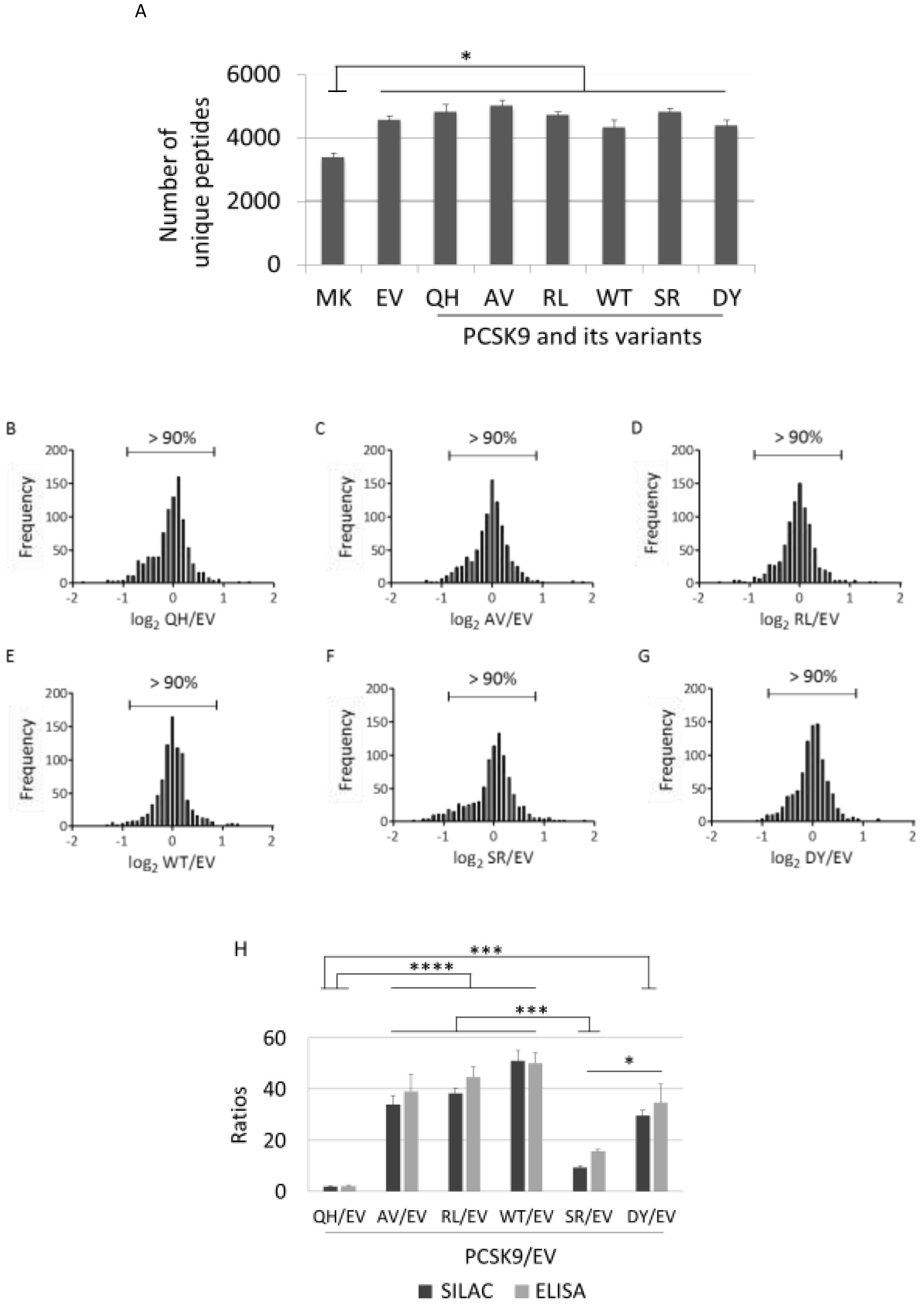
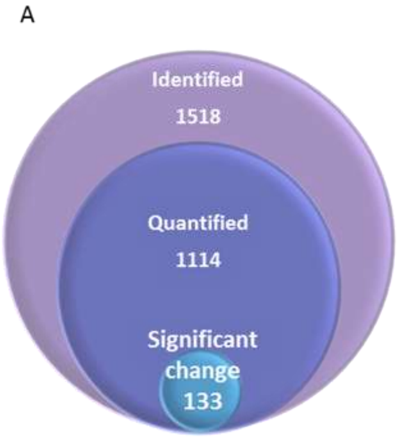

A
*
Number of unique peptides
6000
4000
2000
0
MK
EV
QH
AV
RL
WT
SR
DY
PCSK9 and its variants
B
> 90%
Frequency
200
150
100
50
0
-2
-1
0
1
2
$\log_2$ QH/EV
C
> 90%
$\log_2$ AV/EV
D
> 90%
$\log_2$ RL/EV
E
> 90%
$\log_2$ WT/EV
F
> 90%
$\log_2$ SR/EV
G
> 90%
$\log_2$ DY/EV
H
***
****
***
*
Ratios
60
40
20
0
QH/EV
AV/EV
RL/EV
WT/EV
SR/EV
DY/EV
PCSK9/EV
SILAC
ELISA

Figure 13. Determination of experimental controls and confirmation of the SILAC quantification by comparison of the SILAC ‘ratio of ratios’ to the ELISA results of secreted PCSK9. (A). The number of unique peptides identified in secretome samples from transiently transfected HUH7 cells by MS/MS and MaxQuant analyses (n=3, means ± SEM). Asterisk indicates significant difference assessed by ANOVA and Tukey’s *post hoc* comparisons (* $p<0.05$). (B-G). Evaluation of the quality of secretome from EV as a quantification control by histograms of secretome ‘ratio of ratios’ (B). $\log_2$ QH/EV; (C). $\log_2$ AV/EV; (D). $\log_2$ RL/EV; (E). $\log_2$ WT/EV; (F). $\log_2$ SR/EV; (G). $\log_2$ DY/EV. The frequency distributions of these $\log_2$ PCSK9/EV ratios were all centered at 0. Over 90% of the proteins had a ‘ratio of ratios’ within range of -1 to 1 between secretomes from PCSK9 (WT and variants) and EV. (H). SILAC H/L ratio of EV was divided by H/L ratios of PCSK9 WT and variants to obtain the ‘ratio of ratios’ of QH/EV, AV/EV, RL/EV, WT/EV, SR/EV and DY/EV. PCSK9 concentration from EILSA results of each PCSK9 WT and variants were divided by concentration of EV. Significance was assessed by ANOVA and Tukey’s *post hoc* analyses (* $p < 0.05$). EV, empty vector; QH, Q152H; AV, L10insA53V; RL, R46L; WT, wild type; SR, S127R; DY, D374Y.

quantification, the SILAC ‘ratio of ratios’ of PCSK9 were compared with the ratios of ELISA results of each PCSK9 WT and variants normalized to EV (Fig. 13H). The MS/MS SILAC quantified ratios between WT and variants were consistent with the ELISA results as shown in Figure 13 confirming the accuracy of this approach.

3.2.3. Characterization of Significantly Changed Secretome Proteins by Hierarchical Clustering Analyses

Among the 1532 proteins identified, 1114 proteins were quantified (73%) in greater than 50% of all biological replicates (Appendix 3) and 133 proteins (12%) showed significant changes as assessed by ANOVA (Appendix 4 and Fig.14A). Cellular localization annotation of the 133 proteins was performed using Ingenuity Pathway Analysis (IPA) as previously described. 63 proteins (47.4%) were annotated as “extracellular”, 44 proteins (33.1%) as “cytoplasm” and 12 proteins (10%) as “plasma membrane”. Many proteins annotated as “cytoplasm” are also released to extracellular space through non-classical secretion such as heat shock proteins (HSPA4, HSPA5, HSPB1 and HSPE1) and proteasome subunits (PSMB2, PSMD3) [159-161]. Some proteins annotated as “plasma membrane” are also known to distribute in the extracellular space such as Annexins (AnxA2 and AnxA5) again through non-classical secretion routes [156]. In order to investigate patterns within the significantly changed proteins, z-score normalized medians of $\log_2$ L/H ratios were subject to hierarchical clustering by Perseus as described in the materials and methods (Fig. 14B). The 133 proteins were grouped into five major clusters as shown in Figure 14C. PCSK9 was found in Cluster C, where its abundance was lower in EV and Q152H than other PCSK9s as expected. Except for PCSK9, the proteins annotated “extracellular” in terms of cellular location as their major distribution in IPA all belonged to Cluster E.


A
Identified
1518
Quantified
1114
Significant
change
133

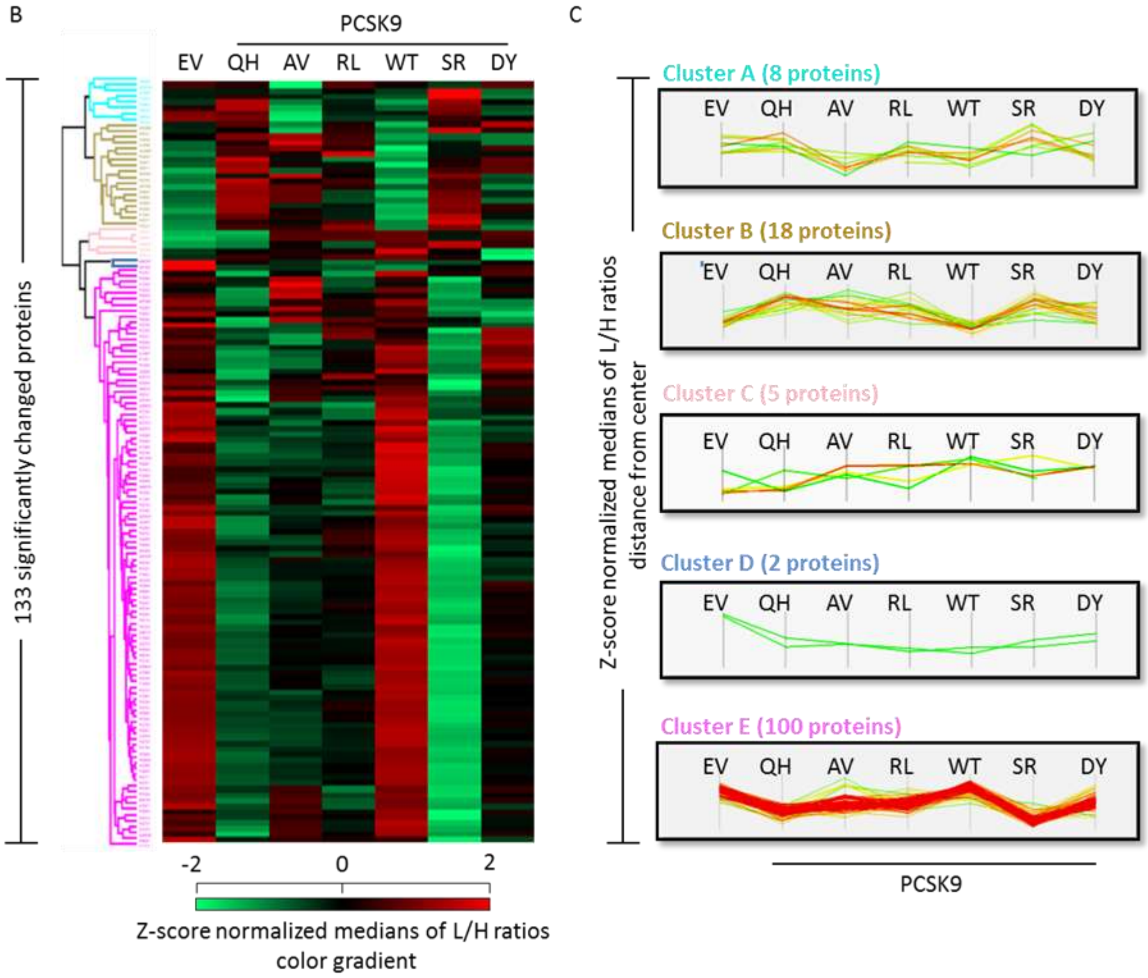

B
PCSK9
EV
QH
AV
RL
WT
SR
DY
133 significantly changed proteins
-2
0
2
Z-score normalized medians of L/H ratios
color gradient
C
Cluster A (8 proteins)
Cluster B (18 proteins)
Cluster C (5 proteins)
Cluster D (2 proteins)
Cluster E (100 proteins)
EV
QH
AV
RL
WT
SR
DY
Z-score normalized medians of L/H ratios
distance from center
PCSK9

Figure 14. Significantly changed secretome proteins and their hierarchical clustering. (A). Number of protein identified by MS/MS (1518), quantified by MaxQuant (1114) and significantly changed assessed by ANOVA (133). (B). Heat map of the 133 significantly changed proteins generated by their z-score normalized medians of $\log_2$ L/H ratios in EV, PCSK9 WT and variants (Q152H (QH), A53V (AV, R46L (RL), S127R (SR), D374Y (DY)). Color gradient from -2 (down regulated, green) to 2 (up regulated, red). (C): Protein abundance changing patterns among EV, PCSK9 WT and variants of the five hierarchical clusters A to E. The curves were based on the distances from center after z-score normalization of $\log_2$ L/H medians.

Cluster E contained 100 proteins with a general changing pattern of higher abundance in WT while lower in Q152H and S127R (Fig. 14C). In contrast, proteins in cluster B demonstrate opposite patterns to cluster E, where WT is decreased, while Q152H and S127R are increased. Cluster A and B share similar pattern except for A53V with decreased level in Cluster A. Cluster D shows proteins with reduced level upon any PCSK9 overexpression. These protein abundance changes observed in the secretome may be affected directly or indirectly by PCSK9 that caused changes in the protein secretion rates and/or rates of protein endocytosis and degradation.

To analyze initially those proteins with the largest fold changes I set up the following criteria: (1) passed ANOVA, (2) 1% FDR, (3) ≥ 2 unique peptides detected and (4) >2 fold changes between the highest L/H ratio and lowest L/H ratio of EV, PCSK9 WT and variants. This narrowed the initial 133 significantly changed secretome proteins to the 20 proteins listed in Table 4. These 20 proteins all belonged to Cluster E (Fig. 14C) except for PCSK9 (Cluster C, Fig. 14C).

Among these, six were apolipoproteins (Apo): ApoE, ApoA1, ApoC2, ApoC-3, ApoM and ApoJ (Clusterin). Like ApoB100 their levels may be affected by PCSK9 interaction along the secretory pathway or by their uptake through LDLR. As well, the reduction might also result from PCSK9's action towards receptors of these apolipoproteins beyond LDLR like VLDL and APOER2 [104, 105]. The different classes of significantly changed apolipoproteins may reveal a broader effect of PCSK9 and its variants on the lipoprotein and apolipoprotein metabolisms beyond LDLR and LDL regulation. As previously mentioned, PCSK9 was also found to promote VLDLR and APOER2 degradation [105], however this regulatory effect on the apolipoprotein level changes has not been thoroughly studied.

Table 4. List of proteins with over 2 folds changes between the highest and lowest L/H ratios of EV, PCSK9 WT and variants in secretome analyes from HUH7 cells overexpressing PCSK9 and its variants

Max fold change ($\log_2$)	Gene name	Protein name	Unique peptides	Median $\log_2$ L/H ratio						
				EV	QH	AV	RL	WT	SR	DY
5.49	PCSK9	proprotein convertase subtilisin/kexin type 9	28	-2.27	-1.46	2.86	2.91	3.22	0.92	2.53
1.82	COL2A1	collagen, type II, alpha 1	22	3.08	1.67	2.08	2.33	3.22	1.40	2.60
1.48	APOE	apolipoprotein E	46	0.64	-0.11	0.46	0.73	1.38	0.16	0.96
1.23	APOC2	apolipoprotein C-II	5	-1.20	-1.83	-1.64	-1.52	-0.75	-1.98	-1.34
1.23	HP	haptoglobin	8	0.73	-0.09	0.03	0.06	0.52	-0.50	0.42
1.22	SERPIND1	serpin peptidase inhibitor, clade D, member 1	12	-0.25	-0.98	-0.71	-0.72	-0.16	-1.38	-0.66
1.21	CLSTN3	calsyntenin 3	8	-0.99	-1.36	-1.26	-1.52	-0.70	-1.91	-1.31
1.19	APOA1	apolipoprotein A-I	6	0.67	-0.04	0.22	0.41	0.81	-0.39	0.37
1.18	APOM	apolipoprotein M	6	1.20	0.70	0.96	0.71	1.50	0.32	0.97
1.17	KNG1	kininogen 1	24	-0.50	-1.12	-1.07	-0.86	-0.40	-1.58	-0.87
1.16	F2	coagulation factor II (thrombin)	12	-0.78	-1.47	-1.22	-1.18	-0.71	-1.87	-1.28
1.15	SERPINA1	serpin peptidase inhibitor, clade A, member 1	29	0.01	-0.62	-0.19	-0.38	0.45	-0.70	-0.14
1.09	AHSG	Alpha-2-HS-glycoprotein	12	-0.19	-0.99	-0.87	-0.75	-0.22	-1.28	-0.67
1.08	CLU	clusterin	31	-0.45	-1.01	-0.80	-0.80	-0.25	-1.32	-0.77
1.06	SERPINF2	serpin peptidase inhibitor, clade F, member 2	12	0.61	-0.02	0.02	0.14	0.72	-0.34	0.17
1.06	F1	coagulation factor X	6	-0.10	-0.77	-0.64	-0.30	0.01	-1.04	-0.45
1.02	BNC2	basonuclin 2	2	-0.12	-1.03	-0.73	-0.64	-0.27	-1.15	-0.65
1.01	SPINK1	serine peptidase inhibitor, Kazal type 1	7	1.28	0.77	0.76	0.96	1.34	0.32	0.99
1.00	APOC3	apolipoprotein C-III	3	0.49	-0.30	0.11	0.25	0.57	-0.44	-0.01
1.00	COL5A2	collagen, type V, alpha 2	18	1.45	1.25	1.22	1.23	1.79	0.79	1.30

Abbreviation: Q152H, QH; A53V, AV; R46L, RL; S127R, SR; D374Y, DY.

Herein, my data showed that the overexpression of PCSK9 and its variants affected apolipoprotein levels differently. This may reflect PCSK9's role in the metabolism of other lipoprotein receptors as well as conversions between lipoproteins.

Six of the 20 proteins with over 2-fold changes were related to coagulation pathway, namely coagulation factor II (thrombin, F2), coagulation factor X (F1), kininogen 1 (KNG1), serpin peptidase inhibitor, clade A, member 1 (SERPINA1), serpin peptidase inhibitor, clade D member 1 (SERPIND1) and serpin peptidase inhibitor clade F member 2 (SERPINF2; Tables 4). This indicates that PCSK9 and its variants might play a role in the coagulation system that has not been reported yet. In the secretome data set, 15 serine peptidase inhibitors (serpins) were detected: A1, A4, A5, A6, B1, B6, B9, C1, D1, E1, F1, F2, G1, H1 and I1 (Appendix 2). Eight of the serpins showed significant changes as assessed by ANOVA: A1, A4, A5, A6, D1, F1, F2 and I1 (Appendix 4). As above mentioned, A1, D1 and F2 exhibited over 2-fold changes among the EV, PCSK9 WT and variants. This showed the effect of PCSK9 on the serpins were specific on different clades. As above mentioned, extracellular serpinA1, A5, D1, F2 are involved in coagulation pathway and serpinA1, A3, D1, F1, F2 are involved in acute phase response signalling pathway [162]. This indicates that PCSK9 and its variants may have a specific effect on these pathways. In fact, PCSK9 has been reported to be up regulated in the acute period of myocardial infarction in rat [163], supporting my observations.

The other proteins showing over 2-fold changes included: collagens (type II, alpha 1 (COL2A1) and type V, alpha 2 (COL5A2)), one of the predominant structural proteins of the liver extracellular matrix and is involved in stem cell proliferation and differentiation [164]; haptoglobin (HP), a regulator of reverse cholesterol transport in diabetes aptoglobin [165];

calsyntenin 3 (CLSTN3), involved in modulating calcium-mediated postsynaptic signals [166]; alpha-2-HS-glycoprotein (AHSG), an important circulating inhibitor of calcification associating with increased risk of cardiovascular disease [167] and insulin resistance [168]; alpha-2-HS-glycoproteinbasonuclin 2 (BNC2), implied to be involved in ovarian cancer [169]; serine peptidase inhibitor, Kazal type 1 (SPINK1), related to pancreatitis disease [170] (Table 4). The underlying involvement of PCSK9 and its variants in the regulation of these proteins and the related effects require further investigation.

3.2.4. Discussion

The ‘secretome’ was defined as a portion of proteins secreted to the extracellular space in the classical secretory pathway [171] and ‘non-classical’ secretory pathways [156], such as proteins contained in secreted organelles such as microvesicles and exosomes [172], as well as plasma membrane proteins whose extracellular domains are shed into the extracellular milieu [159]. In addition, nuclear and cytoplasmic proteins have also been found to be released to the extracellular environment and considered as bona fide components of the secretome [159]. The secretome proteins control and regulate a multitude of biological and physiological processes such as cell migration, cell signaling and communication [173]. This is the first study documenting the total secretome in HUH7 cells and the secretome changes upon PCSK9 overexpression. The objective of this section was to identify changes in secretomes of HUH7 cells overexpressing PCSK9 and its variants to investigate the global effect of PCSK9 in the extracellular matrix and the difference between its variants. Three replicates of the total secretome study have been conducted using the SILAC ‘spike-in’ quantification method. 12% of the proteins have shown significant changes between the control (EV), PCSK9 WT and variants, where 20 proteins exhibited over 2-fold changes.

However, on a global scale, 90% of the proteins identified in the secretomes of each EV, PCSK9 WT and variants were not changing with statistical significance. This indicated that the effects of the PCSK9 variants on the secretome were extremely specific on lipoprotein metabolism and likely on the coagulation system and acute phase response signalling pathway as well. Furthermore, this suggests that the strategies of the pharmaceutical industry towards inhibiting secreted PCSK9 function are not likely to induce vast physiological changes.

3.3. Proteomic Analyses and Identification of the Secreted Interactome in HUH7 Cells of PCSK9 and Its Variants

As previously described, PCSK9 mainly functions as a secreted, escort protein and extracellular accessory proteins have been implicated in PCSK9-mediated LDLR rerouting [98, 99]. The identification of the accessory factors and their functions will further elucidate the exact mechanism of PCSK9's action towards LDLR. To date, several extracellular proteins have been identified that affect secreted PCSK9 action including Annexin A2, ApoB100 and APLP2 [90-92], and pharmaceutical antibody drugs targeting extracellular PCSK9 are playing the role of inhibitory interactors. Whether there would be other endogenous inhibitors that bind to PCSK9 extracellularly is worthy of investigation, as well as the effects caused by presence and loss their interaction.

Herein, my goal is to identify the secreted interactome of PCSK9 and its variants upon their overexpression in the HUH7 cell line. To my knowledge this is the first global study using proteomics to investigate the secreted interaction of PCSK9.

3.3.1. Secreted PCSK9 Interactome Analyses Workflow

The workflow toward identifying the secreted PCSK9 interactome is as demonstrated in Figure 15. HUH7 cells transiently transfected with plasmids expressing V5-tagged EV, PCSK9 WT, Q152H, A53V, R46L, S127R, and D374Y or MK transfected were cultured in serum-reduced media for 48 hours and secretomes harvested as described in materials and methods. Co-immunoprecipitations of freshly harvested secretomes were carried out using anti-V5 affinity agarose beads followed by SDS-PAGE fractionation. One-tenth of each sample was fractioned for immunoblotting to validate PCSK9 immunoprecipitation and the remainder for fractionation and Colloidal Blue staining to visualize co-immunoprecitated proteins, and for gel cutting and in-gel digestion of these proteins as described in materials and methods. The co-immunoprecipitation was successfully completed for 3 replicates each of MK, EV, WT PCSK9 and its variants. In total 24 gel slices/lane × 8 lanes were subject to in-gel digestion and MS/MS measurements. MaxQuant was used for database searching and label-free quantification (LFQ). The quantification was performed using two levels of analyses: (1) by whole lanes and (2) by individual gel slices. As the first step of analyses, quantifying the co-immunoprecipitated proteins by whole lanes allowed me to quickly identify as well as observe interesting protein changes among MK, EV, WT PCSK9 and its variants. This output generated my primary list of potential PCSK9 interactors. The second level of analyses, quantifying by individual gel slices allowed me to match the known molecular weight of the identified interacting PCSK9 proteins or its isoforms with its SDS-PAGE fractionated apparent molecular weight by SDS-PAGE fractionation. This allowed me to determine if its isoforms demonstrated similar or differing interaction patterns with secreted PCSK9.

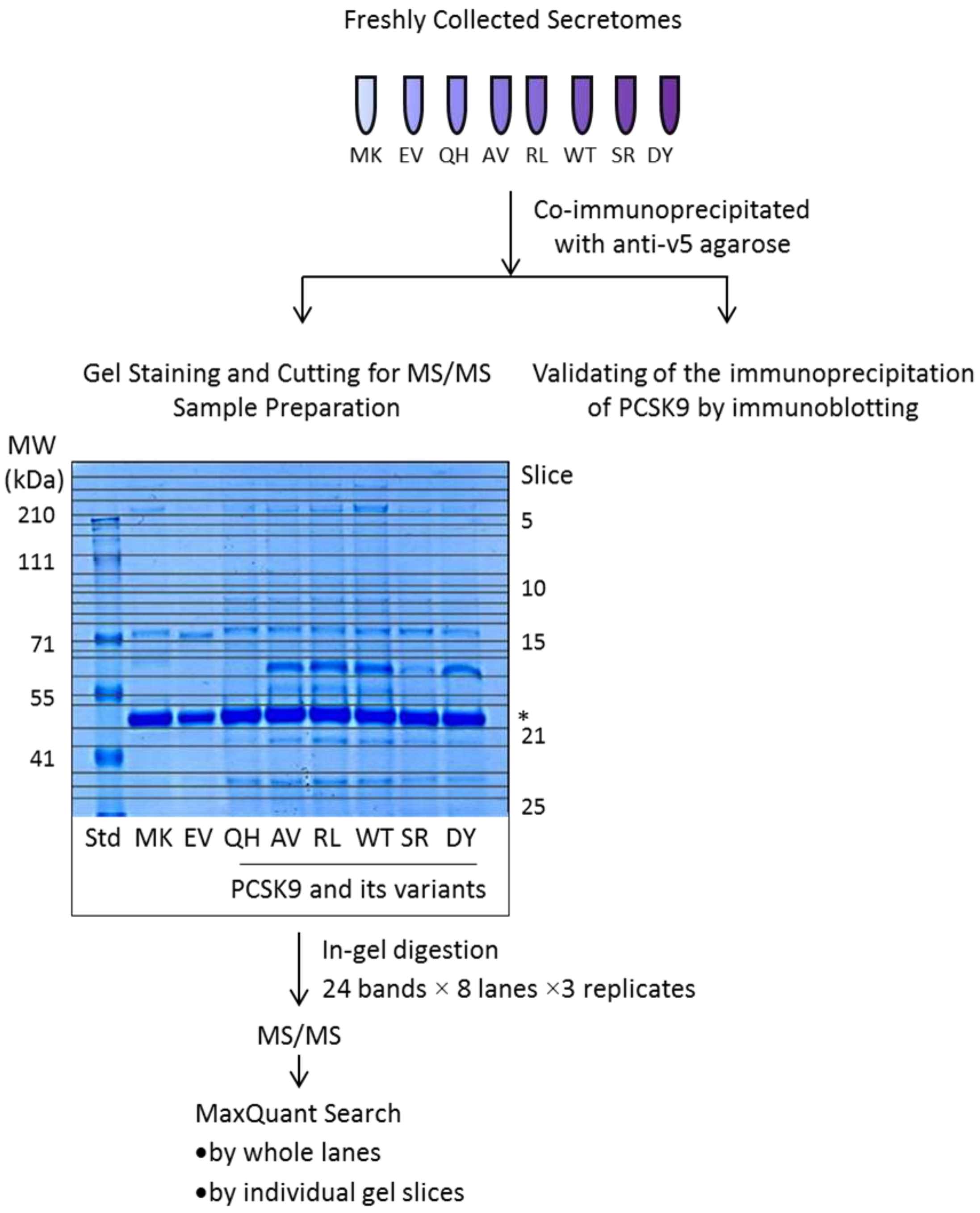

Freshly Collected Secretomes
MK EV QH AV RL WT SR DY
Co-immunoprecipitated with anti-v5 agarose
Gel Staining and Cutting for MS/MS Sample Preparation
Validating of the immunoprecipitation of PCSK9 by immunoblotting
MW (kDa)
210
111
71
55
41
Slice
5
10
15
*
21
25
Std MK EV QH AV RL WT SR DY
PCSK9 and its variants
In-gel digestion
24 bands × 8 lanes ×3 replicates
MS/MS
MaxQuant Search
•by whole lanes
•by individual gel slices

Figure 15. Workflow for proteomic analyses and identification of the secreted interactome of PCSK9 and its variants in HUH7 cells. Co-immunoprecipitations were carried out from the secretomes of transiently transfected HUH7 cells with V5-tagged EV, PCSK9 WT, QH, AV, RL, SR, DY and MK transfection, and confirmed by immunoblotting. SDS-PAGE fractionation, gel cutting and in-gel digestion of co-immunoprecipitated proteins were conducted. Following gel fractionation each lane was cut into 25 slices as indicated by the lines in the gel image. The representative slice numbers are on the right of the gel image. Asterisk indicates the slice (Slice 20) not processed for MS/MS because its major protein was anti-V5 IgG. MS/MS results were searched, and identified proteins were quantified by MaxQuant software using label-free quantification (LFQ) conducted (1) for each whole lane and (2) for individual gel slices. Abbreviations: Molecular weight, MW; mock, MK; empty vector, EV; Q152H, QH; L10insA53V, AV; R46L, RL; wild type, WT; S127R, SR; D374Y, DY; Standard, Std; Molecular weight, MW.

3.3.2. Co-immunoprecipitation of PCSK9 and Its Variants

Prior to in-gel preparations for MS/MS analyses of co-immunoprecipitated proteins shown above, I validated the immunoprecipitation of PCSK9 by immunoblotting with anti-PCSK9 as previously in materials and methods.

The representative immunoblots are as shown in Figure 16A. There is no PCSK9 detected in MK, EV and Q152H (Fig. 16A). Since the co-immunoprecipitation was conducted using anti-V5 agarose beads which should target only the transfected V5 tagged PCSK9 and not the endogenous PCSK9, the result was as expected for MK and EV (no transfected PCSK9) and Q152H (lacks processing; not secreted and remains in the ER). These serve as controls for non-specific binding to V5 agarose beads. Both the mature and furin-cleaved forms of PCSK9 were detected for A53V, R46L and WT. However, only the mature form was detected in S127R and D374Y (Fig. 16A). This is consistent with previous findings that S127R and D374Y demonstrate reduced levels of furin cleavage [45, 88].

The densitometry analyses of the immunoblots against PCSK9 showed that the levels of immuno-detected mature PCSK9 and total PCSK9 were significantly higher in A53V, R46L, WT and D374Y than S127R as assessed by ANOVA and Tukey's *post hoc* test (Fig. 16B and C). This is consistent with the PCSK9 level comparison between each of WT and variants in the secretome from ELISA and SILAC MS/MS quantification results (section 3.2.1.3, Fig. 16). In addition, the densitometry analyses of the detected furin-cleaved PCSK9/total PCSK9 showed no significant changes between A53V, R46L and WT PCSK9 (Fig. 16D).

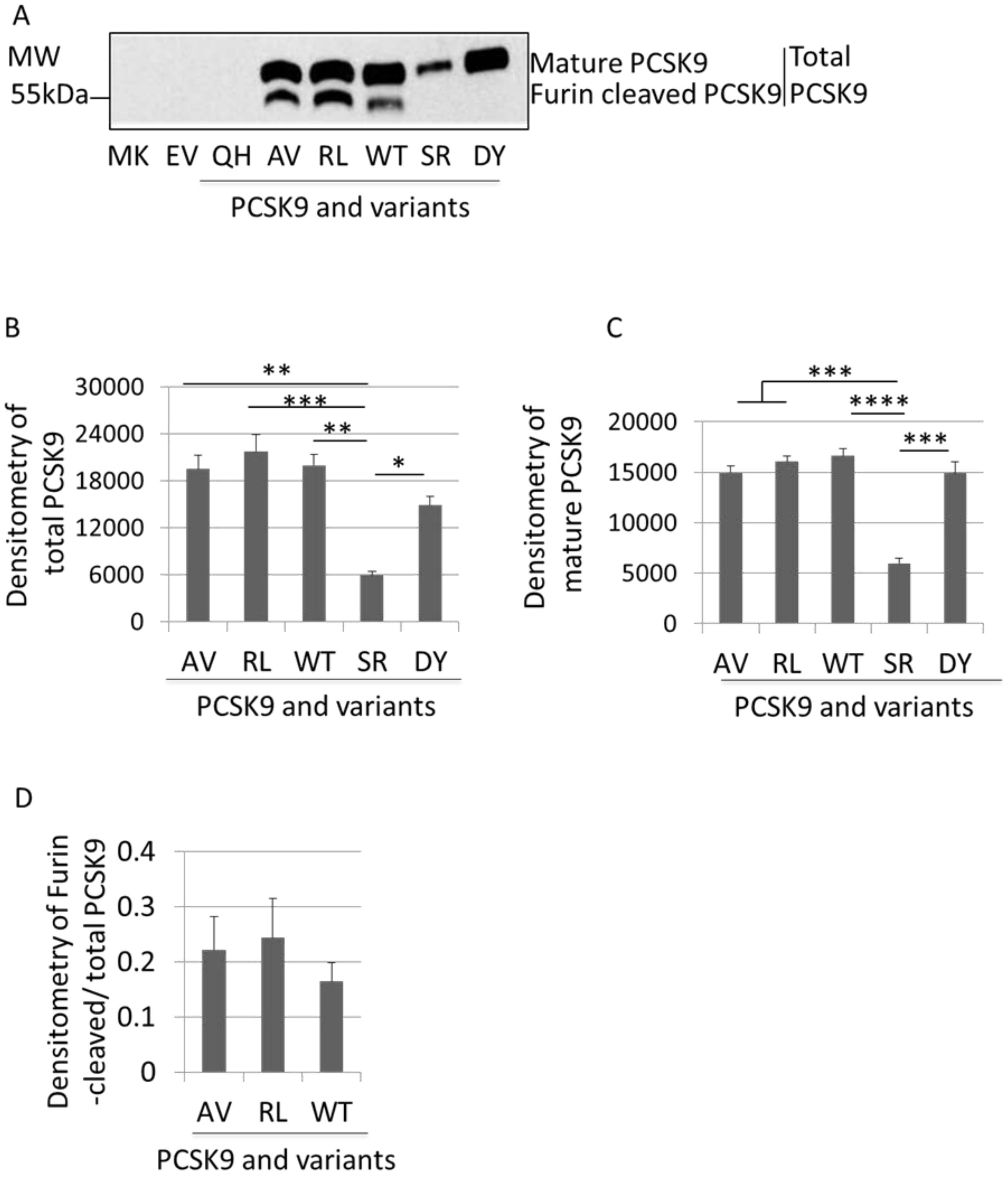

A
MW
55kDa
Mature PCSK9
Furin cleaved PCSK9
Total PCSK9
MK EV QH AV RL WT SR DY
PCSK9 and variants
B
Densitometry of total PCSK9
30000
24000
18000
12000
6000
0
**
***
**
*
AV RL WT SR DY
PCSK9 and variants
C
Densitometry of mature PCSK9
20000
15000
10000
5000
0
***
****
***
AV RL WT SR DY
PCSK9 and variants
D
Densitometry of Furin -cleaved/ total PCSK9
0.4
0.3
0.2
0.1
0
AV RL WT
PCSK9 and variants

Figure 16. Validation of PCSK9 immunoprecipitation by immunoblotting with densitometry analyses. (A). Representative immunoblot against anti-PCSK9. MK, EV and QH did not have detectable PCSK9; SR and DY did not have detectable furin-cleaved PCSK9. (B). Densitometry of total PCSK9 in AV, RL, WT, SR and DY (mature PCSK9 and furin-cleaved PCSK9). (C). Densitometry of mature PCSK9 in AV, RL, WT, SR and DY. (D). Densitometry of furin-cleaved PCSK9/total PCSK9 in AV, RL and WT. Data are presented as the means ± SEM (n=3). Significant differences were assessed by ANOVA with Tukey's *post hoc* test. Significance is indicated as *p<0.05, **p<0.005 and ***p<0.001. PCSK9 wild type (WT), Q152H (QH), L10InsA53V (AV), R46L (RL), S127R (SR), D374Y (DY), empty vector (EV) and mock (MK) transfection; Molecular weight, MW.

3.3.3. First Level Analyses of Significantly Changed Proteins Quantified by Whole Lanes and Hierarchical Clustering

From the first level of analysis by MaxQuant which was quantifying co-immunoprecitated proteins by whole lanes, the LFQ quantification identified 823 proteins. These were quantifiable with group-based values in terms that they have LFQ intensity in at least one replicate of three for each EV, WT PCSK9 and variants. MK was not considered for statistical analyses or comparisons as consistent with the secretome analyses, but the LFQ data and immunoblotting results of MK are still presented. Among the 823 proteins, 208 that showed significant changes as assessed by ANOVA were subject to hierarchical clustering by Perseus as described in materials and methods (Fig. 17A). Three major clusters were generated as shown in Fig. 17B. The protein level patterns of the three clusters were shown in Fig. 17C.

Since EV and Q152H secretomes did not have V5-tagged PCSK9, proteins in these two co-immunoprecipitates with similar intensity as WT PCSK9 and its other variants should be eliminated from potential interactors targeting. Thus proteins identified from the EV and Q152H co-immunoprecipitates were considered as the non-specific ‘control group’. Proteins identified from the co-immunoprecipitates from the secretomes of WT PCSK9 and its other variants were considered as ‘PCSK9-positive group’, where A53V (AV) and R46L (RL) were LOF group, and S127R (SR) and D374Y (DY) were GOF group. Cluster C demonstrated the interesting pattern as potential PCSK9 interactor, with lower protein levels in control group (EV and QH), higher in PCSK9-positive (PCSK9 WT, LOF or GOF) group. There were 159 proteins in the Cluster C as listed in Table 5.

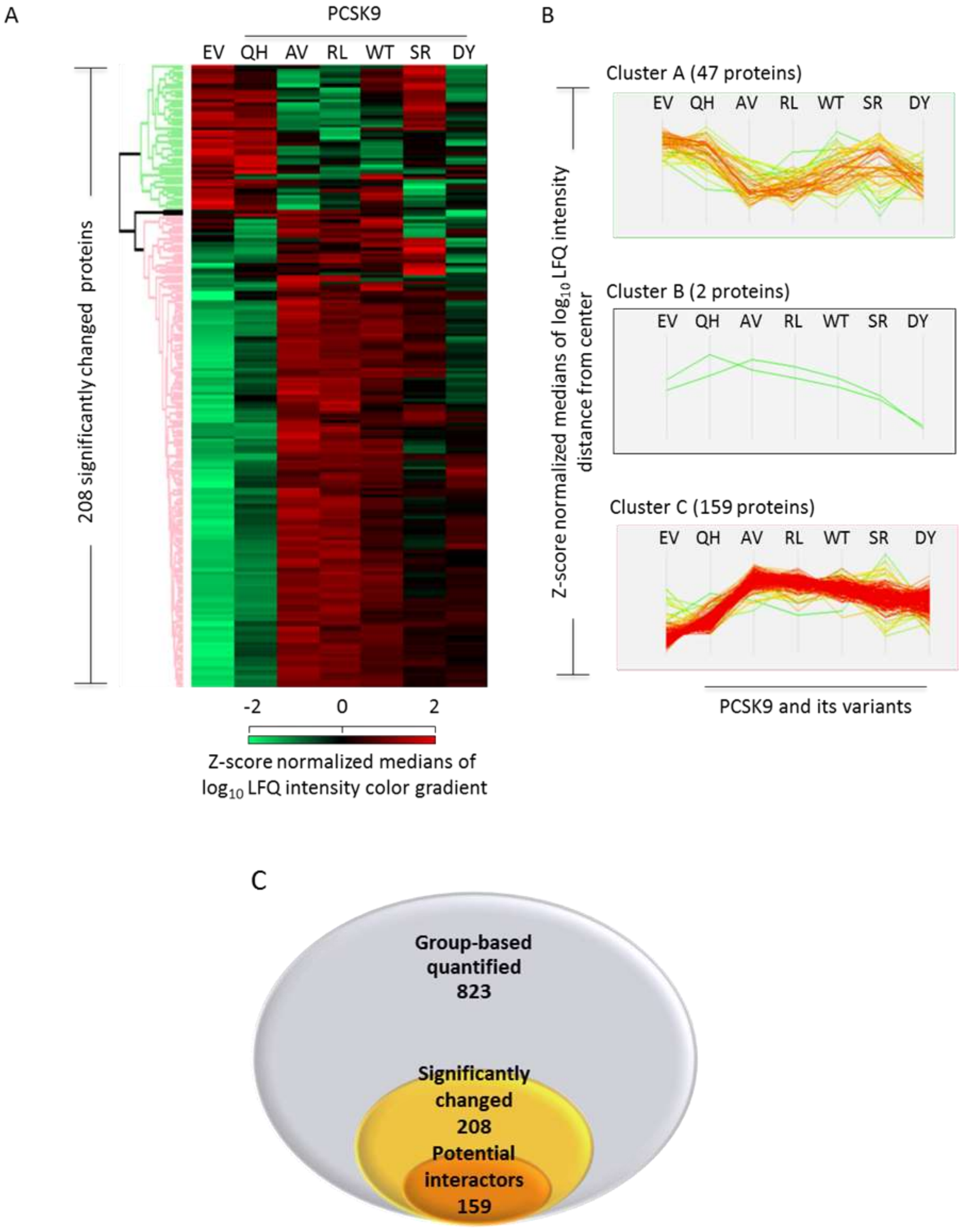

A
PCSK9
EV QH AV RL WT SR DY
208 significantly changed proteins
-2 0 2
Z-score normalized medians of $\log_{10}$ LFQ intensity color gradient
B
Cluster A (47 proteins)
EV QH AV RL WT SR DY
Cluster B (2 proteins)
EV QH AV RL WT SR DY
Cluster C (159 proteins)
EV QH AV RL WT SR DY
Z-score normalized medians of $\log_{10}$ LFQ intensity distance from center
PCSK9 and its variants
C
Group-based quantified 823
Significantly changed 208
Potential interactors 159

Figure 17. Proteins identified from the co-immunoprecipitants and their hierarchical clustering. (A). Heat map and clustering of the 208 significantly changed proteins generated by their z-score normalized medians of $\log_{10}$ LFQ intensity in EV, PCSK9 WT and variants. Color gradient is from -2 (down regulated, green) to 2 (up regulated, red). (B). Protein abundance changing patterns among EV, PCSK9 WT and variants of the three hierarchical clusters A to C. The curves were based on the distances from center after z-score normalization of $\log_{10}$ LFQ intensity medians. The dominant cluster, cluster C, demonstrated potential interactor pattern with lower abundance in control group (EV and QH) and higher level in the PCSK9-positive group (PCSK9 WT, AV, RL, SR and DY). (C). Numbers of quantified proteins by label free quantification (LFQ) with group-based values by MaxQuant (823), significantly changed assessed by ANOVA (208) and showed potential PCSK9 interactor patterns (in Cluster C, 159).

Table 5 List of potential interacting proteins of PCSK9 and its variants in Cluster C in proteomic analyses and identification of the secreted interactome in HUH7 cells

Gene Name	Protein Name	Unique Peptides	Max Fold Change
DYNC1H1	dynein, cytoplasmic 1, heavy chain 1	259	11.7
FASN	fatty acid synthase	157	6.9
PRKDC	protein kinase, DNA-activated, catalytic polypeptide	150	15.7
GCN1L1	GCN1 general control of amino-acid synthesis 1-like 1 (yeast)	103	13.0
MYH9	myosin, heavy chain 9, non-muscle	102	3.3
CLTC	clathrin, heavy chain (Hc)	100	2.2
COPA	coatomer protein complex, subunit alpha	95	7.6
SNRNP200	small nuclear ribonucleoprotein 200kDa (U5)	87	5.4
EEF2	eukaryotic translation elongation factor 2	86	1.5
EIF3A	eukaryotic translation initiation factor 3, subunit A	83	5.9
COPB1	coatomer protein complex, subunit beta 1	70	2.7
ACLY	Valyl-tRNA synthetase	67	2.8
ACACA	acetyl-CoA carboxylase alpha	66	8.2
IARS	isoleucyl-tRNA synthetase	65	5.4
EPRS	glutamyl-prolyl-tRNA synthetase	63	5.5
MTHFD1	C-1-tetrahydrofolate synthase	58	5.1
SMCHD1	structural maintenance of chromosomes flexible hinge domain containing 1	56	5.9
SMC4	structural maintenance of chromosomes 4	56	3.4
DHX9	DEAH (Asp-Glu-Ala-His) box helicase 9	55	2.8
SMC2	structural maintenance of chromosomes 2	53	4.8
LARS	leucyl-tRNA synthetase	52	4.5
PSMD2	proteasome (prosome, macropain) 26S subunit, non-ATPase, 2	51	7.0
MCM4	minichromosome maintenance complex component 4	50	5.0
COPG1	coatomer protein complex, subunit gamma 1	50	2.2
SND1	staphylococcal nuclease and tudor domain containing 1	50	2.1
KIAA0368	KIAA0368	49	5.3
HNRNPM	heterogeneous nuclear ribonucleoprotein M	48	4.4
QDPR	quinoid dihydropteridine reductase	48	3.9
XPO1	exportin 1	48	2.9
DCTN1	dynactin 1	48	2.4
PFAS	phosphoribosylformylglycinamidine synthase	47	2.3
PSMD1	proteasome (prosome, macropain) 26S subunit, non-ATPase, 1	44	6.0
MCM6	minichromosome maintenance complex component 6	44	2.5
UPF1	UPF1 regulator of nonsense transcripts homolog (yeast)	43	4.2
UBR4	ubiquitin protein ligase E3 component n-recognin 4	42	15.3
NAA15	N(alpha)-acetyltransferase 15, NatA auxiliary subunit	42	7.0
UBA1	ubiquitin-like modifier activating enzyme 1	42	2.0

IKBKAP	Elongator complex protein 1	40	11.7
COPB2	coatomer protein complex, subunit beta 2 (beta prime)	40	2.5
DIS3	DIS3 exosome endoribonuclease and 3'-5' exoribonuclease	39	2.6
DHX15	DEAH (Asp-Glu-Ala-His) box helicase 15	39	2.4
MATR3	matrin 3	38	7.3
MARS	methionyl-tRNA synthetase	37	4.3
AHCY	adenosylhomocysteinase	37	1.7
PSME4	proteasome (prosome, macropain) activator subunit 4	36	12.3
KARS	lysyl-tRNA synthetase	36	8.1
EIF3B	eukaryotic translation initiation factor 3, subunit B	36	2.4
PSMC2	proteasome (prosome, macropain) 26S subunit, ATPase, 2	35	3.4
MON2	MON2 homolog (S. cerevisiae)	35	3.2
PRPF8	pre-mRNA processing factor 8	34	4.0
KNTC1	kinetochore associated 1	34	3.0
PCSK9	proprotein convertase subtilisin/kexin type 9	33	187.7
PYGB	phosphorylase, glycogen; brain	33	2.0
PSMD6	proteasome (prosome, macropain) 26S subunit, non-ATPase, 6	32	6.7
PSMD3	proteasome (prosome, macropain) 26S subunit, non-ATPase, 3	32	6.1
PYGL	phosphorylase, glycogen, liver	32	3.9
CAD	Aspartate carbamoyltransferase	31	4.3
USP9X	ubiquitin specific peptidase 9, X-linked	30	6.7
PC	pyruvate carboxylase	30	6.4
AQR	aquarius intron-binding spliceosomal factor	30	4.9
NONO	non-POU domain containing, octamer-binding	30	2.5
CPSF1	cleavage and polyadenylation specific factor 1, 160kDa	29	7.9
RTCB	RNA 2',3'-cyclic phosphate and 5'-OH ligase	28	4.0
SRRT	serrate, RNA effector molecule	28	3.5
HTT	huntingtin	27	9.7
RUVBL1	RuvB-like AAA ATPase 1	27	7.8
MSH2	mutS homolog 2	27	5.2
BM28	ATP-citrate (pro-S-)-lyase	27	3.5
PSMC5	proteasome (prosome, macropain) 26S subunit, ATPase, 5	27	3.4
HYOU1	hypoxia up-regulated 1	27	2.3
MTTP	microsomal triglyceride transfer protein	26	5.7
RPS3	ribosomal protein S3	26	5.4
EIF4A3	eukaryotic translation initiation factor 4A3	26	3.7
POLR2B	polymerase (RNA) II (DNA directed) polypeptide B, 140kDa	26	3.7
CLUH	clustered mitochondria (cluA/CLU1) homolog	25	4.6
PSMC1	proteasome (prosome, macropain) 26S subunit, ATPase, 1	25	4.6
PSMC6	proteasome (prosome, macropain) 26S subunit, ATPase, 6	25	4.4
CTPS1	CTP synthase 1	25	4.2

RRM1	ribonucleotide reductase M1	25	4.1
HDAC2	histone deacetylase 2	25	4.1
XPOT	exportin, tRNA	25	2.6
ACO1	aconitase 1, soluble	24	7.0
DYNC1LI1	dynein, cytoplasmic 1, light intermediate chain 1	24	5.8
GNB2L1	Elongator complex protein 1	24	3.5
SRP68	signal recognition particle 68kDa	24	2.2
HMGCS1	3-hydroxy-3-methylglutaryl-CoA synthase 1 (soluble)	24	1.8
IGF2BP1	insulin-like growth factor 2 mRNA binding protein 1	23	10.7
EFTUD2	elongation factor Tu GTP binding domain containing 2	23	2.6
IGF2R	insulin-like growth factor 2 receptor	23	2.5
ATP6V1A	ATPase, H+ transporting, lysosomal 70kDa, V1 subunit A	22	5.7
FARSA	phenylalanyl-tRNA synthetase, alpha subunit	22	3.4
STRAP	serine/threonine kinase receptor associated protein	22	3.0
RPSA	ribosomal protein SA	22	2.8
STAT1	signal transducer and activator of transcription 1, 91kDa	21	4.5
SKIV2L	superkiller viralicidic activity 2-like (S. cerevisiae)	21	4.0
NANS	N-acetylneuraminic acid synthase	21	3.4
FEN1	flap structure-specific endonuclease 1	21	2.2
DARS	aspartyl-tRNA synthetase	20	5.3
POLR2A	polymerase (RNA) II (DNA directed) polypeptide A, 220kDa	20	4.2
MYBBP1A	MYB binding protein (P160) 1a	20	4.1
FAM115A	family with sequence similarity 115, member A	20	3.9
AP2A1	adaptor-related protein complex 2, alpha 1 subunit	20	2.9
AOF2	Pyridoxal-dependent decarboxylase domain-containing protein 1	19	6.9
GLDC	glycine dehydrogenase (decarboxylating)	19	6.9
VTN	vitronectin	19	1.7
CS	citrate synthase	18	7.9
MSH6	mutS homolog 6	18	6.0
SARS	seryl-tRNA synthetase	18	4.0
NSUN2	NOP2/Sun RNA methyltransferase family, member 2	18	3.2
PPAT	phosphoribosyl pyrophosphate amidotransferase	18	3.1
SEPT9	septin 9	18	3.0
EIF2S1	eukaryotic translation initiation factor 2, subunit 1 alpha, 35kDa	18	3.0
EIF4A1	eukaryotic translation initiation factor 4A1	18	1.5
XPO5	exportin 5	17	5.3
GNB1	guanine nucleotide binding protein (G protein), beta polypeptide 1	17	5.3
BUB3	BUB3 mitotic checkpoint protein	17	4.7
NADK2	NAD kinase 2, mitochondrial	16	8.5
RPS18	ribosomal protein S18	16	5.5
LIN28B	lin-28 homolog B (C. elegans)	15	4.7

EIF3I	eukaryotic translation initiation factor 3, subunit I	15	3.5
UBE4B	ubiquitination factor E4B	15	3.0
RPL5	ribosomal protein L5	14	5.4
RPS11	ribosomal protein S11	13	9.7
RAN	RAN, member RAS oncogene family	13	3.2
GALK1	galactokinase 1	13	2.6
CDK1	cyclin-dependent kinase 1	12	8.7
NQO1	NAD(P)H dehydrogenase, quinone 1	12	7.0
GMPPA	GDP-mannose pyrophosphorylase A	12	4.5
DNAJA2	DnaJ (Hsp40) homolog, subfamily A, member 2	12	2.9
TBCD	tubulin folding cofactor D	12	2.8
RPS16	ribosomal protein S16	11	6.1
OAT	ornithine aminotransferase	11	5.8
MPST	mercaptopyruvate sulfurtransferase	11	3.9
ZC3HAV1L	zinc finger CCCH-type, antiviral 1-like	11	3.3
PCYOX1	prenylcysteine oxidase 1	11	3.0
HSPA2	Nucleosome assembly protein 1-like 4b	11	3.0
NAGK	N-acetylglucosamine kinase	11	2.2
IPO8	importin 8	10	4.3
COASY	CoA synthase	10	3.8
ACTR1A	ARP1 actin-related protein 1 homolog A, centractin alpha	10	2.7
C12orf10	chromosome 12 open reading frame 10	10	2.1
VARS	Valyl-tRNA synthetase	9	12.4
KIAA0745	BRAF35-HDAC complex protein BHC110	9	5.4
HDAC6	histone deacetylase 6	9	5.1
SERPINA5	serpin peptidase inhibitor, clade A member 5	9	5.1
TNPO3	transportin 3	9	4.3
TUBB6	tubulin, beta 6 class V	9	3.4
PCOLCE2	procollagen C-endopeptidase enhancer 2	9	2.2
PPP6C	protein phosphatase 6, catalytic subunit	7	3.5
TRIM24	tripartite motif containing 24	7	2.3
HECTD3	HECT domain containing E3 ubiquitin protein ligase 3	7	1.5
TARDBP	TAR DNA binding protein	6	5.6
KIAA0251	DNA replication licensing factor MCM2	6	4.4
ARPC1B	actin related protein 2/3 complex, subunit 1B, 41kDa	6	3.9
DDX19A	DEAD (Asp-Glu-Ala-Asp) box polypeptide 19A	6	2.0
MAPK3	mitogen-activated protein kinase 3	4	2.5
GNAS	GNAS complex locus	4	2.4
ACTG1	actin, gamma 1	1	4.1
GANAB	glucosidase, alpha; neutral AB	1	4.1

Among these proteins, 23 proteins showed significant difference between control group and each one of PCSK9-positive groups as assessed by *post hoc* analyses ($p<0.05$; Table 6).

Table 6. List of potential PCSK9 interactors with significant difference between control group and each of PCSK9-positive group

Gene Names	Protein Names	Mol. Weight [kDa]	Unique Peptides	Max Fold Change to EV	Max Fold Change to QH
FASN	fatty acid synthase	273.4	157	6.9	3.5
PSME4	proteasome activator subunit 4	211.3	36	12.3	4.6
PSMD3	proteasome 26S subunit, non-ATPase, 3	61.0	32	6.1	4.8
PSMD2	proteasome 26S subunit, non-ATPase, 2	100.2	51	7.0	3.4
PRKDC	protein kinase, DNA-activated, catalytic polypeptide	469.1	150	15.7	7.5
GCN1L1	GCN1 general control of amino-acid synthesis 1-like 1	292.8	103	13.0	4.9
COPA	coatomer protein complex, subunit alpha	138.3	95	7.6	3.6
SNRNP200	small nuclear ribonucleoprotein 200kDa (U5)	244.5	87	5.4	2.3
COPB1	coatomer protein complex, subunit beta 1	107.1	70	2.7	2.2
ACACA	acetyl-CoA carboxylase alpha	270.0	66	8.2	2.9
IARS	isoleucyl-tRNA synthetase	144.5	65	5.4	2.7
SMC4	structural maintenance of chromosomes 4	147.2	56	3.4	2.7
SMC2	structural maintenance of chromosomes 2	135.7	53	4.8	3.8
VARS2	Valyl-tRNA synthetase	140.5	47	12.4	4.3
NAA15	N(alpha)-acetyltransferase 15, NatA auxiliary subunit	101.3	42	7.0	4.4
MARS	methionyl-tRNA synthetase	101.1	37	4.3	2.7
ACO1	aconitase 1, soluble	98.4	24	7.0	5.4
RPSA	ribosomal protein SA	32.9	22	2.7	2.8
SKIV2L	superkiller viralicidic activity 2-like (S. cerevisiae)	137.8	21	4.0	2.9
DDX13	Helicase-like protein	95.2	20	6.9	4.4
TNPO3	transportin 3	108.1	19	4.3	2.4
NSUN2	NOP2/Sun RNA methyltransferase family, member 2	86.5	18	3.1	3.2
HDAC6	histone deacetylase 6	131.4	9	5.1	3.4

3.3.4. Targeting Extracellular FASN and Proteasome-related Subunits as Potential PCSK9 Interactors and Their Dynamic Co-immunoprecipitation Patterns

Since for LFQ analyses, the higher abundance of proteins and larger fold changes are more accurate for statistical analysis [145], fatty acid synthase (FASN) which had largest number of unique peptides was first targeted as my primary focus for further analyses. The histogram of its $\log_{10}$ LFQ intensity quantified by whole lanes is as shown in Figure 18A. It demonstrated a dynamic pattern of FASN co-immunoprecipitation with PCSK9, namely it co-immunoprecipitated significantly less with the GOF group than the LOF group (Fig. 18A). In addition, the proteasome proteins comprised a large portion of the Cluster C with eight proteasome 26S subunits (PSMs) and one proteasome activator subunit (PSME; Table 7). Since the PSME4 and PSMD 2 and 3 were also in the list of proteins with significant difference between control group and each of PCSK9-positive group (Table 6), they were also included for further analyses. The histograms of their $\log_{10}$ LFQ intensity quantified by whole lanes are as shown in Figure 18B-D (PSME4, PSMD2 and PSMD3, respectively). PSME4 and PSMD2 showed dynamic patterns of co-immunoprecipitation with PCSK9 (Fig. 18B and C). PSME4 co-immunoprecipitated more with PCSK9 LOF group (A53V and R46L) than with PCSK9 GOF variant D374Y (Fig. 18B). PSMD2 co-immunoprecipitated significantly more with PCSK9 LOF group (A53V and R46L) than with PCSK9 GOF group (S127R and D374Y). In addition, PSMD2 was also co-immunoprecipitated significantly more with PSCK9 WT than S127R (Fig. 18C). Although not statistically significant, PSMD3 showed a slight decreasing pattern in terms of co-immunoprecipitation with PCSK9 from LOF variants to GOF variants (Fig. 18D).

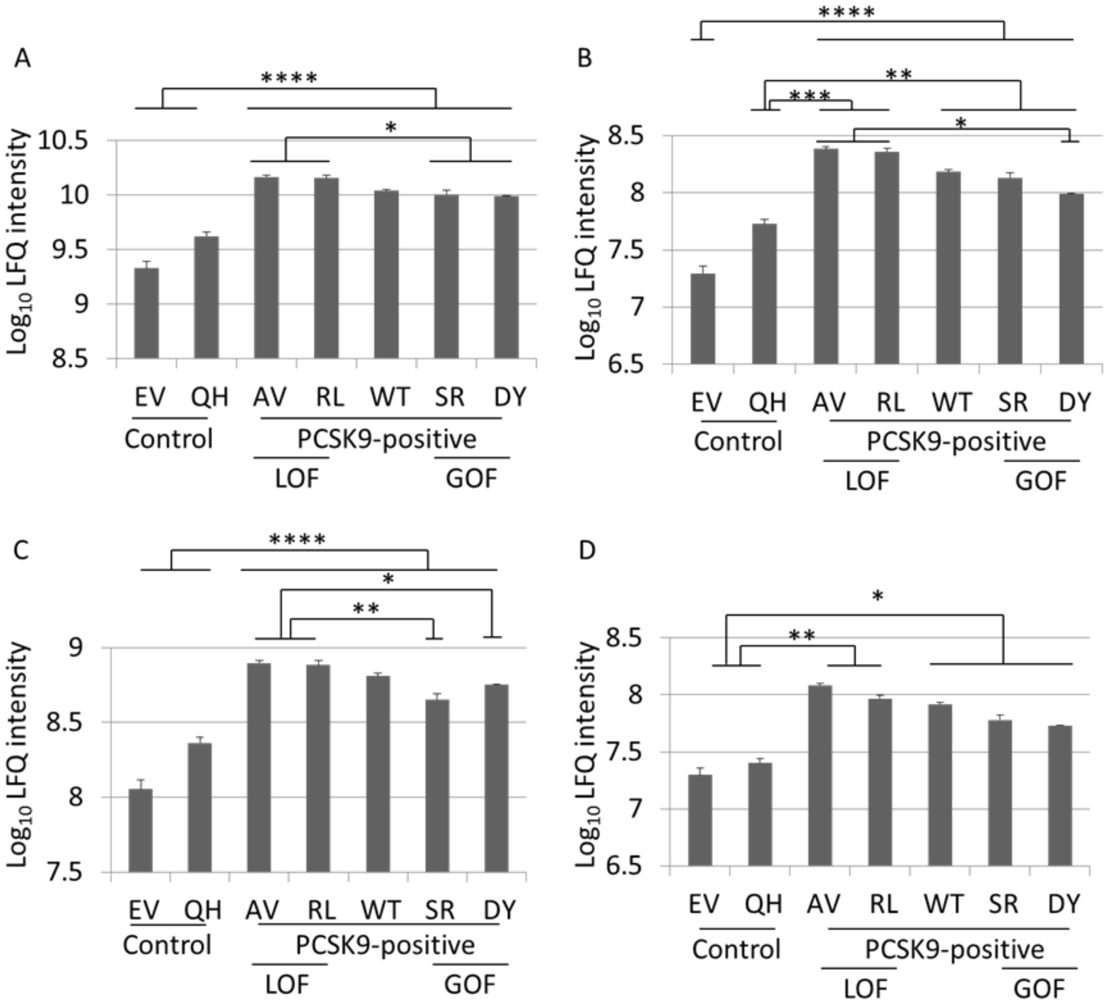

A
B
C
D
Log$_{10}$ LFQ intensity
****
**
***
*
EV
QH
AV
RL
WT
SR
DY
Control
PCSK9-positive
LOF
GOF
10.5
10
9.5
9
8.5
8
7.5
7
6.5

Figure 18. Protein abundance of V5 co-immunoprecipitated FASN, PSME4, PSMD2 and PSMD3 quantified by whole lanes. The histograms of $\log_{10}$ LFQ intensity of: (A). FASN; (B.) PSME4; (C). PSMD2; (D). PSMD3. Data are presented as medians of $\log_{10}$ LFQ intensity ± SEM, n=3). Significant changes were assessed by ANOVA and *post hoc* analyses (*$p<0.05$, **$p<0.005$ and ***$p<0.001$). Abbreviations: empty vector (EV), Q152H (QH), PCSK9 wild type (WT), L10InsA53V (AV), R46L (RL), S127R (SR) and D374Y (DY); fatty acid synthase (FASN), Proteasome activator subunit (PSME4), Proteasome 26S subunits 2 (PSMD2) and 3 (PSMD3); Loss of function (LOF), gain of function (GOF).

3.3.5. Characterization and Validation of Interaction Potential PCSK9 Interactors

3.3.5.1. Characterization of The Potential Interactors by Second Level Analyses of Cluster C Proteins Quantified by Individual Gel Slices

As the second level analyses, MaxQuant quantification by individual gel slices was carried out for the co-immuneprecipitated proteins. As previously mentioned, this second level analyses allowed me to collect information on the molecular weight and isoforms of the potential interactors to further analyze and validate their identification and interaction with PCSK9. The LFQ intensity histograms of these proteins quantified by gel slices were generated for each of EV, PCSK9 WT and variants. The LFQ intensity of proteins in MK was also included in the histograms although it was not considered for statistical analyses. The LFQ histograms of FASN, PSME4, PSMD2 and PSMD3 as I targeted for analyses are as shown in Figure 19. First, these histograms demonstrated that the SDS-PAGE fractionated apparent molecular weights of the identified PCSK9 interactors matched their known molecular weights: ~210 kDa for FASN mainly distributed in gel slices 4 and 5; ~97kDa for PSMD2 in gel slice 11; ~61kDa for PSMD3 mainly in gel slice 18 and ~211 kDa for PSME4 mainly in slice 6. Second, they further confirmed that the distribution of these co-immunoprecipitated proteins had very low abundance in the control groups compared to the PCSK9-positive groups.

Representative histograms of the top 20 proteins identified during co-immunoprecipitation with respect to the largest fold changes between the highest and lowest LFQ intensities, as well as, the highest numbers of unique peptides are shown in Appendix 4.

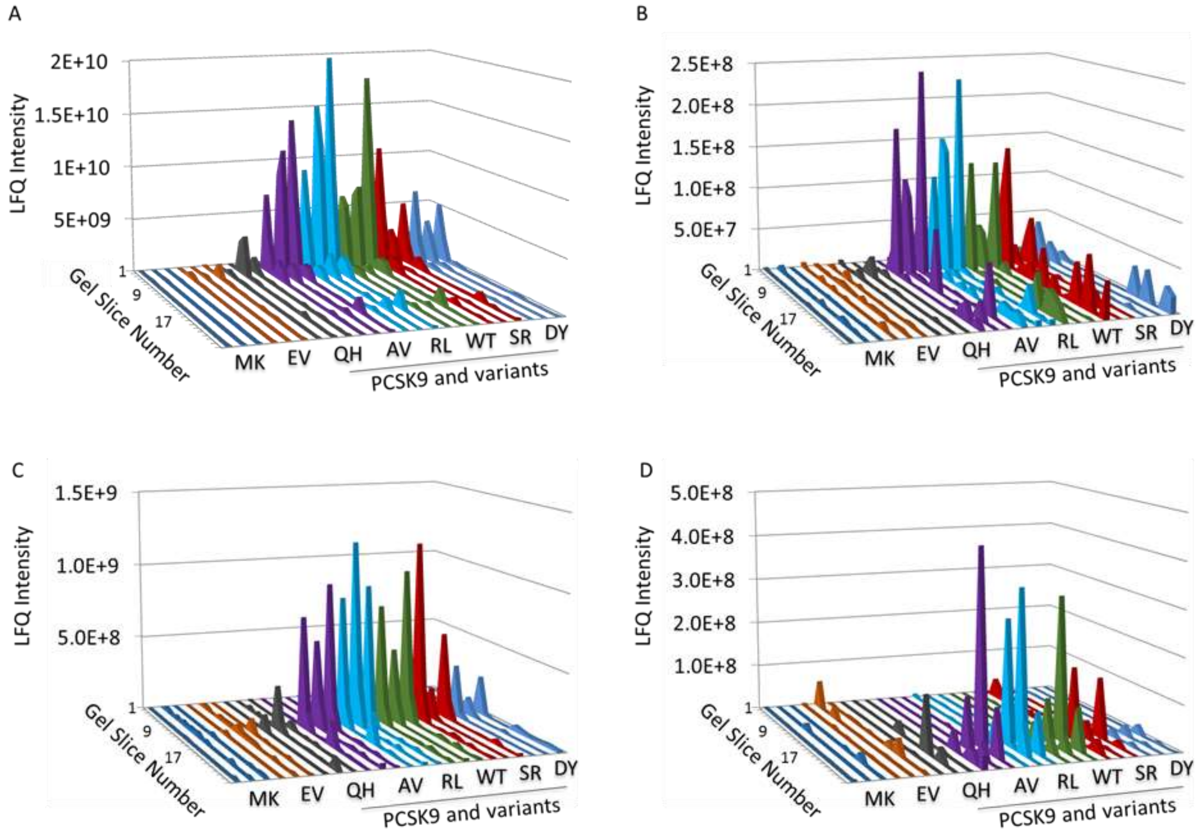

A
LFQ Intensity
2E+10
1.5E+10
1E+10
5E+09
1
9
17
Gel Slice Number
MK
EV
QH
AV
RL
WT
SR
DY
PCSK9 and variants
B
LFQ Intensity
2.5E+8
2.0E+8
1.5E+8
1.0E+8
5.0E+7
1
9
17
Gel Slice Number
MK
EV
QH
AV
RL
WT
SR
DY
PCSK9 and variants
C
LFQ Intensity
1.5E+9
1.0E+9
5.0E+8
1
9
17
Gel Slice Number
MK
EV
QH
AV
RL
WT
SR
DY
PCSK9 and variants
D
LFQ Intensity
5.0E+8
4.0E+8
3.0E+8
2.0E+8
1.0E+8
1
9
17
Gel Slice Number
MK
EV
QH
AV
RL
WT
SR
DY
PCSK9 and variants

Figure 19. The protein abundance distribution of potential PCSK9 interactors quantified by individual gel slices. LFQ intensity histograms of: (A). FASN; (B). PSME4; (C). PSMD2; D. PSMD3. Empty vector (EV), Q152H (QH), PCSK9 wild type (WT), L10InsA53V (AV), R46L (RL), S127R (SR) and D374Y (DY); fatty acid synthase (FASN), Proteasome activator subunit (PSME4), Proteasome 26S subunits 2 (PSMD2) and 3 (PSMD3); n=3.

3.3.5.1. Secretion Levels of the Potential Interactors

In order to confirm that the co-immunoprecipitation level differences of the potential PCSK9 interactors between control group and PCSK9-positive group did not result from secretion level differences (i.e. secretion level is significantly lower in EV or Q152H than PCSK9 WT) which may result in false positive results when nonspecific bound to anti-V5 beads, the protein abundance of FASN and PSMD2 in the secretomes were analyzed by SILAC quantification as shown in Figure 20A and B. There was no significant difference between control group and PCSK9-positive group as assessed by ANOVA for both FASN and PSMD2 protein abundance in the secretomes. This confirms that their co-immunoprecipitation with PCSK9-V5 was not due to a possible correlation with changes in their secretion patterns. The SILAC quantification of PSME4 and PSMD3 were not available for more than half of the replicates and therefore statistical analyses in my total secretome data set. This likely reflects the fact that these are low abundant proteins in the total secretome. Thus they were likely to co-immunoprecipitate with PCSK9 with high specificity. FASN and PSMD2 were chosen for further validation of their interaction with PCSK9.

3.3.5.2. Validation of the Potential Interactors by Immunoblotting Following V5 Co-immunoprecipitation of Secretomes from HUH7 Cells Overexpressing PCSK9 and Its Variants

To validate the PCSK9-FASN and the PCSK9-PSMD2 interactions, co-immunoprecitation of secreted PCSK9 was carried out from the secretomes of HUH7 cells transiently transfected with EV and PCSK9 Q152H (control group) as well as PCSK9 WT, A53V, R46L, S127R and D374Y (PCSK9-positive group) followed by immunoblotting against FASN and PSMD2 (Fig. 20C). MK was also included in the immunoblotting although it was

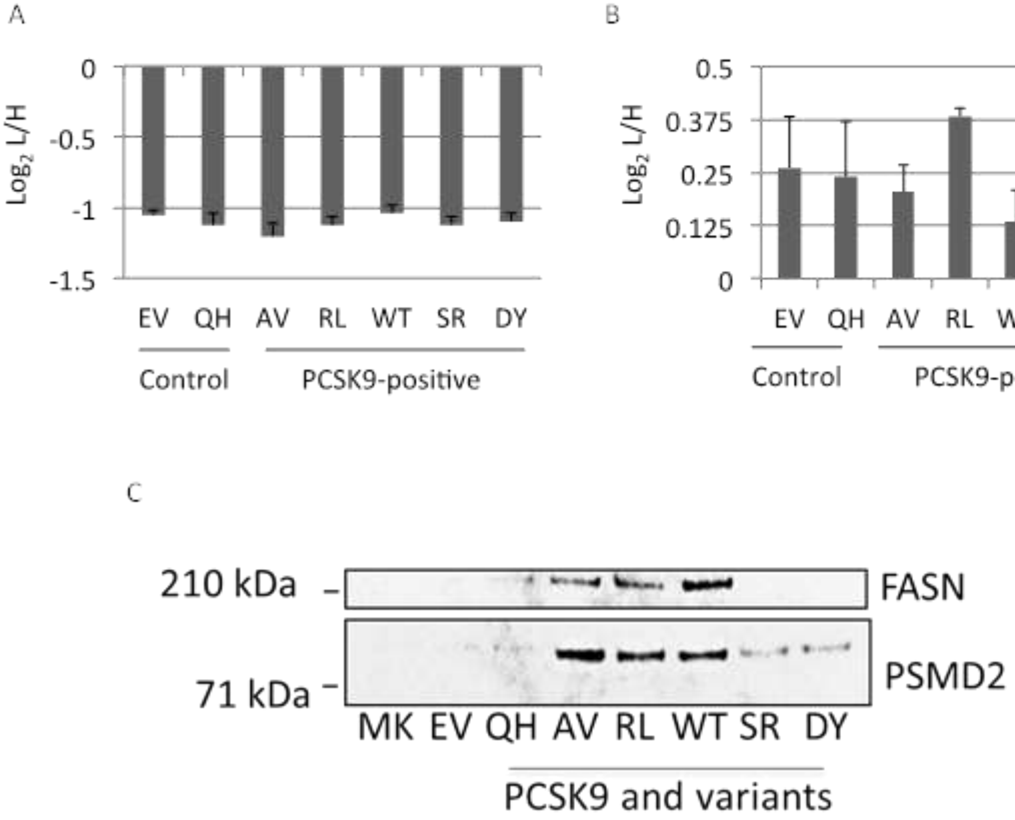

A
Log2 L/H
0
-0.5
-1
-1.5
EV
QH
AV
RL
WT
SR
DY
Control
PCSK9-positive
B
Log2 L/H
0.5
0.375
0.25
0.125
0
EV
QH
AV
RL
WT
SR
DY
Control
PCSK9-positive
C
210 kDa
71 kDa
FASN
PSMD2
MK
EV
QH
AV
RL
WT
SR
DY
PCSK9 and variants

Figure 20. Protein abundance of FASN and PSMD2 in the secretome and validation of the their interaction with PCSK9 and its variants. (A-B) Log_2 L/H ratios of FASN and PSMD2, respectively, in the secretomes of EV, PCSK9 WT and variants transfections by MS/MS SILAC quantification (medians ±SEM, n=3). (C). Representative immunoblots of FASN (upper panel) and PSMD2 (lower panel) following V5 Co-immunoprecipitation of secretomes from HUH7 cells overexpressing PCSK9 and its variants. Mock (MK), Empty vector (EV), Q152H (QH), PCSK9 wild type (WT), L10InsA53V (AV), R46L (RL), S127R (SR) and D374Y (DY); fatty acid synthase (FASN), Proteasome 26S subunits 2 (PSMD2).

not considered for analyses. FASN was detected in A53V, R46L and WT and was excluded in MK, EV and Q152H, confirming extracellular FASN is interacting with PCSK9, either directly or indirectly (Fig. 20C upper panel). It was also excluded in S127R and D374Y, with the MS/MS LFQ data that it co-immunoprecipitated significantly less with the GOF group (S127R and D374Y) than the LOF group (A53V and R46L, Fig. 18A). PSMD2 was also detected in A53V, R46L and WT and was excluded in MK, EV and Q152H, confirming its interaction with PCSK9 (Fig. 20C upper panel). In addition, it was also detected in GOF group (S127R and D374Y; in the other 2 replicates not detected) with much weaker bands compared to LOF group (A53V and R46L), further proved its dynamic interaction pattern with PCSK9 as demonstrated in the MS/MS LFQ data (Fig. 19C).

3.3.6. Discussion

To identify PCSK9 interacting proteins that may be of both high and low abundance, I carried out a non-stringent co-immunoprecipitation of V5-tagged PCSK9 from the various secretomes then I used SDS-PAGE fractionation and cut the whole lanes into gel bands for trypsin digestion. This approach (as opposed to in solution digestion) also allowed me to collect information on their molecular weight and isoforms in the secretome.

The first pass analyses identified 159 potential PCSK9 interactors of either WT or variants. Two candidates, fatty acid synthase (FASN) and proteasome 26S non-ATPase subunit 2 (PSMD2) were validated for their interaction with PCSK9.

FASN is synthesized by the liver and plays a crucial role in the fatty acid synthesis [174] and has been investigated as a possible oncogene [175]. Identification of extracellular FASN has been reported in several circumstances *in vivo* and *in vitro* and was related to hepatitis C virus (HCV) infection and human epidermal growth factor receptor 2 (HER2) shedding and signaling [176-181]. However, the mechanism regulating FASN release and the function of extracellular FASN have not been reported so far.

Proteasomes has been detected and measured in cell culture supernatants, in serum of patients with certain types of malignant diseases [182-186]. In alveolar space the extracellular proteasomes were reported to be proteolytic active in patients with acute respiratory distress syndrome [187]. As well, in healthy human cerebrospinal fluid they were also found to be proteolytic active [188]. The composition of extracellular proteasomes have been characterized by mass spectrometry that contain most of the 26S proteasome subunits including the three subunits in my dataset (PSMD2, PSMD3 and PSME4) [189]. However, functions of extracellular proteasomes especially of those expressed by liver remain unclear.

In this section, 'label-free' quantification (LFQ) was utilized for MS/MS data analyses. This method compares peptide intensities between separate LC-MS/MS runs. Compared to the SILAC ‘spiked-in’ quantification used for secretome analyses, the LFQ has certain accuracy limitations; as the samples are processed and analyzed separately, the quantification accuracy is compromised as the number of unpaired steps in the procedure increases. However, the label-free algorithms that are used in the MaxQuant software can approximate the protein ratios between samples and can be quite appropriate when large fold changes are expected [145].

4. General Discussion and Future Direction

Proprotein convertase subtilisin/kexin type 9 (PCSK9) plays an important role in mediating degradation of cell surface LDLRs through the lysosomal pathway, and its 'gain of function' and 'loss of function' variants associate with hyper- and hypo-cholesterolemia [22, 77, 190].

To date, several proteins affecting PCSK9's trafficking (such as Sec 24A and APLP2), as well as proteins being affected by PCSK9 (such as ApoB100), have been reported in the literature and associate with specific subcellular locations [91, 191]. A global study of influence between PCSK9 and other proteins on each other at a subcellular level is warranted especially with the presence of functionally different PCSK9 variants with unknown mechanisms of action. The first objective of my study was to develop methodology for identifying changes in subcellular distribution of proteins affected by PCSK9 variants. I have developed a proteomic based, quantitative method for studying the changes in subcellular distribution of proteins upon stable expression of PCSK9 LOF (Q152H) and GOF (D374Y) variants. Although this required a significant amount of work on method design, development including the optimization and normalization of the data, it could identify and quantify the subcellular distribution of 1000s of proteins in a single mass spectrometry run. Herein, I compared two experimental conditions but because the fractionations are carried out separately the number of conditions/samples that can be compared is unlimited. This is the advantage of incorporating the 'SILAC spike in' reference for quantification. Within the scope of this thesis, method development and optimization time constraints restricted my subcellular distribution studies of PCSK9 LOF versus GOF variants to a single comparison however several protein changes were observed. This method is ready for biological replication to determine statistical significance in the changes observed as well as expanding to other PCSK9 variants as part of our future directions.

As previously mentioned, PCSK9 largely functions from the ‘outside-in’ in terms of its effect on LDLR levels [23, 76-78], however the global effect of PCSK9 and its variants on the secretome has not been thoroughly studied. My second objective then was to identify changes in secretomes of cells overexpressing PCSK9 and its variants. I have demonstrated that in the secretomes from HUH7 cell lines transiently transfected with PCSK9 WT and variants the majority (90%) of the proteins were not significantly changing. Notably, I showed that the mock-transfected cell line (often used as a control for transfections) was not a suitable control as the levels of proteins it secreted in comparison to PCSK9 overexpression differed by up to 40%. However, transfections with EV were suitable controls as shown in Figure 13. The SILAC ‘spike in’ was shown as a sensitive and accurate quantification method and the relative abundance of PCSK9 quantified by this method was consistent with my ELISA results. As mentioned previously, changes in protein secretion rates and/or rates of protein endocytosis and degradation may affect the protein abundance changes observed in the secretome. In this study, several apolipoproteins showed significant difference in protein abundance between PCSK9 WT and variants, as well as proteins related to the coagulation pathway and acute phase response. The significant changes I noted above and in Table 4 may result from one or a combined effect of protein secretion, uptake and degradation. My current study does not differentiate between these possibilities. Following validation of these secretome changes by ELISA or immunoblotting, the dynamic equilibrium of the combined effect should be the major future direction for the secretome analysis. For example, the question of whether the changes of apolipoproteins resulted from direct PCSK9 regulation (that is changes in production and secretion rates) or from indirect effect through receptors (that is uptake and subsequent degradation) or from a combined effect remained to be answered. Production and secretion rates of proteins of interest should be studied by pulse-chase analysesin the presence and absence of PCSK9 variants. Alternatively, PCSK9 can be knocked-down to observe the effect of loss of PCSK9 in the pulse-chase analyses. In terms of protein of interest-receptor uptake, I can measure the protein uptake in the presence and absence of PCSK9 by immunoflurosecence or by using commercially available uptake assays when available. For example, there is a kit available to monitor ApoB100 uptake through LDLR. In cases where a particular receptor is implicated, experiments using knock-down or overexpression could be conducted to examine the effects and to determine whether the regulation of receptors contributes to the secretome protein changes. These validations and experiments would expand our knowledge of the role of PCSK9 and its particular variants. In addition, mouse models of overexpressing or knockout of *PCSK9* and its variations can be generated and used for validating the protein changes observed from my cell culture data.

Since extracellular proteins have been identified and implicated in the mechanism of PCSK9's action towards LDLR [82, 98, 99, 158], identification of novel secreted interactors of PCSK9 was the third objective of my study. I have identified a list of potential secreted PCSK9 interactors using MS/MS LFQ following co-immunoprecipitation of the secretomes from HUH7 cell lines transiently transfected with PCSK9 WT and variants (Table 7). Furthermore, two interactors, namely FASN and PSMD2, were validated by immunoblotting and demonstrated a dynamic interacting pattern with PCSK9 WT and variants. For both of these proteins, interaction was strongest for WT PCSK9 and the two LOF variants (A53V and R46L) while barely detectable for the two GOF variants (S127R and D374Y). It is unclear whether these proteins interact more readily with the furin-cleaved form (~55kDa; Fig 16) of mature form of PCSK9 (~63kDa; Fig 16) or equally with both. To determine whether the decrease in binding to SR and DY is due to lack of the furin-cleaved form, we can co-transfect a furin-containing plasmid with PCSK9, resulting in 100% secretion of the furin-cleaved form and repeat our co-IPs. Another way to determine if the interactors were interacting with both forms of PCSK9 is to inactivate the gene of furin so that all the PCSK9 and its variant remain in the mature form [88] and to examine if the dynamic interaction pattern changes for PCSK9 interactors. In addition, to further validate the co-immunoprecipitation results, reciprocal experiment of co-immunoprecipitation using antibodies against the above mentioned candidates should be carried out to see if PCSK9 can be co-precipitated. Alternatively these proteins may interact through PCSK9's prodomain. In that case, we can replace PCSK9 prodomain with a family member prodomain as a chimeric protein or co-express the prodomain with a catalytic domain in *trans* and co-immunoprecipitate the replaced or co-expressed prodomain to examine if the above mentioned candidates can co-immunoprecipitate. Furthermore, for future investigation on the effects of the observed interaction on PCSK9 and LDLR biology, studies on models with knock-down or knock out of the interacting proteins or genes should be carried out.

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Appendices

Appendix 1. List of proteins identified by MS/MS in global protein subcellular distribution analysis on a global proteomic scale

ID	Protein Names	Gene Names	SILAC Quantified	Majority Protein Uniprot IDs	Unique Peptides
1	Cytoplasmic dynein 1 heavy chain 1	DHC1	+	Q14204	251
2	Desmoyokin	AHNAK	+	Q09666	220
3	DNA-dependent protein kinase catalytic subunit	HYRC	+	P78527	177
4	fatty acid synthase	FASN	+	P49327	128
5	Bifunctional aminoacyl-tRNA synthetase	EPRS	+	P07814	99
6	eIF3 p167	EIF3A	+	Q14152	94
7	Cellular myosin heavy chain, type A	MYH9	+	P35579	93
8	600 kDa retinoblastoma protein-associated factor	KIAA0462	+	Q5T4S7-2	92
9	Clathrin heavy chain 1	CLH17	+	Q00610	85
10	GCN1-like protein 1	GCN1L1	+	Q92616	83
11	220 kDa U5 snRNP-specific protein	PRPC8	+	Q6P2Q9	79
12	Colonic and hepatic tumor over-expressed gene protein	CKAP5	+	Q14008-3	75
13	Actin-binding protein 280	FLN	+	P21333	73
14	High density lipoprotein-binding protein	HBP	+	Q00341	71
15	Nuclear mitotic apparatus protein 1	NUMA	+	Q14980	70
16	Aspartate carbamoyltransferase	CAD	+	P27708	69
17	Activating signal cointegrator 1 complex subunit 3-like 1	ASCC3L1	+	O75643	68
18	ATP-dependent RNA helicase A	DDX9	+	Q08211	67
19	250/210 kDa paraneoplastic pemphigus antigen	DSP	+	P15924	64
20	Talin-1	KIAA1027	+	Q9Y490	63
21	Isoleucine--tRNA ligase	IARS	+	P41252	62
22	180 kDa ribosome receptor homolog	KIAA1398	+	Q9P2E9	61
23	Proteasome-associated protein ECM29 homolog	ECM29	+	Q5VYK3	61
24	Heterogeneous nuclear ribonucleoprotein M	HNRNPM	+	P52272	60
25	15S Mg(2+)-ATPase p97 subunit	VCP	+	P55072	59
26	Elongation factor 2	EEF2	+	P13639	57
27	Nucleolin	NCL	+	P19338	57
28	NAD(+) ADP-ribosyltransferase 1	ADPRT	+	P09874	55
29	Leucine--tRNA ligase	KIAA1352	+	Q9P2J5	54
30	BAT2 domain-containing protein 1	BAT2D1	+	E7EPN9	53
31	Chromosome-associated polypeptide C	CAPC	+	Q9NTJ3	53

Continuing Appendix 1. List of proteins identified by MS/MS in global protein subcellular distribution analysis on a global proteomic scale

32	Arginine--tRNA ligase	RARS	+	P54136	52
33	Vimentin	VIM	+	P08670	52
34	26S proteasome non-ATPase regulatory subunit 1	PSMD1	+	Q99460	51
35	26S proteasome non-ATPase regulatory subunit 2	PSMD2	+	Q13200	51
36	Alpha-coat protein	COPA	+	P53621	51
37	Glutamine--tRNA ligase	QARS	+	P47897	51
38	Microtubule-associated protein 4	MAP4	+	E7EVA0	51
39	60S ribosomal protein L1	RPL1	+	P36578	50
40	CCT-theta	C21orf112	+	P50990	50
41	86 kDa subunit of Ku antigen	G22P2	+	P13010	49
42	CCT-beta	99D8.1	+	P78371	49
43	Hemidesmosomal protein 1	PLEC1	+	Q15149	49
44	Pre-mRNA-splicing factor SF3b 155 kDa subunit	SAP155	+	O75533	49
45	Sb1.8	DXS423E	+	Q14683	49
46	150 kDa dynein-associated polypeptide	DCTN1	+	Q14203	48
47	CCT-eta	CCT7	+	Q99832	48
48	CCR4-associated factor 1	AD-005	+	A5YKK6	46
49	CCT-epsilon	CCT5	+	P48643	46
50	Chromosome-associated protein E	CAPE	+	O95347	46
51	Nestin	Nbla00170	+	P48681	46
52	ATP-dependent RNA helicase DDX3X	DBX	+	O00571	45
53	70 kDa subunit of Ku antigen	G22P1	+	P12956	44
54	CCT-gamma	CCT3	+	P49368	44
55	KRAB-associated protein 1	KAP1	+	Q13263	44
56	Activating signal cointegrator 1 complex subunit 3	ASCC3	+	Q8N3C0	43
57	ATP-dependent helicase RENT1	KIAA0221	+	Q92900	43
58	MAP1 light chain LC1	MAP1B	+	P46821	43
59	60S ribosomal protein L3	OK/SW-cl.32	+	P39023	42
60	100 kDa coactivator	SND1	+	Q7KZF4	41
61	26S proteasome non-ATPase regulatory subunit 3	PSMD3	+	O43242	41
62	620 kDa actin-binding protein	ABP620	+	H3BQK9	41
63	ARF-binding protein 1	HSPC272	+	Q7Z6Z7	41
64	Eukaryotic translation initiation factor 5B	EIF5B	+	O60841	41
65	Alpha-II spectrin	SPTA2	+	Q13813	40
66	CDC46 homolog	CDC46	+	P33992	40
67	CXXC-type zinc finger protein 9	AIM	+	F5GX68	40
68	40S ribosomal protein S4, X isoform	CCG2	+	P62701	39
69	Aspartate--tRNA ligase	DARS	+	P14868	39
70	Cold shock domain-containing protein E1	CSDE1	+	G5E9Q2	39

Continuing Appendix 1. List of proteins identified by MS/MS in global protein subcellular distribution analysis on a global proteomic scale

71	DNA polymerase alpha holoenzyme-associated protein P1	MCM3	+	P25205	39
72	Moloney leukemia virus 10 protein	KIAA1631	+	Q9HCE1	39
73	100 kDa DNA-pairing protein	PSF	+	P23246	38
74	Basement membrane-associated chondroitin proteoglycan	BAM	+	Q9UQE7	38
75	Coding region determinant-binding protein	CRDBP	+	Q9NZI8	38
76	Death-associated protein 5	DAP5	+	P78344	38
77	Methionine--tRNA ligase	MARS	+	P56192	38
78	Pre-mRNA-splicing factor SF3b 145 kDa subunit	SAP145	+	Q13435	38
79	Valyl-tRNA synthetase	DAAP-21F2.2-001	+	B0V043	38
80	116 kDa U5 small nuclear ribonucleoprotein component	EFTUD2	+	Q15029	37
81	26S protease regulatory subunit 7	MSS1	+	P35998	37
82	4-alpha-glucanotransferase	AGL	+	P35573	37
83	54 kDa nuclear RNA- and DNA-binding protein	NONO	+	Q15233	37
84	Autoantigen Ge-1	EDC4	+	Q6P2E9	37
85	CCT-delta	CCT4	+	P50991	37
86	Cell proliferation-inducing gene 46 protein	CYK18	+	P05783	37
87	Cullin-associated and neddylation-dissociated protein 1	CAND1	+	Q86VP6	37
88	eIF3 p110	EIF3B	+	P55884-2	37
89	Lysine--tRNA ligase	KARS	+	Q15046	37
90	ATP-binding cassette 50	ABC50	+	Q8NE71	36
91	ATP-dependent helicase CHD4	CHD4	+	Q14839-2	36
92	Cell cycle protein p38-2G4 homolog	EBP1	+	Q9UQ80	36
93	Deleted in breast cancer gene 1 protein	DBC1	+	Q8N163	36
94	La ribonucleoprotein domain family member 1	KIAA0731	+	Q6PKG0	36
95	60S ribosomal protein L5	MSTP030	+	P46777	35
96	Beta-coat protein	COPB	+	P53618	35
97	Constitutive coactivator of PPAR-gamma-like protein 1	C9orf10	+	Q9NZB2-6	35
98	eIF3 p110	EIF3C	+	Q99613	35
99	Hsc70/Hsp90-organizing protein	STIP1	+	P31948	35
100	26S protease regulatory subunit 4	PSMC1	+	P62191	34
101	300 kDa nuclear matrix antigen	HSPC075	+	Q9UQ35	34
102	23 kDa highly basic protein	RPL13A	+	P40429	33
103	40S ribosomal protein S3	OK/SW-cl.26	+	P23396	33
104	Cell division cycle 5-like protein	CDC5L	+	Q99459	33
105	Eukaryotic translation initiation factor 2 subunit 2	EIF2B	+	P20042	33
106	Far upstream element-binding protein 2	FUBP2	+	Q92945	33

Continuing Appendix 1. List of proteins identified by MS/MS in global protein subcellular distribution analysis on a global proteomic scale

107	60S ribosomal protein L6	RPL6	+	Q02878	32
108	Cell proliferation-inducing gene 54 protein	KIAA0648	+	Q29RF7	32
109	Cellular apoptosis susceptibility protein	CAS	+	P55060	32
110	Conventional kinesin heavy chain	KIF5B	+	P33176	32
111	Cytosolic thyroid hormone-binding protein	OIP3	+	P14618	32
112	Damage-specific DNA-binding protein 1	DDB1	+	Q16531	32
113	DNA replication licensing factor MCM2	BM28	+	F5H1E9	32
114	DNA replication licensing factor MCM6	MCM6	+	Q14566	32
115	Echinoderm microtubule-associated protein-like 4	C2orf2	+	Q9HC35	32
116	Importin subunit beta-1	KPNB1	+	Q14974	32
117	Pre-mRNA-splicing factor SF3b 130 kDa subunit	KIAA0017	+	Q15393	32
118	26S protease regulatory subunit 6B	MIP224	+	P43686	31
119	26S proteasome non-ATPase regulatory subunit 11	PSMD11	+	O00231	31
120	78 kDa glucose-regulated protein	GRP78	+	P11021	31
121	ATP-dependent RNA helicase DDX1	DDX1	+	Q92499	31
122	CDC47 homolog	CDC47	+	P33993	31
123	Chromosome condensation-related SMC-associated protein 1	CAPD2	+	Q15021	31
124	DEAD box protein 46	DDX46	+	Q7L014	31
125	Eukaryotic translation initiation factor 2 subunit 1	EIF2A	+	P05198	31
126	26S proteasome non-ATPase regulatory subunit 12	PSMD12	+	O00232	30
127	60S ribosomal protein L7a	RPL7A	+	P62424	30
128	ACC-alpha	ACAC	+	Q13085-4	30
129	Acute morphine dependence-related protein 2	CCT6	+	P40227	30
130	Annexin A2	ANX2	+	P07355-2	30
131	C-1-tetrahydrofolate synthase, cytoplasmic	MTHFC	+	F5H2F4	30
132	CCT-alpha	CCT1	+	P17987	30
133	Eukaryotic translation initiation factor 2 subunit 3	EIF2G	+	P41091	30
134	5-3 exoribonuclease 2	XRN2	+	Q9H0D6	29
135	60S ribosomal protein L7	RPL7	+	P18124	29
136	ATP-dependent RNA helicase #46	DBP1	+	O43143	29
137	DEAD box protein 5	DDX5	+	P17844	29
138	Deubiquitinating enzyme 7	HAUSP	+	Q93009	29
139	eEF-1B gamma	EEF1G	+	P26641	29
140	Gem-associated protein 5	GEMIN5	+	Q8TEQ6	29
141	HLA-B-associated transcript 2	BAT2	+	P48634	29
142	IMPDH-II	IMPD2	+	P12268	29
143	Polyadenylate-binding protein 1	PAB1	+	P11940	29
144	Pre-mRNA-processing factor 6	C20orf14	+	O94906	29

Continuing Appendix 1. List of proteins identified by MS/MS in global protein subcellular distribution analysis on a global proteomic scale

145	Putative uncharacterized protein MATR3	MATR3	+	A8MXP9	29
146	Tight junction protein 2	TJP2	+	Q9UDY2	29
147	Zinc finger antiviral protein	PRO1677	+	Q7Z2W4	29
148	182 kDa tankyrase-1-binding protein	KIAA1741	+	Q9C0C2	28
149	26S protease regulatory subunit 10B	PSMC6	+	P62333	28
150	26S protease regulatory subunit 8	PSMC5	+	P62195	28
151	300 kDa mannose 6-phosphate receptor	IGF2R	+	P11717	28
152	Adenovirus early region 1B-associated protein 5	E1BAP5	+	Q9BUJ2	28
153	Asparagine--tRNA ligase	ASNS	+	O43776	28
154	Coatomer subunit gamma	COPG	+	Q9Y678	28
155	E3 ubiquitin/ISG15 ligase TRIM25	EFP	+	Q14258	28
156	Elongator complex protein 1	ELP1	+	O95163	28
157	p90	TFRC	+	P02786	28
158	100 kDa U5 snRNP-specific protein	DDX23	+	Q9BUQ8	27
159	40S ribosomal protein S2	RPS2	+	P15880	27
160	40S ribosomal protein S6	OK/SW-cl.2	+	P62753	27
161	60S ribosomal protein L8	RPL8	+	P62917	27
162	cDNA FLJ61021, highly similar to Far upstream element-binding protein 1	FUBP1	+	B4E0X8	27
163	DEAD box polypeptide 17 isoform p82 variant	DDX17	+	Q59F66	27
164	eIF-3 p48	EIF3E	+	P60228	27
165	Eukaryotic translation initiation factor 3, subunit E interacting protein	AL022311.1-001	+	B0QY89	27
166	26S proteasome non-ATPase regulatory subunit 13	PSMD13	+	Q9UNM6	26
167	60S ribosomal protein L13	BBC1	+	P26373	26
168	60S ribosomal protein L18a	RPL18A	+	Q02543	26
169	Ataxin-2 domain protein	A2D	+	Q8WWM7	26
170	ATP-dependent helicase SKIV2L2	KIAA0052	+	P42285	26
171	Beta-coat protein	COPB2	+	P35606	26
172	CDC21 homolog	CDC21	+	P33991	26
173	Cellular thyroid hormone-binding protein	ERBA2L	+	P07237	26
174	Far upstream element-binding protein 3	FBP3	+	Q96I24	26
175	Rho/rac guanine nucleotide exchange factor (GEF) 2	ARHGEF2	+	Q5VY93	26
176	Ribonucleases P/MRP protein subunit POP1	KIAA0061	+	Q99575	26
177	SF3a120	SAP114	+	Q15459	26
178	40S ribosomal protein S7	RPS7	+	P62081	25
179	48 kDa TATA box-binding protein-interacting protein	CGI-46	+	Q9Y230	25
180	94 kDa glucose-regulated protein	GRP94	+	P14625	25
181	Adenylyl cyclase-associated protein 1	CAP	+	Q01518	25

Continuing Appendix 1. List of proteins identified by MS/MS in global protein subcellular distribution analysis on a global proteomic scale

182	Archain 1, isoform CRA_a	ARCN1	+	B0YIW6	25
183	Cytovillin	EZR	+	P15311	25
184	Importin subunit beta-3	IPO5	+	O00410-3	25
185	Leucine-rich repeat-containing protein 47	KIAA1185	+	Q8N1G4	25
186	Radixin isoform b	hCG_39182	+	A7YIJ8	25
187	Tripeptidyl aminopeptidase	TPP2	+	P29144	25
188	UDP-glucose 6-dehydrogenase	UGDH	+	O60701	25
189	26S protease regulatory subunit 6A	PSMC3	+	P17980	24
190	26S proteasome non-ATPase regulatory subunit 6	KIAA0107	+	Q15008	24
191	40S ribosomal protein S11	RPS11	+	P62280	24
192	40S ribosomal protein S9	RPS9	+	P46781	24
193	ATP-binding cassette sub-family F member 2	ABCF2	+	Q9UG63	24
194	ATP-dependent DNA helicase Q1	RECQ1	+	P46063	24
195	DNA topoisomerase 1	TOP1	+	P11387	24
196	DNA-directed RNA polymerase II subunit A	POLR2	+	P24928	24
197	eIF3 p66	EIF3D	+	O15371	24
198	Heat shock 86 kDa	HSP90A	+	P07900-2	24
199	IGF-II mRNA-binding protein 3	IGF2BP3	+	O00425	24
200	Kinesin-2	KIF2	+	O00139-2	24
201	50 kDa dynein-associated polypeptide	DCTN2	+	F5H2S7	23
202	Caprin-1	CAPRIN1	+	Q14444	23
203	DEAH box protein 30	DDX30	+	Q7L2E3	23
204	Deubiquitinating enzyme 14	TGT	+	P54578	23
205	Deubiquitinating enzyme FAF-X	DFFRX	+	Q93008	23
206	Diaphanous-related formin-1	DIAP1	+	H9KV28	23
207	Gastric cancer antigen Ga19	GA19	+	Q9BXJ9	23
208	Heterogeneous nuclear ribonucleoprotein U-like protein 2	HNRNPUL2	+	Q1KMD3	23
209	SNU66 homolog	SART1	+	O43290	23
210	TATA box-binding protein-associated factor 2S	CA150	+	O14776	23
211	2-5-oligoadenylate-binding protein	ABCE1	+	P61221	22
212	40S ribosomal protein S16	RPS16	+	P62249	22
213	40S ribosomal protein S18	D6S218E	+	P62269	22
214	49 kDa TATA box-binding protein-interacting protein	INO80H	+	Q9Y265	22
215	63 kDa membrane protein	CKAP4	+	Q07065	22
216	65 kDa eukaryotic translation initiation factor 2A	CDA02	+	Q9BY44	22
217	Adapter-related protein complex 3 subunit beta-1	ADTB3A	+	O00203	22
218	AIR carboxylase	ADE2	+	P22234	22
219	Cell proliferation-inducing gene 19 protein	LDHA	+	P00338-3	22

Continuing Appendix 1. List of proteins identified by MS/MS in global protein subcellular distribution analysis on a global proteomic scale

220	Cellular myosin heavy chain, type B	MYH10	+	F8VTL3	22
221	Chromosome region maintenance 1 protein homolog	CRM1	+	O14980	22
222	CLASP1 protein	CLASP1	+	B7ZLX3	22
223	CLIP-associating protein 2	CLASP2	+	F5H604	22
224	DNA-directed RNA polymerase II 140 kDa polypeptide	POLR2B	+	P30876	22
225	Eukaryotic translation initiation factor 4B	EIF4B	+	E7EX17	22
226	Exosome complex exonuclease RRP44	DIS3	+	Q9Y2L1	22
227	Glycoprotein p43	HNRPG	+	P38159	22
228	GRB10-interacting GYF protein 2	GIGYF2	+	Q6Y7W6	22
229	Heat shock 70 kDa protein 1/2	HSPA1	+	H0YG33	22
230	Heterogeneous nuclear ribonucleoproteins A2/B1	HNRNPA2B1	+	P22626	22
231	La autoantigen	SSB	+	P05455	22
232	Membrane-organizing extension spike protein	MSN	+	P26038	22
233	Metastasis-associated 1-like 1	MTA1L1	+	O94776	22
234	Protein NICE-4	KIAA0144	+	Q14157-5	22
235	Protein RCC2	KIAA1470	+	Q9P258	22
236	RRP12-like protein	KIAA0690	+	Q5JTH9	22
237	Signal recognition particle 68 kDa protein	SRP68	+	Q9UHB9	22
238	UPF0027 protein C22orf28	C22orf28	+	Q9Y3I0	22
239	136 kDa double-stranded RNA-binding protein	ADAR	+	E7ENU4	21
240	40S ribosomal protein S8	OK/SW-cl.83	+	P62241	21
241	60S ribosomal protein L14	RPL14	+	P50914	21
242	60S ribosomal protein L24	RPL24	+	P83731	21
243	D-3-phosphoglycerate dehydrogenase	PGDH3	+	O43175	21
244	Deubiquitinating enzyme 10	KIAA0190	+	Q14694-2	21
245	Developmentally-regulated GTP-binding protein 1	DRG1	+	Q9Y295	21
246	D-fructose-6-phosphate amidotransferase 1	GFAT	+	Q06210	21
247	DNase IV	FEN1	+	P39748	21
248	E3 ubiquitin-protein ligase UBR5	EDD	+	O95071	21
249	Glyceraldehyde-3-phosphate dehydrogenase	CDABP0047	+	P04406	21
250	Glycylpeptide N-tetradecanoyltransferase 1	NMT	+	P30419	21
251	Heat shock 84 kDa	HSP90AB1	+	P08238	21
252	Heterogeneous nuclear ribonucleoprotein L	HNRNPL	+	P14866	21
253	Nuclear matrix protein 200	NMP200	+	Q9UMS4	21
254	Pericentriolar material 1 protein	PCM1	+	Q15154	21
255	Pre-mRNA-splicing factor 3	HPRP3	+	O43395	21
256	U1 small nuclear ribonucleoprotein 70 kDa	RNPU1Z	+	P08621	21
257	40S ribosomal protein S10	RPS10	+	P46783	20
258	40S ribosomal protein S3a	FTE1	+	P61247	20

Continuing Appendix 1. List of proteins identified by MS/MS in global protein subcellular distribution analysis on a global proteomic scale

259	60S ribosomal protein L10a	NEDD6	+	P62906	20
260	60S ribosomal protein L15	EC45	+	P61313	20
261	60S ribosomal protein L18	RPL18	+	Q07020	20
262	82 kDa FMRP-interacting protein	KIAA1321	+	Q7Z417	20
263	Activated-platelet protein 1	APP1	+	Q13310-3	20
264	Adapter-related protein complex 3 subunit delta-1	AP3D1	+	O14617-5	20
265	ATP-dependent RNA helicase p54	DDX6	+	P26196	20
266	Autoantigen PM/Scl 2	EXOSC10	+	Q01780	20
267	cDNA FLJ53542, highly similar to Heterogeneous nuclear ribonucleoproteins C	HNRNPC	+	P07910-2	20
268	DEAH box protein 36	DDX36	+	Q9H2U1	20
269	Eukaryotic translation initiation factor 2-alpha kinase 2	EIF2AK2	+	P19525	20
270	FK506-binding protein 12-rapamycin complex-associated protein 1	FRAP	+	P42345	20
271	Fragile X mental retardation syndrome-related protein 1	FXR1	+	P51114	20
272	Fructose-bisphosphate aldolase A	ALDA	+	P04075	20
273	Squamous cell carcinoma antigen recognized by T-cells 3	KIAA0156	+	Q15020	20
274	Transketolase	TKT	+	E7EPA7	20
275	Zinc finger protein 598	ZNF598	+	Q86UK7	20
276	Breast cancer-associated gene 1 protein	BCG1	+	Q9UNF1	19
277	30 kDa prosomal protein	HC2	+	P25786-2	19
278	40S ribosomal protein S19	RPS19	+	P39019	19
279	450 kDa epidermal antigen	EPIPL	+	P58107	19
280	57 kDa RNA-binding protein PPTB-1	PTB	+	P26599-3	19
281	60S ribosomal protein L17	RPL17	+	P18621	19
282	60S ribosomal protein L19	RPL19	+	P84098	19
283	Adenosylhomocysteinase	AHCY	+	P23526	19
284	Alternative-splicing factor 1	ASF	+	Q07955	19
285	Arsenite-resistance protein 2	ARS2	+	Q9BXP5	19
286	cDNA FLJ51909, highly similar to Serine-threonine kinase receptor-associatedprotein	MAWD	+	B4DNJ6	19
287	Cytoplasmic dynein 1 light intermediate chain 1	DNCLI1	+	Q9Y6G9	19
288	ELAV-like protein 1	ELAVL1	+	Q15717	19
289	Hepatocellular carcinoma autoantigen p62	IGF2BP2	+	F8W930	19
290	Lymphoid-specific helicase	HELLS	+	Q9NRZ9	19
291	Macropain subunit C7-I	PSMB2	+	P49721	19
292	Medium tumor antigen-associated 61 kDa protein	PPP2R1A	+	P30153	19
293	Proteasome activator complex subunit 4	KIAA0077	+	Q14997	19
294	Protein flightless-1 homolog	FLII	+	Q13045	19

Continuing Appendix 1. List of proteins identified by MS/MS in global protein subcellular distribution analysis on a global proteomic scale

295	2-5-oligoadenylate synthase 3	OAS3	+	Q9Y6K5	18
296	40S ribosomal protein S13	RPS13	+	P62277	18
297	40S ribosomal protein S5	RPS5	+	P46782	18
298	60S ribosomal protein L21	RPL21	+	P46778	18
299	60S ribosomal protein L28	RPL28	+	P46779	18
300	60S ribosomal protein L32	PP9932	+	F8W727	18
301	6-phosphofructokinase type C	PFKF	+	Q01813	18
302	AKAP-Lbc	AKAP13	+	Q12802-2	18
303	Alpha-taxilin	TXLN	+	P40222	18
304	APEX nuclease	APE	+	P27695	18
305	ATP-dependent RNA helicase DDX48	DDX48	+	P38919	18
306	ATP-dependent RNA helicase DHX29	DDX29	+	Q7Z478	18
307	CCAAT-binding transcription factor I subunit A	NSEP1	+	P67809	18
308	cDNA FLJ59751, weakly similar to Mus musculus spermatogenesis associated, serine-rich 2 (Spats2), mRNA	DNAPTP6	+	B4DT67	18
309	Elongation factor 1-alpha 1	EEF1A	+	P68104	18
310	Glycogen [starch] synthase, muscle	GYS	+	P13807	18
311	Inactive ubiquitin-specific peptidase 39	CGI-21	+	Q53GS9	18
312	Interleukin enhancer-binding factor 2	ILF2	+	Q12905	18
313	Macropain subunit C8	HC8	+	P25788	18
314	p195	IQGAP1	+	P46940	18
315	Proteasome subunit alpha type-7	HSPC	+	O14818	18
316	Splicing factor U2AF 65 kDa subunit	U2AF2	+	P26368	18
317	Tetratricopeptide repeat protein 37	KIAA0372	+	Q6PGP7	18
318	Threonine--tRNA ligase	TARS	+	P26639	18
319	Zinc finger CCCH domain-containing protein 4	C19orf7	+	Q9UPT8	18
320	25 kDa FK506-binding protein	FKBP25	+	Q00688	17
321	26S proteasome non-ATPase regulatory subunit 8	PSMD8	+	P48556	17
322	27 kDa prosomal protein	PROS27	+	G3V5Z7	17
323	40S ribosomal protein S17	RPS17	+	P08708	17
324	60S ribosomal protein L23a	RPL23A	+	P62750	17
325	60S ribosomal protein L9	OK/SW-cl.103	+	P32969	17
326	Activator 1 37 kDa subunit	RFC4	+	P35249	17
327	Aminoacyl tRNA synthase complex-interacting multifunctional protein 1	AIMP1	+	Q12904-2	17
328	ATP-dependent DNA helicase VIII	G3BP	+	Q13283	17
329	Bcl-2-associated transcription factor 1	BCLAF1	+	Q9NYF8	17
330	Cofactor of BRCA1	COBRA1	+	Q8WX92	17

Continuing Appendix 1. List of proteins identified by MS/MS in global protein subcellular distribution analysis on a global proteomic scale

331	Component of gems 3	DDX20	+	Q9UHI6	17
332	DEAD box protein 21	DDX21	+	Q9NR30	17
333	DEAD box protein 41	ABS	+	Q9UJV9	17
334	eIF3 p40 subunit	EIF3H	+	O15372	17
335	Eukaryotic translation initiation factor 3 subunit M	EIF3M	+	Q7L2H7	17
336	Gamma-ring complex protein 103 kDa	GCP2	+	F5H4L0	17
337	Helicase SKI2W	DDX13	+	Q15477	17
338	Initiation factor 2-associated 67 kDa glycoprotein	METAP2	+	P50579	17
339	MIDAS-containing protein	KIAA0301	+	Q9NU22	17
340	Myb-binding protein 1A	MYBBP1A	+	Q9BQG0-2	17
341	Natural killer cell-enhancing factor A	PAGA	+	Q06830	17
342	NOL1/NOP2/Sun domain family member 2	NSUN2	+	Q08J23	17
343	PAT1-like protein 1	OK/KNS-cl.5	+	Q86TB9-4	17
344	Probable ATP-dependent RNA helicase YTHDC2	YTHDC2	+	Q9H6S0	17
345	Sodium pump subunit alpha-1	ATP1A1	+	F5H3A1	17
346	TRIP12 protein	TRIP12	+	Q14CA3	17
347	Tyrosyl--tRNA ligase	YARS	+	P54577	17
348	UPF0568 protein C14orf166	C14orf166	+	Q9Y224	17
349	40S ribosomal protein S25	RPS25	+	P62851	16
350	60 kDa poly(U)-binding-splicing factor	FIR	+	Q9UHX1-2	16
351	6-phosphofructokinase, muscle type	PFKM	+	P08237	16
352	Activator 1 140 kDa subunit	RFC1	+	P35251	16
353	Activator 1 38 kDa subunit	RFC3	+	P40938	16
354	Aldehyde dehydrogenase family 1 member A1	ALDC	+	P00352	16
355	Basic leucine zipper and W2 domain-containing protein 1	BZAP45	+	Q7L1Q6-3	16
356	Chromosome-associated protein G	CAPG	+	Q9BPX3	16
357	Cleavage and polyadenylation specificity factor 68 kDa subunit	CFIM68	+	Q16630-2	16
358	Component of gems 4	GEMIN4	+	P57678	16
359	DNA mismatch repair protein Msh2	MSH2	+	P43246	16
360	Dnm1p/Vps1p-like protein	DLP1	+	O00429-6	16
361	DRG family-regulatory protein 1	DFRP1	+	Q8WU90	16
362	ELKS/Rab6-interacting/CAST family member 1	ELKS	+	Q8IUD2	16
363	GAP SH3 domain-binding protein 2	G3BP2	+	Q9UN86	16
364	Heterogeneous nuclear ribonucleoprotein R	HNRNPR	+	O43390-2	16
365	Importin subunit alpha-2	KPNA2	+	P52292	16
366	Negative elongation factor A	NELFA	+	Q9H3P2	16
367	Pescadillo homolog	PES1	+	O00541	16

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368	Phenylalanine--tRNA ligase beta chain	FARSB	+	Q9NSD9	16
369	Protein A1S9	A1S9T	+	P22314	16
370	Thyroid hormone receptor-associated protein 3	THRAP3	+	Q9Y2W1	16
371	U2 small nuclear ribonucleoprotein A	SNRPA1	+	P09661	16
372	140 kDa Ser/Arg-rich domain protein	KIAA0332	+	O15042	15
373	32 kDa thioredoxin-related protein	TRP32	+	O43396	15
374	40S ribosomal protein S12	RPS12	+	P25398	15
375	60S ribosomal protein L27	RPL27	+	P61353	15
376	Aldehyde dehydrogenase family 16 member A1	ALDH16A1	+	Q8IZ83	15
377	Antiapoptosis clone 11 protein	API5	+	Q9BZZ5-2	15
378	APOBEC1-binding protein 1	ABBP1	+	Q99729-3	15
379	Baculoviral IAP repeat-containing protein 6	BIRC6	+	Q9NR09	15
380	Bystin	BYSL	+	Q13895	15
381	Cadherin-associated Src substrate	CTNND1	+	O60716-3	15
382	cAMP regulatory element-associated protein 1	CREAP1	+	O95232	15
383	CBF5 homolog	DKC1	+	O60832	15
384	cDNA FLJ46199 fis, clone TESTI4007965, highly similar to AP-1 complex subunit gamma-1	ADTG	+	B3KXW5	15
385	cDNA FLJ53647, highly similar to Four and a half LIM domains protein 1	FHL1	+	Q13642-5	15
386	Death-associated transcription factor 1	C20orf158	+	Q9BTC0	15
387	DNA repair protein RAD50	RAD50	+	Q92878-2	15
388	DnaJ homolog subfamily A member 1	DNAJ2	+	P31689	15
389	eIF-2B GDP-GTP exchange factor subunit beta	EIF2B2	+	P49770	15
390	eIF-2B GDP-GTP exchange factor subunit epsilon	EIF2B5	+	Q13144	15
391	Eukaryotic translation initiation factor 5	EIF5	+	P55010	15
392	High mobility group protein 2	HMG2	+	P26583	15
393	Leucine zipper protein 1	LUZP1	+	Q86V48	15
394	Macropain subunit C9	HC9	+	P25789	15
395	mRNA export factor TAP	NXF1	+	Q9UBU9	15
396	Npw38-binding protein	NPWBP	+	Q9Y2W2	15
397	Paraspeckle component 1	PSP1	+	Q8WXF1	15
398	PHD finger protein 6	KIAA1823	+	E9PC97	15
399	Protein lin-28 homolog B	CSDD2	+	Q6ZN17	15
400	Signal recognition particle 72 kDa protein	SRP72	+	O76094	15
401	37 kDa laminin receptor precursor	LAMBR	+	P08865	14
402	60S ribosomal protein L12	RPL12	+	P30050	14
403	72 kDa ICln-binding protein	HRMT1L5	+	O14744	14
404	Actin-like protein 3	ACTR3	+	P61158	14

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405	Aminoacyl tRNA synthase complex-interacting multifunctional protein 2	AIMP2	+	Q13155	14
406	Arginine/proline-rich coiled-coil domain-containing protein 1	KIAA1187	+	Q3KQU3	14
407	Ataxin-2	ATX2	+	Q99700	14
408	ATP-dependent RNA helicase #3	DBP2	+	O60231	14
409	Barren homolog protein 1	BRRN	+	Q15003	14
410	CATX-11	CATX11	+	O76021	14
411	cDNA FLJ36192 fis, clone TESTI2027450, highly similar to Eukaryotic translation initiation factor 3 subunit 5	hCG_1784554	+	B3KSH1	14
412	cDNA FLJ56180, highly similar to Negative elongation factor E	NELFE	+	B4DYX9	14
413	Centrosomal protein of 170 kDa	CEP170	+	Q5SW79	14
414	DEAD box protein 47	DDX47	+	Q9H0S4	14
415	DnaJ homolog subfamily C member 2	DNAJC2	+	Q99543	14
416	Dolichyl-diphosphooligosaccharide--protein glycosyltransferase 67 kDa subunit	RPN1	+	P04843	14
417	Dynamin-2	DNM2	+	P50570	14
418	eIF3 p42	EIF3G	+	O75821	14
419	GC-binding factor 2	GCF2	+	Q32MZ4-3	14
420	Heat shock 70 kDa protein 4	APG2	+	P34932	14
421	Heterochromatin protein 1-binding protein 3	HP1BP3	+	Q5SSJ5	14
422	Heterogeneous nuclear ribonucleoprotein A0	HNRNPA0	+	Q13151	14
423	Importin-7	IPO7	+	O95373	14
424	LDH heart subunit	LDHB	+	P07195	14
425	MORC family CW-type zinc finger protein 2	KIAA0852	+	Q9Y6X9	14
426	Nucleoprotein TPR	TPR	+	P12270	14
427	Protein LTV1 homolog	C6orf93	+	Q96GA3	14
428	Protein pelota homolog	CGI-17	+	Q9BRX2	14
429	SF3a60	SAP61	+	Q12874	14
430	UDP-glucose pyrophosphorylase	UGP1	+	Q16851	14
431	14-3-3 protein zeta/delta	YWHAZ	+	E7EX29	13
432	18S rRNA dimethylase	DIMT1	+	Q9UNQ2	13
433	26S proteasome non-ATPase regulatory subunit 14	POH1	+	O00487	13
434	40S ribosomal protein S20	RPS20	+	P60866	13
435	46 kDa DNA polymerase delta interaction protein	KIAA1649	+	Q9BY77	13
436	47 kDa mannose 6-phosphate receptor-binding protein	M6PRBP1	+	O60664	13
437	60 kDa chaperonin	HSP60	+	P10809	13
438	60S ribosomal protein L17	RPL23	+	P62829	13
439	60S ribosomal protein L35a	GIG33	+	P18077	13

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440	ABP125	HSPC275	+	D6REX3	13
441	ABT1-associated protein	ABTAP	+	Q9H501	13
442	Alpha-adducin	ADD1	+	P35611-3	13
443	Androgen-induced proliferation inhibitor	APRIN	+	Q9NTI5	13
444	Antigen NY-CO-25	HSP105	+	Q92598	13
445	APC-binding protein EB1	MAPRE1	+	Q15691	13
446	Astrin	SPAG5	+	Q96R06	13
447	Condensin-2 complex subunit D3	CAPD3	+	P42695	13
448	Cytoplasmic dynein 1 intermediate chain 2	DNCI2	+	Q13409-2	13
449	eIF3 p36	EIF3I	+	Q13347	13
450	eIF-4D	EIF5A	+	I3L504	13
451	ELKL motif kinase 1	EMK1	+	Q7KZI7	13
452	Elongation factor 1-beta	EEF1B	+	P24534	13
453	Fanconi anemia group I protein	FANCI	+	Q9NVI1	13
454	GTP-binding protein 1	GTPBP1	+	O00178	13
455	Hepatocellular carcinoma protein 1	HCC1	+	Q14498	13
456	High mobility group protein 1	HMG1	+	P09429	13
457	HLA-B-associated transcript 2-like 1	BAT2L	+	Q5JSZ5	13
458	Inosine-5-monophosphate dehydrogenase	IMPDH1	+	C9JV30	13
459	Interferon receptor 1-bound protein 4	HMT2	+	G5E9B6	13
460	La ribonucleoprotein domain family member 4	LARP4	+	Q71RC2-4	13
461	Long-chain acyl-CoA synthetase 4	ACS4	+	O60488	13
462	M-phase phosphoprotein homolog	ZFR	+	Q96KR1	13
463	Neural precursor cell expressed developmentally down-regulated protein 5	DIFF6	+	Q15019-2	13
464	Paraspeckle protein 2	RBM14	+	Q96PK6	13
465	Pre-mRNA-processing factor 31	PRP31	+	Q8WWY3	13
466	Putative RNA-binding protein Luc7-like 2	CGI-59	+	Q9Y383	13
467	Putative uncharacterized protein GATAD2A	GATAD2A	+	B5MC40	13
468	Small nuclear ribonucleoprotein Sm D2	SNRPD1	+	P62316	13
469	Smu-1 suppressor of mec-8 and unc-52 protein homolog	SMU1	+	Q2TAY7	13
470	26S proteasome non-ATPase regulatory subunit 7	MOV34L	+	P51665	12
471	358 kDa nucleoporin	NUP358	+	P49792	12
472	60S acidic ribosomal protein P2	D11S2243E	+	P05387	12
473	60S ribosomal protein L27a	RPL27A	+	P46776	12
474	60S ribosomal protein L35	RPL35	+	P42766	12
475	60S ribosomal protein L36	RPL36	+	Q9Y3U8	12
476	80 kDa nuclear cap-binding protein	CBP80	+	Q09161	12

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477	ABP-278	FLN1L	+	O75369-8	12
478	Alpha-CP1	PCBP1	+	Q15365	12
479	Autoantigen p542	HNRPCL2	+	Q9UKM9	12
480	Breast carcinoma-amplified sequence 2	BCAS2	+	O75934	12
481	cAMP-dependent protein kinase type II-alpha regulatory subunit	PKR2	+	P13861	12
482	Chromosome-associated protein G2	LUZP5	+	Q86XI2-2	12
483	Chronic renal failure gene protein	CRFG	+	Q9BZE4	12
484	CLL-associated antigen KW-14	HGRG8	+	Q9Y5A9	12
485	CML33	FARS	+	Q9Y285	12
486	Coiled-coil domain-containing protein 124	CCDC124	+	Q96CT7	12
487	Collapsin response mediator protein 2	CRMP2	+	Q16555	12
488	Cyclophilin B	CYPB	+	P23284	12
489	DEAH box protein 57	DHX57	+	Q6P158	12
490	Dedicator of cytokinesis protein 7	DOCK7	+	Q96N67	12
491	DNA mismatch repair protein Msh6	GTBP	+	P52701	12
492	EBI3-associated protein of 60 kDa	ORCA	+	Q13501	12
493	eIF-2B GDP-GTP exchange factor subunit gamma	EIF2B3	+	Q9NR50	12
494	Endoplasmic reticulum resident protein 70	ERP70	+	P13667	12
495	eNOS-interacting protein	CGI-25	+	Q9Y314	12
496	Erythrocyte 65 kDa protein	KDP	+	F5H2M7	12
497	Eukaryotic peptide chain release factor GTP-binding subunit ERF3A	ERF3A	+	P15170-2	12
498	Exportin-5	KIAA1291	+	Q9HAV4	12
499	Fas ligand-associated factor 1	FBP11	+	O75400	12
500	G patch domain and KOW motifs-containing protein	GPATC5	+	Q92917	12
501	General transcription factor 3C polypeptide 1	GTF3C1	+	Q12789	12
502	Hepatocellular carcinoma-associated antigen 90	HCA90	+	Q96RR5	12
503	Heterogeneous nuclear ribonucleoprotein F	HNRNPF	+	P52597	12
504	Histone deacetylase complex subunit Sin3a	SIN3A	+	Q96ST3	12
505	HLA-B-associated transcript 3	BAT3	+	G3V1Z5	12
506	Importin-4	IMP4B	+	Q8TEX9-2	12
507	LIM and SH3 domain protein 1	LASP1	+	Q14847	12
508	Mammalian branch point-binding protein	SF1	+	Q15637	12
509	Mitotic checkpoint protein BUB3	BUB3	+	O43684	12
510	Myotubularin-related protein 5	MTMR5	+	H7BXU8	12
511	NOP2 protein	NOP2	+	Q3KQS4	12
512	Phosphorylation regulatory protein HP-10	ART4	+	Q9ULX3	12
513	Pre-mRNA-splicing factor SRP55	SFRS6	+	Q13247	12

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514	Protein transport protein Sec23A	SEC23A	+	Q15436	12
515	PRP4 homolog	PRP4	+	O43172	12
516	Ran GTPase-activating protein 1	KIAA1835	+	P46060	12
517	RNA-binding motif protein 27	KIAA1311	+	Q9P2N5	12
518	Serine/arginine-rich protein-specific kinase 1	SRPK1	+	Q96SB4	12
519	Synaptic vesicle membrane protein VAT-1 homolog	VAT1	+	Q99536	12
520	14-3-3 protein epsilon	YWHAE	+	P62258	11
521	28 kDa heat- and acid-stable phosphoprotein	HASPP28	+	Q13442	11
522	34 kDa nucleolar scleroderma antigen	FBL	+	P22087	11
523	40S ribosomal protein S14	PRO2640	+	P62263	11
524	41 kDa phosphoribosypyrophosphate synthetase-associated protein	PRPSAP2	+	O60256	11
525	4F2 cell-surface antigen heavy chain	MDU1	+	P08195-4	11
526	58 kDa glucose-regulated protein	ERP57	+	P30101	11
527	60S ribosomal protein L30	RPL30	+	P62888	11
528	75 kDa DNA-pairing protein	FUS	+	H3BPE7	11
529	Actin-binding protein anillin	ANLN	+	Q9NQW6	11
530	Activator 1 40 kDa subunit	RFC2	+	P35250	11
531	Adapter-related protein complex 3 mu-1 subunit	AP3M1	+	Q9Y2T2	11
532	Adaptor protein complex AP-1 mu-1 subunit	AP1M1	+	Q9BXS5-2	11
533	Adipophilin	ADFP	+	Q99541	11
534	Alpha-actinin-4	ACTN4	+	O43707	11
535	ATP-dependent RNA helicase DDX42	DDX42	+	Q86XP3	11
536	Autoantigen NGP-1	GNL2	+	Q13823	11
537	BMP2-induced 3-kb gene protein	BIG3	+	P61964	11
538	BPY2-interacting protein 1	BPY2IP1	+	Q66K74	11
539	Calcium pump 2	ATP2A2	+	P16615	11
540	Capping protein (Actin filament) muscle Z-line, beta	CAPZB	+	E7EQ40	11
541	Clathrin light chain B	CLTB	+	P09497	11
542	Cleavage and polyadenylation specificity factor 160 kDa subunit	CPSF1	+	Q10570	11
543	Cleavage and polyadenylation specificity factor 59 kDa subunit	CPSF7	+	Q8N684-3	11
544	Coiled-coil domain-containing protein 86	CCDC86	+	Q9H6F5	11
545	Cyclophilin A	CYPA	+	P62937	11
546	DnaJ homolog subfamily C member 9	DNAJC9	+	Q8WXX5	11
547	E2-induced gene 3 protein	E2IG3	+	Q9BVP2	11
548	Elongator complex protein 3	ELP3	+	Q9H9T3	11
549	Endothelial differentiation-related factor 1	EDF1	+	O60869	11
550	Eukaryotic translation initiation factor 2C 2	AGO2	+	Q9UKV8	11

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551	Eukaryotic translation initiation factor 4H	EIF4H	+	Q15056	11
552	Fragile X mental retardation syndrome-related protein 2	FMR1L2	+	P51116	11
553	Glycogen phosphorylase, brain form	PYGB	+	P11216	11
554	HCG2045902, isoform CRA_b	hCG_2045902	+	Q8IWZ2	11
555	Kinesin-like protein 6	KIF2C	+	Q99661	11
556	Macropain chain Z	PSMB7	+	Q99436	11
557	Macropain subunit C3	HC3	+	P25787	11
558	Macropain subunit C5	PSC5	+	P20618	11
559	Modulator of non-genomic activity of estrogen receptor	HMX3	+	I3L3A8	11
560	mRNA turnover protein 4 homolog	C1orf33	+	Q9UKD2	11
561	N-acetyltransferase 10	ALP	+	Q9H0A0	11
562	Natural killer cell-enhancing factor B	NKEFB	+	P32119	11
563	Neurofibromatosis-related protein NF-1	NF1	+	P21359	11
564	Nucleolar protein 56	NOL5A	+	O00567	11
565	O-GlcNAc transferase subunit p110	OGT	+	O15294	11
566	Protein ftsJ homolog 3	FTSJ3	+	Q8IY81	11
567	Pseudouridylate synthase 7 homolog	KIAA1897	+	Q96PZ0	11
568	Putative uncharacterized protein RAN	hCG_1744585	+	B5MDF5	11
569	RNA polymerase II-associated protein 3	RPAP3	+	Q9H6T3	11
570	Sec1 family domain-containing protein 1	C14orf163	+	Q8WVM8	11
571	Vacuolar ATPase isoform VA68	ATP6A1	+	P38606	11
572	Zinc finger CCHC domain-containing protein 3	C20orf99	+	Q9NUD5	11
573	110 kDa cell membrane glycoprotein	ADRM1	+	Q16186	10
574	18 kDa Alu RNA-binding protein	SRP14	+	P37108	10
575	26S protease subunit S5 basic	KIAA0072	+	Q16401	10
576	40S ribosomal protein S23	RPS23	+	P62266	10
577	40S ribosomal protein S24	RPS24	+	E7EPK6	10
578	59 kDa serine/threonine-protein kinase	ILK	+	Q13418	10
579	60S ribosomal protein L31	RPL31	+	P62899	10
580	6-phosphofructokinase, liver type	PFKL	+	P17858	10
581	Activator 1 36 kDa subunit	RFC5	+	P40937	10
582	Alanine--tRNA ligase	AARS	+	P49588	10
583	ALG-2-interacting protein 1	AIP1	+	E9PFU1	10
584	Alpha E-catenin	CTNNA1	+	P35221-2	10
585	Apoptotic chromatin condensation inducer in the nucleus	ACIN1	+	Q9UKV3	10
586	ATP-dependent RNA helicase eIF4A-1	DDX2A	+	P60842	10

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587	Calcyclin-binding protein	CACYBP	+	Q9HB71	10
588	CAP-Gly domain-containing linker protein 2	CLIP2	+	F5GYW3	10
589	CDC10 protein homolog	CDC10	+	Q16181	10
590	cDNA FLJ41124 fis, clone BRACE2014850, highly similar to Small nuclear ribonucleoprotein-associated protein N	COD	+	P14678-3	10
591	Cell division control protein 2 homolog	CDC2	+	P06493	10
592	Clathrin light chain A	CLTA	+	P09496	10
593	Coatomer subunit epsilon	COPE	+	O14579	10
594	COP9 signalosome complex subunit 4	COPS4	+	D6RAX7	10
595	CSNK2A1 protein	CSNK2A1	+	Q5U5J2	10
596	Cytokine-induced protein of 29 kDa	HCC1	+	P82979	10
597	DEAD box protein 27	DDX27	+	Q96GQ7	10
598	Delayed-early protein HRS	HRS	+	Q13243	10
599	Dendritic cell-derived IFNG-induced protein	MOP5	+	Q9Y3Z3	10
600	DNA polymerase	POLA1	+	A6NMQ1	10
601	Dynactin subunit 4	DCTN4	+	Q9UJW0	10
602	eIF3 p35	EIF3J	+	O75822	10
603	Enhancer of rudimentary homolog	ERH	+	P84090	10
604	ERFS	HBS1	+	Q9Y450	10
605	Exportin-7	KIAA0745	+	E9PEN8	10
606	Fortilin	TPT1	+	P13693	10
607	Gamma-1-tubulin	TUBG	+	P23258	10
608	Gamma-ring complex protein 104 kDa	GCP3	+	Q96CW5	10
609	General transcription factor 3C polypeptide 5	CDABP0017	+	Q9Y5Q8-3	10
610	Glutamate-rich WD repeat-containing protein 1	GRWD	+	Q9BQ67	10
611	Guanine nucleotide-binding protein-like 3-like protein	GNL3L	+	Q9NVN8	10
612	Heat shock 70 kDa protein 14	HSP60	+	Q0VDF9	10
613	Heat shock 70 kDa protein 8	HSC70	+	P11142	10
614	Helix-destabilizing protein	HNRNPA1	+	P09651	10
615	High mobility group protein 2a	HMG2A	+	O15347	10
616	Histone H1x	H1FX	+	Q92522	10
617	HIV Tat-specific factor 1	HTATSF1	+	O43719	10
618	Kinesin-like protein 2	HSET	+	Q9BW19	10
619	Leucine-rich repeat-containing protein 40	LRRC40	+	Q9H9A6	10
620	Macropain zeta chain	PSMA5	+	P28066	10
621	Mineral dust-induced gene protein	MDIG	+	Q8IUF8	10
622	N-ethylmaleimide-sensitive fusion protein	NSF	+	P46459	10
623	Phosphodeoxyriboaldolase	CGI-26	+	Q9Y315	10

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624	Proteasome chain 13	PSMB3	+	P49720	10
625	Putative uncharacterized protein MLLT4	MLLT4	+	P55196-5	10
626	Ras-related protein Rab-14	RAB14	+	P61106	10
627	Ras-related protein Rab-7a	RAB7	+	P51149	10
628	Replication factor A protein 1	REPA1	+	P27694	10
629	Ribonucleoside-diphosphate reductase large subunit	RR1	+	P23921	10
630	Ribosome maturation protein SBDS	CGI-97	+	Q9Y3A5	10
631	Serine/arginine-rich protein-specific kinase 2	SRPK2	+	P78362-2	10
632	Seryl-tRNA synthetase	RP11-352P4.2-004	+	Q5T5C7	10
633	SF3a66	SAP62	+	Q15428	10
634	Signal recognition particle 54 kDa protein	SRP54	+	P61011	10
635	Smoothelin	SMSMO	+	P53814-6	10
636	Splicing factor 9G8	SFRS7	+	Q16629	10
637	TESS	TES	+	H0Y2M4	10
638	Transcription elongation factor A protein 1	GTF2S	+	P23193	10
639	Triosephosphate isomerase	TPI	+	P60174	10
640	TTF-I-interacting peptide 20	ZNF787	+	Q6DD87	10
641	U1 small nuclear ribonucleoprotein A	SNRPA	+	P09012	10
642	Ubiquitin-protein ligase E3C	KIAA0010	+	Q15386	10
643	Zinc finger CCCH domain-containing protein 11A	KIAA0663	+	O75152	10
644	11S regulator complex subunit gamma	PSME3	+	P61289-2	9
645	14-3-3 protein tau	YWHAQ	+	P27348	9
646	230 kDa bullous pemphigoid antigen	BPAG1	+	Q03001	9
647	26 kDa prosomal protein	PROS26	+	P28070	9
648	28 kDa heat shock protein	HSP27	+	P04792	9
649	40S ribosomal protein S28	RPS28	+	P62857	9
650	5-3 exoribonuclease 1	01-Sep	+	Q8IZH2	9
651	56 kDa U2AF65-associated protein	BAT1	+	F8VQ10	9
652	60S ribosomal protein L22	RPL22	+	P35268	9
653	60S ribosomal protein L37a	RPL37A	+	P61513	9
654	70 kDa lamin	LMN1	+	P02545	9
655	80K-H protein	G19P1	+	P14314	9
656	Actin-related protein 10	ACTR10	+	Q9NZ32	9
657	Activated RNA polymerase II transcriptional coactivator p15	PC4	+	P53999	9
658	Activating signal cointegrator 1 complex subunit 2	ASC1P100	+	Q9H1I8	9
659	Activating signal cointegrator 1	TRIP4	+	Q15650	9
660	Alpha-1-fetoprotein	AFP	+	P02771	9

Continuing Appendix 1. List of proteins identified by MS/MS in global protein subcellular distribution analysis on a global proteomic scale

661	Alpha-glucosidase 2	G2AN	+	Q14697-2	9
662	Ataxin-10	ATXN10	+	Q9UBB4	9
663	Autocrine motility factor	GPI	+	P06744-2	9
664	Brix domain-containing protein 2	BRIX	+	Q8TDN6	9
665	Cardiac lineage protein 1	CLP1	+	O94992	9
666	CCR4-associated factor 3	CNOT3	+	O75175	9
667	cDNA FLJ56329, highly similar to Myosin light polypeptide 6	MYL6	+	B7Z6Z4	9
668	Cell growth-regulating nucleolar protein	LYAR	+	Q9NX58	9
669	CF-1 64 kDa subunit	CSTF2	+	E7EWR4	9
670	COP9 signalosome complex subunit 1	COPS1	+	Q13098-7	9
671	Cullin-3	CUL3	+	Q13618	9
672	Diadenosine tetraphosphate synthetase	GARS	+	P41250	9
673	DNA polymerase delta catalytic subunit	POLD	+	P28340	9
674	eIF-2B GDP-GTP exchange factor subunit alpha	EIF2B1	+	Q14232	9
675	eIF-4-gamma II	EIF4G3	+	H7BYW0	9
676	Embryonal carcinoma differentiation-regulated protein	EDR	+	Q86TG7	9
677	Eukaryotic translation initiation factor 1A, X-chromosomal	EIF1A	+	P47813	9
678	Ewing sarcoma breakpoint region 1 protein	EWS	+	Q01844-5	9
679	Exosome complex exonuclease MTR3	EXOSC6	+	Q5RKV6	9
680	GAP-associated tyrosine phosphoprotein p62	KHDRBS1	+	Q07666	9
681	Golgi-specific brefeldin A-resistance guanine nucleotide exchange factor 1	GBF1	+	Q92538	9
682	GTP-binding protein YPT3	RAB11B	+	Q15907	9
683	Histone H4	H4/A	+	P62805	9
684	Kinesin-like protein KIF2	KIAA1708	+	F8WCP6	9
685	Leucine-rich repeat-containing protein 59	LRRC59	+	Q96AG4	9
686	Lissencephaly-1 protein	LIS1	+	P43034	9
687	Long-chain acyl-CoA synthetase 3	ACS3	+	O95573	9
688	Negative elongation factor C/D	HSPC130	+	Q8IXH7	9
689	Nonsense mRNA reducing factor 2	KIAA1408	+	Q9HAU5	9
690	Nucleolysin TIAR	TIAL1	+	Q01085-2	9
691	Pre-mRNA-splicing factor SRp30C	SFRS9	+	Q13242	9
692	Protein FAM50A	DXS9928E	+	Q14320	9
693	Protein PR264	SFRS2	+	H0YG49	9
694	Protein transport protein Sec24C	KIAA0079	+	P53992	9
695	Putative RNA-binding protein Luc7-like 1	LUC7L	+	Q9NQ29	9
696	Putative uncharacterized protein FAM98B	FAM98B	+	A8MUW5	9

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697	RING finger protein 213	C17orf27	+	Q63HN8-4	9
698	Sucrose nonfermenting protein 2 homolog	SMARCA5	+	O60264	9
699	TAR DNA-binding protein 43	TARDBP	+	Q13148-2	9
700	Treacher Collins syndrome protein	TCOF1	+	E9PHK9	9
701	Tumor protein D52-like 2	hCG_22755	+	Q5JWU6	9
702	Ubiquitin-associated protein 2	KIAA1491	+	Q5T6F2	9
703	Uncharacterized protein C7orf50	C7orf50	+	Q9BRJ6	9
704	UPF0488 protein C8orf33	C8orf33	+	Q9H7E9-2	9
705	100 kDa coated vesicle protein A	ADTAA	+	O95782	8
706	11S regulator complex subunit alpha	IFI5111	+	Q06323	8
707	14-3-3 protein eta	YWHA1	+	Q04917	8
708	39 kDa phosphoribosypyrophosphate synthase-associated protein	PRPSAP1	+	Q14558-2	8
709	50 kDa nuclear polyadenylated RNA-binding protein	BRUNOL2	+	G5EA30	8
710	51 kDa FK506-binding protein	FKBP4	+	Q02790	8
711	60S ribosomal protein L29	RPL29	+	P47914	8
712	60S ribosomal protein L34	RPL34	+	P49207	8
713	60S ribosomal protein L37	RPL37	+	P61927	8
714	6-phosphogluconate dehydrogenase, decarboxylating	PGD	+	P52209	8
715	Actin-like protein 2	ACTR2	+	E9PF41	8
716	Alien homolog	COPS2	+	P61201-2	8
717	Anaphase-promoting complex subunit 1	ANAPC1	+	Q9H1A4	8
718	ATP-binding protein associated with cell differentiation	APACD	+	O14530	8
719	ATP-dependent RNA helicase DDX24	DDX24	+	Q9GZR7	8
720	Aurora kinase A	AIK	+	O14965	8
721	Bleomycin hydrolase	BLMH	+	Q13867	8
722	Calcium homeostasis endoplasmic reticulum protein	CHERP	+	Q8IWX8	8
723	Calcium/calmodulin-dependent protein kinase type II subunit delta	CAMK2D	+	E9PF82	8
724	Calmodulin	CALM	+	H0Y7A7	8
725	Calmodulin-regulated spectrin-associated protein 3	CAMSAP3	+	Q9P1Y5	8
726	Calnexin	CANX	+	P27824	8
727	Calregulin	CALR	+	P27797	8
728	CapZ alpha-1	CAPZA1	+	P52907	8
729	Cell differentiation protein RCD1 homolog	RCD1	+	Q92600	8
730	Cell migration-inducing gene 10 protein	MIG10	+	P00558	8
731	CF-1 77 kDa subunit	CSTF3	+	Q12996	8
732	Cleavage and polyadenylation specificity factor 25 kDa subunit	CFIM25	+	O43809	8

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733	Cofactor required for Sp1 transcriptional activation, subunit 3, 130kDa	CRSP3	+	H7BYY3	8
734	Component of gems 1	SMN	+	E7EQZ4	8
735	COP9 signalosome complex subunit 3	COPS3	+	Q9UNS2	8
736	Cullin-2	CUL2	+	G3V1S2	8
737	Cyclin	PCNA	+	P12004	8
738	Cyclin-G-associated kinase	GAK	+	O14976	8
739	Cysteine and glycine-rich protein 1	CSRP	+	P21291	8
740	Cytoplasmic dynein 1 light intermediate chain 2	DNCLI2	+	O43237	8
741	Cytoplasmic FMR1-interacting protein 1	CYFIP1	+	E7EQ04	8
742	DnaJ homolog subfamily C member 8	DNAJC8	+	O75937	8
743	Double-stranded RNA-binding protein Staufen homolog 2	STAU2	+	E9PF26	8
744	EBNA1-binding protein 2	EBNA1BP2	+	H7C2Q8	8
745	Enhancer of mRNA-decapping protein 3	EDC3	+	Q96F86	8
746	Eukaryotic peptide chain release factor subunit 1	ERF1	+	P62495	8
747	G patch domain-containing protein 9	DXS8237E	+	P98175	8
748	General transcription factor 3C polypeptide 3	GTF3C3	+	Q9Y5Q9	8
749	Hepatoma-derived growth factor	HDGF	+	P51858	8
750	Interferon-inducible double stranded RNA-dependent protein kinase activator A	HSD14	+	O75569	8
751	La ribonucleoprotein domain family member 4B	KIAA0217	+	Q92615	8
752	Macropain delta chain	LMPY	+	P28072	8
753	Myristoylated alanine-rich C-kinase substrate	MACS	+	P29966	8
754	Nucleolar and spindle-associated protein 1	ANKT	+	Q9BXS6	8
755	Nucleolar protein 5	HSPC120	+	Q9Y2X3	8
756	Nucleolar protein 9	NOL9	+	Q5SY16	8
757	p125	MSTP053	+	Q9Y6Y8	8
758	Phosphatidylinositol-4-phosphate 3-kinase C2 domain-containing subunit alpha	PIK3C2A	+	O00443	8
759	Phosphoinositol 3-phosphate-binding protein 2	KIAA1686	+	E9PHQ3	8
760	Pre-mRNA-splicing factor 38B	PRPF38B	+	Q5VTL8	8
761	Pre-mRNA-splicing factor SYF1	HCNP	+	Q9HCS7	8
762	Protein BUD31 homolog	BUD31	+	P41223	8
763	Protein clk-2 homolog	KIAA0683	+	Q9Y4R8	8
764	Protein enabled homolog	ENAH	+	Q8N8S7-2	8
765	Protein FAM91A1	FAM91A1	+	Q658Y4	8
766	Protein raver-1	KIAA1978	+	E9PAU2	8
767	Putative uncharacterized protein SRRM1	RP11-373M8.1-002	+	E9PCT1	8
768	RING finger protein 109	RNF109	+	Q9BRZ2	8

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769	RNA-binding protein PNO1	PNO1	+	Q9NRX1	8
770	Secretory carrier-associated membrane protein 3	C1orf3	+	O14828	8
771	SMARCA4 isoform 2	hCG_29955	+	F5H0X5	8
772	YTH domain family protein 3	YTHDF3	+	Q7Z739	8
773	130 kDa leucine-rich protein	LRP130	+	P42704	7
774	24-dehydrocholesterol reductase	DHCR24	+	Q15392	7
775	38 kDa nuclear protein containing a WW domain	JM26	+	O60828	7
776	40S ribosomal protein S15	RIG	+	P62841	7
777	40S ribosomal protein S15a	OK/SW-cl.82	+	P62244	7
778	60S ribosomal protein L38	RPL38	+	P63173	7
779	8 kDa dynein light chain	DLC1	+	P63167	7
780	95 kDa retinoblastoma-associated protein	BRE1B	+	O75150	7
781	Activator of 90 kDa heat shock protein ATPase homolog 1	AHSA1	+	O95433	7
782	AKAP 120-like protein	AKAP350	+	Q99996-6	7
783	A-kinase anchor protein 8-like	AKAP8L	+	Q9ULX6	7
784	Alpha-NAC	HSD48	+	E9PAV3	7
785	Alpha-soluble NSF attachment protein	NAPA	+	P54920	7
786	Aminoacyl tRNA synthetase complex-interacting multifunctional protein 3	AIMP3	+	O43324	7
787	Amplified in breast cancer gene 2 protein	ABC2	+	Q8ND04-2	7
788	Anaphase-promoting complex subunit 7	ANAPC7	+	Q9UJX3	7
789	Arginine/serine-rich-splicing factor 14	KIAA0365	+	Q8IX01	7
790	Arginine-rich 54 kDa nuclear protein	SFRS11	+	Q05519	7
791	Aspartyl aminopeptidase	ASPEP	+	E7ETB3	7
792	Atlastin-3	ATL3	+	Q6DD88	7
793	ATP-dependent 61 kDa nucleolar RNA helicase	DDX21	+	Q9NY93	7
794	Auberger B antigen	BCAM	+	P50895	7
795	Beta-Pix	ARHGEF7	+	Q14155	7
796	Bromodomain and WD repeat-containing protein 2	BRWD2	+	Q9BZH6	7
797	BTB/POZ domain-containing protein KCTD12	C13orf2	+	Q96CX2	7
798	C-2K	CDC2L4	+	P50750-2	7
799	Casein kinase I isoform alpha	CSNK1A1	+	P48729-2	7
800	Casein kinase II subunit alpha	CK2A2	+	P19784	7
801	CD2 antigen cytoplasmic tail-binding protein 2	CD2BP2	+	O95400	7
802	cDNA FLJ56102, highly similar to Homo sapiens calpastatin (CAST), transcript variant 8, mRNA	CAST	+	P20810-6	7
803	Cell migration-inducing gene 5 protein	MIG5	+	P63000-2	7
804	Cell proliferation-inducing gene 21 protein	GNB2L1	+	P63244	7
805	Cellubrevin	SYB3	+	Q15836	7

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806	Clathrin-coated vesicle/synaptic vesicle proton pump 116 kDa subunit	ATP6N1	+	Q93050-3	7
807	Cohesin subunit SA-2	SA2	+	Q8N3U4-2	7
808	Coiled-coil and C2 domain-containing protein 1A	CC2D1A	+	Q6P1N0	7
809	Coronin-1B	CORO1B	+	Q9BR76	7
810	Crooked neck homolog	CGI-201	+	Q9BZJ0	7
811	Cyclophilin-like protein PPIL4	PPIL4	+	Q8WUA2	7
812	Cytokine IK	IK	+	Q13123	7
813	Double-strand-break repair protein rad21 homolog	HR21	+	O60216	7
814	Dual specificity protein phosphatase 9	DUSP9	+	Q99956	7
815	Dynactin complex subunit 22 kDa subunit	DCTN22	+	O75935	7
816	eIF-4F 25 kDa subunit	EIF4E	+	P06730-2	7
817	Elongation factor sec	EEFSEC	+	P57772	7
818	Enigma homolog	ENH	+	Q96HC4	7
819	ER-Golgi intermediate compartment 53 kDa protein	ERGIC53	+	P49257	7
820	Exonuclease XPMC2	PMC2	+	Q9GZR2	7
821	FSHD region gene 1 protein	FRG1	+	Q14331	7
822	Glycogen phosphorylase, liver form	PYGL	+	P06737	7
823	Heterogeneous nuclear ribonucleoprotein methyltransferase-like protein 3	HRMT1L3	+	O60678	7
824	Histidine--tRNA ligase	HARS	+	P12081	7
825	Histone deacetylase 2	HDAC2	+	Q92769	7
826	Histone H1.5	H1F5	+	P16401	7
827	Histone H1	H1F0	+	P07305	7
828	HSP27 estrogen response element-TATA box-binding protein	HAP	+	Q15424	7
829	hTREX120	CXorf3	+	Q8NI27	7
830	L1880	RAB5C	+	F8W1H5	7
831	Lariat debranching enzyme	DBR1	+	Q9UK59	7
832	LAR-interacting protein 1	LIP1	+	Q13136	7
833	Nonsense mRNA reducing factor 3B	RENT3B	+	Q9BZI7	7
834	Nucleolar protein EMG1 homolog	C2F	+	Q92979	7
835	Nucleosome assembly protein 1-like 4b	NAP1L4	+	A8MZ22	7
836	p59scr	Nbla00526	+	H7BYM7	7
837	p76	TM9SF2	+	Q99805	7
838	Pre-mRNA-splicing factor SRP20	SFRS3	+	P84103	7
839	Pre-rRNA-processing protein TSR1 homolog	KIAA1401	+	Q2NL82	7
840	Profilin I	PFN1	+	P07737	7
841	Programmed cell death protein 5	PDCD5	+	O14737	7
842	Protein CDV3 homolog	CDV3	+	Q9UKY7	7

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843	Protein FAM61A	C19orf13	+	Q8ND56	7
844	Protein FAM98A	FAM98A	+	Q8NCA5	7
845	Protein LAS1 homolog	LAS1L	+	Q9Y4W2	7
846	Protein quaking	HKQ	+	Q96PU8	7
847	PRP4 kinase	KIAA0536	+	Q13523	7
848	Putative uncharacterized protein PLRG1	PLRG1	+	A8MW61	7
849	Ras-related protein Rab-1A	RAB1	+	P62820	7
850	Regulator of differentiation 1	ROD1	+	O95758-4	7
851	Ribonuclease P protein subunit p30	RNASEP2	+	E9PB02	7
852	Ribosome assembly protein BMS1 homolog	BMS1	+	Q14692	7
853	RIO kinase 2	RIOK2	+	Q9BVS4	7
854	RNA-binding motif protein 42	RBM42	+	Q9BTD8	7
855	RNA-binding motif protein 47	RBM47	+	A0AV96	7
856	Rotavirus X-associated non-structural protein	KIAA1031	+	Q9UGR2	7
857	SET domain-containing protein 3	C14orf154	+	Q86TU7	7
858	Syntaxin-7	STX7	+	O15400	7
859	TNF receptor-associated factor 2	TRAF2	+	Q12933-2	7
860	TRM1-like protein	C1orf25	+	Q7Z2T5	7
861	Tubulin beta-6 chain	TUBB6	+	Q9BUF5	7
862	Zinc finger CCHC domain-containing protein 8	ZCCHC8	+	Q6NZY4	7
863	Zinc finger protein 326	ZNF326	+	Q5BKZ1	7
864	140 kDa nuclear and cell adhesion-related phosphoprotein	DRS	+	Q9H307	6
865	214 kDa nucleoporin	CAIN	+	P35658-5	6
866	22 kDa neuronal tissue-enriched acidic protein	BASP1	+	P80723	6
867	2-phospho-D-glycerate hydro-lyase	ENO1	+	P06733	6
868	3-alkyladenine DNA glycosylase	AAG	+	P29372	6
869	40S ribosomal protein S21	RPS21	+	P63220	6
870	46 kDa mannose 6-phosphate receptor	M6PR	+	P20645	6
871	47 kDa heat shock protein	CBP1	+	P50454	6
872	5F7	BSG	+	I3NI00	6
873	60S ribosomal protein L26	RPL26	+	P61254	6
874	93 kDa nucleoporin	KIAA0095	+	Q8N1F7	6
875	A18 hnRNP	A18HNRNP	+	Q14011	6
876	A190	POLR1A	+	O95602	6
877	Actin-related protein 2/3 complex subunit 1B	ARC41	+	O15143	6
878	Actin-related protein 2/3 complex subunit 2	ARC34	+	O15144	6
879	Adapter-related protein complex 2 beta subunit	ADTB2	+	P63010-2	6
880	Adducin-like protein 70	ADD3	+	Q9UEY8	6

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881	AH receptor-interacting protein	AIP	+	O00170	6
882	Anaphase-promoting complex subunit 3	ANAPC3	+	G3V1C4	6
883	Androgen receptor cofactor p44	HKMT1069	+	Q9BQA1	6
884	Angiotensin-II type 2 receptor-interacting protein	ATBP	+	Q9ULD2	6
885	Apolipoprotein E	APOE	+	P02649	6
886	Basic leucine zipper and W2 domain-containing protein 2	BZW2	+	Q9Y6E2	6
887	Beta-catenin-like protein 1	C20orf33	+	Q8WYA6	6
888	Brain-specific angiogenesis inhibitor 1-associated protein 2-like protein 1	BAIAP2L1	+	Q9UHR4	6
889	Calpactin I light chain	ANX2LG	+	P60903	6
890	Calponin, acidic isoform	CNN3	+	Q15417	6
891	cAMP-dependent protein kinase catalytic subunit alpha	PKACA	+	P17612	6
892	Cell division cycle 40 homolog	CDC40	+	O60508	6
893	Coatomer subunit zeta-1	CGI-120	+	F8VVA7	6
894	Coronin-1C_i2 protein	CORO1C	+	A7MAP0	6
895	CUL4- and DDB1-associated WDR protein 2	CDW2	+	Q9H7D7	6
896	Cullin-1	CUL1	+	Q13616	6
897	Cyclin-A/CDK2-associated protein p19	EMC19	+	E5RJR5	6
898	Cysteine--tRNA ligase	CARS	+	P49589-3	6
899	Cytosol aminopeptidase	LAP3	+	P28838	6
900	DAZ-associated protein 1	DAZAP1	+	Q96EP5	6
901	Density-regulated protein	DENR	+	O43583	6
902	DNA ligase 3	LIG3	+	P49916	6
903	DNA primase 58 kDa subunit	PRIM2	+	P49643	6
904	E2-EPF	E2EPF	+	Q16763	6
905	EH domain-containing protein 4	EHD4	+	Q9H223	6
906	eIF-3 p25	ARG134	+	Q9UBQ5	6
907	Endomembrane proton pump 58 kDa subunit	ATP6B2	+	P21281	6
908	Exosome complex exonuclease RRP4	EXOSC2	+	Q13868	6
909	Exosome complex exonuclease RRP41	EXOSC4	+	Q9NPD3	6
910	Exosome complex exonuclease RRP43	EXOSC8	+	Q96B26	6
911	Extended synaptotagmin-1	ESYT1	+	Q9BSJ8-2	6
912	Farnesyl-diphosphate farnesyltransferase	FDFT1	+	P37268	6
913	Fibronectin receptor subunit beta	FNRB	+	P05556	6
914	Focal adhesion kinase 1	FAK	+	E7ESA6	6
915	Foocen	KIAA0886	+	Q9NQC3	6
916	GAPex-5	GAPEX5	+	Q14C86-6	6
917	GATA zinc finger domain-containing protein 2B	GATAD2B	+	Q8WXI9	6

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918	General transcription factor IIF 74 kDa subunit	GTF2F1	+	P35269	6
919	General transcription factor TFIIB	GTF2B	+	Q00403	6
920	Glycine-rich protein	LSM4	+	Q9Y4Z0	6
921	Glycogenin-1	GYG	+	P46976	6
922	Heterogeneous nuclear ribonucleoprotein 2H9	HNRNPH3	+	P31942	6
923	Histone deacetylase 1	HDAC1	+	Q13547	6
924	HIV-1-induced protein HIN-1	HIN1	+	G3V0I6	6
925	Hsc70-interacting protein	FAM10A1	+	P50502	6
926	HTPHLP	PDCL3	+	Q9H2J4	6
927	Kinesin light chain 1	KLC	+	E7EVH7	6
928	Kinesin light chain 2	KLC2	+	Q9H0B6	6
929	Male germ cell RacGap	KIAA1478	+	Q9H0H5	6
930	Meiotic recombination REC14 protein homolog	WDR61	+	Q9GZS3	6
931	Membrane-associated protein HEM-2	HEM2	+	Q9Y2A7-2	6
932	Methyl-CpG-binding domain protein 3	MBD3	+	O95983	6
933	Microsomal triglyceride transfer protein large subunit	MTP	+	E9PBP6	6
934	Mitochondrial distribution and morphology protein 20	C12orf30	+	Q14CX7	6
935	MLC-2A	MRLC2	+	O14950	6
936	mRNA (guanine-N(7)-)-methyltransferase	KIAA0398	+	O43148-2	6
937	Multiple myeloma tumor-associated protein 2	C1orf35	+	Q9BU76	6
938	N-alpha-acetyltransferase 10, NatA catalytic subunit	ARD1	+	P41227	6
939	NF-kappa-B-activating protein	NKAP	+	Q8N5F7	6
940	NOL1/NOP2/Sun domain family member 5	NSUN5	+	Q96P11-4	6
941	Nucleolar complex protein 14	C4orf9	+	P78316	6
942	Nucleolar phosphoprotein B23	NPM	+	P06748	6
943	Nucleolar protein 130	NUC130	+	Q9NVU7	6
944	One-twenty two protein 1	OTT	+	Q96T37	6
945	Partner of ARF	C9orf86	+	Q3YEC7-2	6
946	PHD finger-like domain-containing protein 5A	PHF5A	+	Q7RTV0	6
947	Phosphoinositide 3-kinase adaptor protein	PIK3R4	+	Q99570	6
948	Placental ribonuclease inhibitor	PRI	+	P13489	6
949	Polyposis locus protein 1-like 1	C19orf32	+	Q96HR9	6
950	Pre-mRNA-splicing factor RBM22	199G4	+	Q9NW64	6
951	Pre-mRNA-splicing factor SRP75	SFRS4	+	Q08170	6
952	Protein DEK	DEK	+	P35659	6
953	Protein linking IAP with cytoskeleton 1	DA41	+	Q9UMX0	6
954	Purine-rich single-stranded DNA-binding protein alpha	PUR1	+	Q00577	6
955	Putative TRAF4-associated factor 1	C15orf23	+	Q9Y448	6
956	Putative uncharacterized protein ARPC4	ARPC4	+	C9JWM7	6

Continuing Appendix 1. List of proteins identified by MS/MS in global protein subcellular distribution analysis on a global proteomic scale

957	Putative uncharacterized protein RAB3GAP1	RAB3GAP1	+	C9J837	6
958	RING finger protein 160	C21orf10	+	O94822	6
959	Signal recognition particle 9 kDa protein	SRP9	+	P49458	6
960	Small nuclear ribonucleoprotein Sm D3	SNRPD3	+	P62318	6
961	Splicing factor U2AF 35 kDa subunit	U2AF1	+	Q01081	6
962	Target of EGR1 protein 1	TOE1	+	Q96GM8	6
963	Testis-expressed sequence 10 protein	L18	+	Q9NXF1	6
964	ThiFP1	UBA5	+	Q9GZZ9	6
965	U2 small nuclear ribonucleoprotein B	SNRPB2	+	P08579	6
966	Uncharacterized protein C3orf26	C3orf26	+	Q9BQ75	6
967	WD repeat-containing protein 18	WDR18	+	Q9BV38	6
968	11S regulator complex subunit beta	PSME2	+	Q9UL46	5
969	14-3-3 protein gamma	YWHAG	+	P61981	5
970	21 kDa transmembrane-trafficking protein	TMED10	+	P49755	5
971	33 kDa VAMP-associated protein	VAP33	+	Q9P0L0-2	5
972	3-hydroxy-3-methylglutaryl coenzyme A synthase	HMGCS	+	Q01581	5
973	40S ribosomal protein S26	RPS26	+	P62854	5
974	40S ribosomal protein S27a	RPS27A	+	P62979	5
975	40S ribosomal protein S29	RPS29	+	P62273	5
976	40S ribosomal protein S30	FAU	+	E9PR30	5
977	5-aminoimidazole-4-carboxamide ribonucleotide formyltransferase	ATIC	+	P31939	5
978	60S ribosomal protein L10	DXS648E	+	P27635	5
979	7SK snRNA methylphosphate capping enzyme	BCDIN3	+	Q7L2J0	5
980	8 kDa dynein light chain b	DLC2	+	Q96FJ2	5
981	Acetyl-CoA acetyltransferase, cytosolic	ACAT2	+	Q9BWD1	5
982	Activator-recruited cofactor 205 kDa component	ARC205	+	Q15648	5
983	Adapter-related protein complex 2 mu subunit	AP2M1	+	E9PFW3	5
984	Amphiregulin-associated protein	MDK	+	E9PLM6	5
985	Antigen NY-CO-13	P53	+	P04637	5
986	Arg/Glu/Asp-rich protein of 120 kDa	RBM25	+	P49756	5
987	Associate of Myc 1	AMY1	+	Q99417	5
988	ATPase family protein 2 homolog	AFG2	+	Q8NB90	5
989	ATP-dependent RNA helicase DDX18	DDX18	+	Q9NVP1	5
990	ATP-dependent RNA helicase DDX54	DDX54	+	Q8TDD1-2	5
991	ATP-dependent RNA helicase DHX8	DDX8	+	Q14562	5
992	Atypical PKC isotype-specific-interacting protein	PAR3	+	Q8TEW0	5
993	Autoantigen p27	SSSCA1	+	O60232	5
994	BAG family molecular chaperone regulator 2	BAG2	+	O95816	5

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995	Beta-II spectrin	SPTB2	+	Q01082	5
996	BRAF35-HDAC complex protein BHC110	AOF2	+	F6S0T5	5
997	CASP10	PPIG	+	Q13427	5
998	CCR4-NOT transcription complex subunit 10	CNOT10	+	F8WAF2	5
999	cDNA FLJ38069 fis, clone CTONG2015434, highly similar to DOUBLE-STRAND BREAK REPAIR PROTEIN MRE11A	HNGS1	+	B3KTC7	5
1000	cDNA FLJ52765, highly similar to Calponin-2	CNN2	+	B4DUT8	5
1001	Cell cycle and apoptosis regulatory protein 1	CARP1	+	Q8IX12	5
1002	Centromere protein V	CENPV	+	Q7Z7K6	5
1003	Chromatin assembly factor 1 subunit C	RBAP48	+	Q09028	5
1004	Chromosome 20 open reading frame 4	C20orf4	+	A2A2Q9	5
1005	Chromosome-associated protein H2	CAPH2	+	Q6IBW4-4	5
1006	CMP-N-acetylneuraminic acid synthase	CMAS	+	Q8NFW8	5
1007	Connectin	TTN	+	Q8WZ42-8	5
1008	DEAH box protein 40	ARG147	+	Q8IX18	5
1009	Dermatomyositis associated with cancer putative autoantigen 1	C20orf21	+	Q9BYJ9	5
1010	DNA primase 49 kDa subunit	PRIM1	+	P49642	5
1011	DNA topoisomerase 2	TOP2A	+	P11388-4	5
1012	DNA-directed RNA polymerase III largest subunit	POLR3A	+	O14802	5
1013	DnaJ homolog subfamily B member 1	DNAJ1	+	P25685	5
1014	E3 ubiquitin-protein ligase BRE1A	BRE1A	+	Q5VTR2	5
1015	E3 ubiquitin-protein ligase KCMF1	FIGC	+	Q9P0J7	5
1016	Elongin 15 kDa subunit	TCEB1	+	Q15369	5
1017	ERT1	ERK2	+	P28482	5
1018	Exosome complex exonuclease RRP42	EXOSC7	+	Q15024	5
1019	Factor interacting with PAP	FIP1	+	Q6UN15-3	5
1020	G patch domain-containing protein 6	GPATC6	+	Q8N5A5	5
1021	Gamma-taxilin	CXorf15	+	Q9NUQ3	5
1022	Gene associated with retinoic and interferon-induced mortality 12 protein	GRIM12	+	Q16881	5
1023	Glucose-6-phosphate 1-dehydrogenase	G6PD	+	P11413-2	5
1024	Glutamate decarboxylase	GLNS	+	P15104	5
1025	Glutathione S-transferase P	FAEES3	+	P09211	5
1026	H/ACA ribonucleoprotein complex subunit 1	GAR1	+	Q9NY12	5
1027	HBV pre-S2 trans-regulated protein 3	CGI-117	+	Q9Y3C1	5
1028	HEAT repeat-containing protein 2	HEATR2	+	Q86Y56	5
1029	Histone acetyltransferase type B subunit 2	RBAP46	+	Q16576	5
1030	Histone H1.2	H1F2	+	P16403	5

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1031	Histone H1.4	H1F4	+	P10412	5
1032	Homogentisate 1,2-dioxygenase	HGD	+	Q93099	5
1033	Importin beta-2	KPNB2	+	Q92973	5
1034	Keratinocyte protein IEF SSP 9502	PWP1	+	Q13610	5
1035	La ribonucleoprotein domain family member 7	HDCMA18P	+	Q4G0J3	5
1036	Leydig cell tumor 10 kDa protein homolog	C19orf53	+	Q9UNZ5	5
1037	Malignant T cell-amplified sequence 1	MCT1	+	Q9ULC4-3	5
1038	Maternal embryonic leucine zipper kinase	KIAA0175	+	Q14680	5
1039	MKI67 FHA domain-interacting nucleolar phosphoprotein	MKI67IP	+	Q9BYG3	5
1040	mRNA-decapping enzyme 1A	DCP1A	+	Q9NPI6	5
1041	NEDD8 carrier protein	UBC12	+	P61081	5
1042	Nuclear protein SkiP	SKIIP	+	G3V3A4	5
1043	Nucleoside diphosphate kinase	hCG_2001850	+	Q32Q12	5
1044	Plastin-3	PLS3	+	P13797	5
1045	Polyadenylate-binding protein-interacting protein 1	PAIP1	+	Q9H074	5
1046	PP2A B subunit isoform B56-delta	PPP2R5D	+	Q14738	5
1047	PP2A subunit B isoform alpha	PPP2R2A	+	P63151-2	5
1048	Pre-mRNA cleavage complex 2 protein Pcf11	KIAA0824	+	O94913	5
1049	Protein FAM115A	FAM115A	+	Q9Y4C2	5
1050	Protein FAM96B	CGI-128	+	Q9Y3D0	5
1051	Protein KIAA0664	KIAA0664	+	O75153	5
1052	Protein LSM12 homolog	LSM12	+	Q3MHD2-2	5
1053	Protein MON2 homolog	KIAA1040	+	Q7Z3U7	5
1054	Protein SEC13 homolog	D3S1231E	+	P55735	5
1055	Protein Smaug homolog 2	SAMD4B	+	Q5PRF9	5
1056	Protein SMG9	C19orf61	+	Q9H0W8	5
1057	Pumilio homolog 2	KIAA0235	+	Q8TB72	5
1058	Putative RNA-binding protein 3	RBM3	+	P98179	5
1059	Putative uncharacterized protein DBN1	DBN1	+	A8MV58	5
1060	Putative uncharacterized protein TCEB2	hCG_1778969	+	B7WPD3	5
1061	Rab11 family-interacting protein 1	RAB11FIP1	+	Q6WKZ4	5
1062	Ras-related protein Rab-18	RAB18	+	Q9NP72	5
1063	Ras-related protein Rab-1B	RAB1B	+	Q9H0U4	5
1064	Ras-related protein Rab-2A	RAB2	+	P61019	5
1065	Reticulocalbin-1	RCN	+	Q15293	5
1066	RNA-binding protein Musashi homolog 1	MSI1	+	O43347	5
1067	Secretory carrier-associated membrane protein 1	SCAMP	+	O15126	5

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1068	Sodium/potassium-dependent ATPase subunit beta-1	ATP1B	+	P05026	5
1069	Symplekin	SPK	+	Q92797	5
1070	Synaptobrevin-like protein 1	SYBL1	+	P51809-2	5
1071	U1 small nuclear ribonucleoprotein C	SNRPC	+	P09234	5
1072	U6 snRNA-associated Sm-like protein LSm3	LSM3	+	P62310	5
1073	U6 snRNA-associated Sm-like protein LSm6	LSM6	+	P62312	5
1074	Uncharacterized protein C1orf77	C1orf77	+	Q9Y3Y2-3	5
1075	Vacuolar proton pump subunit G 1	ATP6G	+	O75348	5
1076	Zinc finger protein 207	ZNF207	+	H0Y3M2	5
1077	Zinc finger ZZ-type and EF-hand domain-containing protein 1	KIAA0399	+	O43149	5
1078	18 kDa phosphoprotein	CFL	+	E9PK25	4
1079	1-phosphatidylinositol-4,5-bisphosphate phosphodiesterase beta-3	PLCB3	+	Q01970	4
1080	30 kDa splicing factor SMNrp	SMNDC1	+	O75940	4
1081	3-5 exonuclease ERI1	3EXO	+	Q8IV48	4
1082	45 kDa-splicing factor	RBM17	+	Q96I25	4
1083	60S acidic ribosomal protein P1	RPLP1	+	P05386	4
1084	60S ribosomal protein L36a	GIG15	+	P83881	4
1085	68 kDa TATA-binding protein-associated factor	RBP56	+	Q92804	4
1086	A34.5	ASE1	+	O15446-2	4
1087	Adapter protein CMS	CD2AP	+	Q9Y5K6	4
1088	Adenylate cyclase-stimulating G alpha protein	GNAS	+	Q5JWF2	4
1089	Adenylosuccinase	ADSL	+	P30566	4
1090	Akt substrate of 160 kDa	AS160	+	O60343	4
1091	Antioxidant enzyme AOE372	PRDX4	+	Q13162	4
1092	Apolipoprotein B-100	APOB	+	P04114	4
1093	ATP-dependent helicase IGHMBP2	IGHMBP2	+	P38935	4
1094	ATP-dependent RNA helicase DDX19A	DDX19A	+	Q9NUU7	4
1095	ATP-dependent RNA helicase DDX39	DDX39	+	O00148	4
1096	ATP-dependent RNA helicase ROK1-like	DDX52	+	Q9Y2R4	4
1097	AU-rich element RNA-binding protein 1	AUF1	+	Q14103	4
1098	Autoantigen PM/Scl 1	EXOSC9	+	Q06265-2	4
1099	B(2)GCN homolog	EIF3A	+	P56537	4
1100	Barrier-to-autointegration factor	BAF	+	O75531	4
1101	Bax antagonist selected in saccharomyces 1	C21orf50	+	P18583-9	4
1102	Beta-adrenergic receptor kinase 1	ADRBK1	+	P25098	4
1103	Binder of OVCA1-1	HSPC114	+	Q9Y5S9	4
1104	Bloom syndrome protein	BLM	+	P54132	4

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1105	Calcium/calmodulin-dependent protein kinase (CaM kinase) II gamma	CAMK2G	+	Q13555-4	4
1106	Calumenin	CALU	+	O43852-3	4
1107	Cancer/testis antigen 3.2	MAGEB2	+	O15479	4
1108	Cancer-associated Sm-like	CASM	+	O15116	4
1109	Casein kinase 2 beta polypeptide	CSNK2B	+	Q5SRQ6	4
1110	Casein kinase I isoform epsilon	CSNK1E	+	P49674	4
1111	CCR4-associated factor 2	CDC36	+	Q9NZN8	4
1112	Cdc2-related kinase, arginine/serine-rich	CDK12	+	Q9NYV4	4
1113	CDKN2A-interacting protein N-terminal-like protein	CDKN2AIPNL	+	Q96HQ2	4
1114	cDNA FLJ59311, highly similar to Long-chain-fatty-acid--CoA ligase 1 (EC 6.2.1.3)	ACSL1	+	B7Z452	4
1115	Cell division control protein 42 homolog	CDC42	+	P60953	4
1116	Chromosome 21 leucine-rich protein	C21LRP	+	O95456	4
1117	Chronic myelogenous leukemia tumor antigen 28	CML28	+	Q9NQT4	4
1118	Citron Rho-interacting kinase	CIT	+	O14578-4	4
1119	Coatomer subunit gamma-2	COPG2	+	Q9UBF2	4
1120	Coiled-coil domain-containing protein 55	CCDC55	+	Q9H0G5	4
1121	Coiled-coil domain-containing protein 72	CCDC72	+	Q9Y2S6	4
1122	Coiled-coil domain-containing protein 8	CCDC8	+	Q9H0W5	4
1123	Cold shock domain-containing protein A	CSDA	+	P16989	4
1124	Component of gems 2	GEMIN2	+	O14893	4
1125	COP9 signalosome complex subunit 6	COPS6	+	Q7L5N1	4
1126	CTCL tumor antigen se20-10	CKAP2	+	Q8WWK9	4
1127	DCN1-like protein 5	DCUN1D5	+	Q9BTE7	4
1128	DEAD box protein 49	DDX49	+	Q9Y6V7	4
1129	DnaJ homolog subfamily A member 5	DNAJA5	+	Q5F1R6-2	4
1130	Dynein, light chain, roadblock-type 1	DYNLRB1	+	B1AKR6	4
1131	eIF4E-like protein 4E-LP	EIF4E2	+	O60573	4
1132	Embryonic growth-associated protein homolog	EGAP	+	Q5VZE5	4
1133	Enaptin	C6orf98	+	E7EQI5	4
1134	Endobrevin	VAMP8	+	Q9BV40	4
1135	ER-Golgi SNARE of 24 kDa	SEC22B	+	O75396	4
1136	Forkhead box protein K1	FOXK1	+	P85037	4
1137	Friend of EBNA2 protein	FOE	+	Q7Z5K2-3	4
1138	G2/mitotic-specific cyclin-B1	CCNB	+	P14635	4
1139	Gamma-ring complex protein 76 kDa	76P	+	Q9UGJ1	4
1140	Gastric cancer-related protein VRG107	FAM2C	+	P84101	4
1141	General transcription factor 3C polypeptide 2	GTF3C2	+	Q8WUA4	4

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1142	General transcription factor IIE subunit 2	GTF2E2	+	P29084	4
1143	General vesicular transport factor p115	USO1	+	O60763-2	4
1144	Glycoprotein 25L2	GP25L2	+	Q9BVK6	4
1145	Guanosine diphosphate dissociation inhibitor 2	GDI2	+	E7EU23	4
1146	HCNPpp	PBP	+	P30086	4
1147	High mobility group nucleosome-binding domain-containing protein 2	HMG17	+	P05204	4
1148	Histone H1.1	H1F1	+	Q02539	4
1149	Hsp90 chaperone protein kinase-targeting subunit	CDC37	+	Q16543	4
1150	Importin alpha Q2	KPNA3	+	O00505	4
1151	Intracellular hyaluronan-binding protein 4	HABP4	+	Q5JVS0	4
1152	LIM mineralization protein	ENIGMA	+	Q9NR12	4
1153	Macropain epsilon chain	LMPX	+	P28074	4
1154	MAP1 light chain LC2	MAP1A	+	E9PGC8	4
1155	Maternal-embryonic 3	MEM3	+	Q96QK1	4
1156	Methionine adenosyltransferase 2	AMS2	+	P31153	4
1157	Molecule associated with JAK3 N-terminus	KIAA0216	+	Q92614	4
1158	MORF-related gene 15 protein	FWP006	+	Q9UBU8	4
1159	MORF-related gene X protein	KIAA0026	+	Q15014	4
1160	Muscleblind-like protein 1	EXP	+	Q9NR56	4
1161	N-alpha-acetyltransferase 38, NatC auxiliary subunit	LSM8	+	O95777	4
1162	Nercc1 kinase	KIAA1995	+	Q8TD19	4
1163	Neurochondrin	KIAA0607	+	Q9UBB6-3	4
1164	NHP2 non-histone chromosome protein 2-like 1 (S. cerevisiae)	CTA-216E10.8-008	+	B1AHD1	4
1165	NSFL1 cofactor p47	NSFL1C	+	Q9UNZ2-5	4
1166	Oxidative stress-responsive 1 protein	KIAA1101	+	O95747	4
1167	Parvulin-14	PIN4	+	Q9Y237-2	4
1168	Polo-like kinase 1	PLK	+	P53350	4
1169	Polycomb protein EED	EED	+	O75530-2	4
1170	Pre-mRNA branch site protein p14	CGI-110	+	Q9Y3B4	4
1171	Proline-rich protein 8	PRR8	+	Q96EV2	4
1172	Protein A6	PTK9	+	Q12792-3	4
1173	Protein CoREST	KIAA0071	+	Q9UKL0	4
1174	Protein disulfide isomerase P5	PDIA6	+	Q15084-2	4
1175	Protein FAM61B	C20orf40	+	Q9BX40	4
1176	Protein kinase C-like 2	PKN2	+	Q16513	4
1177	Protein lin-41 homolog	LIN41	+	Q2Q1W2	4
1178	Protein regulator of cytokinesis 1	PRC1	+	O43663	4

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1179	Protein transport protein Sec16A	KIAA0310	+	O15027-5	4
1180	Protein ZYP	RBX1	+	P62877	4
1181	Putative uncharacterized protein APEH	APEH	+	C9JIF9	4
1182	Putative uncharacterized protein FAM128B	FAM128B	+	B8ZZ87	4
1183	Rhotekin	RTKN	+	Q9BST9	4
1184	RNA 3-terminal phosphate cyclase	RPC	+	O00442-2	4
1185	RNA-binding motif protein 28	RBM28	+	Q9NW13	4
1186	Serine/threonine-protein phosphatase PP1-alpha catalytic subunit	PPP1A	+	P62136	4
1187	Sm protein F	PBSCF	+	P62306	4
1188	Splicing factor, arginine/serine-rich 10	SFRS10	+	P62995	4
1189	Surfeit locus protein 4	SURF4	+	O15260	4
1190	Syntaxin-12	STX12	+	Q86Y82	4
1191	Translin	TSN	+	Q15631	4
1192	Tubulin beta chain	OK/SW-cl.56	+	P07437	4
1193	Tubulin beta-2A chain	TUBB2	+	Q13885	4
1194	Uncharacterized protein KIAA0406	KIAA0406	+	O43156	4
1195	UPF0534 protein C4orf43	C4orf43	+	H0Y9X1	4
1196	Villin-1	VIL	+	P09327	4
1197	Zinc finger protein 593	ZNF593	+	O00488	4
1198	115 kDa guanine nucleotide exchange factor	ARHGEF1	+	Q92888-3	3
1199	1-Cys peroxiredoxin	AOP2	+	P30041	3
1200	2,3-cyclic-nucleotide 3-phosphodiesterase	CNP	+	P09543	3
1201	32 kDa accessory protein	ATP6D	+	F5GYQ1	3
1202	3-5 exoribonuclease CSL4 homolog	CGI-108	+	Q9Y3B2	3
1203	40S ribosomal protein S27	MPS1	+	P42677	3
1204	40S ribosomal protein S27-like	RPS27L	+	H0YMV8	3
1205	5-AMP-activated protein kinase catalytic subunit alpha-1	AMPK1	+	Q13131-2	3
1206	60 kDa SS-A/Ro ribonucleoprotein	RO60	+	P10155	3
1207	60S ribosomal protein L26-like 1	RPL26L1	+	Q9UNX3	3
1208	6C6-AG tumor-associated antigen	BAP31	+	P51572-2	3
1209	90 kDa ribosomal protein S6 kinase 3	ISPK1	+	P51812	3
1210	Actin-related protein 2/3 complex subunit 1A	ARPC1A	+	Q92747	3
1211	Activating signal cointegrator 1 complex subunit 1	ASCC1	+	F5H874	3
1212	Actopaxin	MXRA2	+	Q9NVD7	3
1213	Adapter-related protein complex 1 sigma-1A subunit	AP19	+	H7C1E4	3
1214	Adapter-related protein complex 3 sigma-1 subunit	AP3S1	+	Q92572	3
1215	ADP-ribosylation factor 3	ARF3	+	P61204	3

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1216	Aging-associated gene 10 protein	AAG10	+	Q9Y6H1	3
1217	Alpha-tubulin 6	TUBA1C	+	F5H5D3	3
1218	Anaphase-promoting complex subunit 4	ANAPC4	+	E9PCR4	3
1219	Ankyrin repeat and zinc finger domain-containing protein 1	ANKZF1	+	Q9H8Y5	3
1220	APG9-like 1	APG9L1	+	Q7Z3C6	3
1221	APOBEC1 complementation factor	A1CF	+	Q9NQ94	3
1222	Aryl sulfotransferase 1	OK/SW-cl.88	+	P50225	3
1223	ATL-derived factor	TRDX	+	P10599	3
1224	Atypical protein kinase C-lambda/iota	DXS1179E	+	P41743	3
1225	Basic transcription factor 2 80 kDa subunit	ERCC2	+	P18074	3
1226	B-CK	CKB	+	P12277	3
1227	BPM90	RANBP9	+	Q96S59	3
1228	BRG1-associated factor 155	BAF155	+	Q92922	3
1229	BTG1-binding factor 1	CAF1	+	Q9UIV1	3
1230	cAMP-dependent protein kinase type I-alpha regulatory subunit	PKR1	+	P10644	3
1231	CapZ alpha-2	CAPZA2	+	P47755	3
1232	CD107 antigen-like family member A	LAMP1	+	P11279	3
1233	CDC42 GTPase-activating protein	ARHGAP1	+	Q07960	3
1234	cDNA FLJ52228, highly similar to Mps one binder kinase activator-like 1A	MOB4A	+	B4DRY3	3
1235	cDNA FLJ61322, highly similar to RNA-binding motif,single-stranded-interacting protein 1	C2orf12	+	B4DN88	3
1236	Cell cycle progression restoration gene 3 protein	CPR3	+	O60884	3
1237	Chloride channel ABP	CLIC1	+	O00299	3
1238	Chromosome 1 open reading frame 57	C1orf57	+	Q5TDF0	3
1239	Cleavage and polyadenylation specificity factor 100 kDa subunit	CPSF100	+	Q9P2I0	3
1240	CLL-associated antigen KW-7	DFS70	+	O75475	3
1241	Coilin	CLN80	+	P38432	3
1242	COPII-associated small GTPase	SAR1	+	Q9NR31	3
1243	DDB1- and CUL4-associated factor 7	DCAF7	+	P61962	3
1244	Delta(6) fatty acid desaturase	FADS2	+	O95864	3
1245	DNA polymerase alpha 70 kDa subunit	POLA2	+	Q14181	3
1246	Dopamine-responsive gene 1 protein	C6orf55	+	Q9NP79	3
1247	Dual specificity mitogen-activated protein kinase kinase 2	MAP2K2	+	P36507	3
1248	Elongation factor 1-alpha 2	EEF1A2	+	Q05639	3
1249	Ezrin-radixin-moesin-binding phosphoprotein 50	NHERF	+	O14745	3
1250	G protein subunit beta-2	GNB2	+	P62879	3

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1251	General transcription factor 3C polypeptide 4	GTF3C4	+	Q9UKN8	3
1252	General transcription factor IIE 56 kDa subunit	GTF2E1	+	P29083	3
1253	GTP-binding protein smg p21B	OK/SW-cl.11	+	P61224	3
1254	HECT domain and RCC1-like domain-containing protein 2	HERC2	+	O95714	3
1255	HECT domain-containing protein 3	HECTD3	+	Q5T447	3
1256	Heterogeneous nuclear ribonucleoprotein A3	HNRNPA3	+	P51991	3
1257	Heterogeneous nuclear ribonucleoprotein L-like	BLOCK24	+	Q8WVV9	3
1258	Histone H1.3	H1F3	+	P16402	3
1259	Histone H3.1	H3FA	+	P68431	3
1260	HLA-DR-associated protein II	SET	+	Q01105	3
1261	Importin subunit alpha-1	KPNA1	+	P52294	3
1262	Importin-9	HSPC273	+	Q96P70	3
1263	Lamina-associated polypeptide 2, isoform alpha	LAP2	+	P42166	3
1264	LIV-1 subfamily of ZIP zinc transporter 4	KIAA0062	+	Q15043	3
1265	Lung cancer antigen NY-LU-12	DEF3	+	P78332	3
1266	MST156	KIAA1341	+	Q9P2K5	3
1267	N-acetyltransferase 13	MAK3	+	Q9GZZ1	3
1268	Nef-binding protein 1	ATP6V1H	+	Q9UI12	3
1269	Neural precursor cell expressed developmentally down-regulated protein 1	NEDD1	+	G3V3F1	3
1270	Nuclear poly(A)-binding protein 1	PAB2	+	Q86U42	3
1271	Nucleoporin NUP188 homolog	KIAA0169	+	Q5SRE5	3
1272	Oxysterol-binding protein 1	OSBP	+	P22059	3
1273	Phosphorylase b kinase regulatory subunit beta	PHKB	+	Q93100	3
1274	Poly(ADP-ribose) glycohydrolase	PARG	+	Q86W56	3
1275	Pre-mRNA-splicing factor SF3b 49 kDa subunit	SAP49	+	Q15427	3
1276	Proteasome inhibitor PI31 subunit	PSMF1	+	Q92530	3
1277	Protein FAM195B	FAM195B	+	I3L4Q0	3
1278	Protein G7b	C6orf28	+	Q9Y333	3
1279	Protein LAPS18-like	C12orf31	+	Q9BRT6	3
1280	Protein transport protein Sec23B	SEC23B	+	Q15437	3
1281	Protein VAC14 homolog	TAX1BP2	+	Q08AM6	3
1282	Putative ubiquitin-conjugating enzyme E2 N-like	UBE2NL	+	Q5JXB2	3
1283	Putative uncharacterized protein KIF20B	KIF20B	+	Q96Q89-4	3
1284	Putative uncharacterized protein NMD3	NMD3	+	C9JA08	3
1285	Putative uncharacterized protein PCYT1A	PCYT1A	+	C9JEJ2	3
1286	Putative uncharacterized protein TNPO3	TNPO3	+	C9J7E5	3
1287	Ras-related protein Rab-21	KIAA0118	+	Q9UL25	3

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1288	Rho cDNA clone 9	ARH9	+	E9PN11	3
1289	Ribosome biogenesis regulatory protein homolog	KIAA0112	+	Q15050	3
1290	RNA 3-terminal phosphate cyclase-like protein	HSPC338	+	Q9Y2P8	3
1291	RNA polymerase B transcription factor 3	BTF3	+	P20290	3
1292	RNA-binding protein with serine-rich domain 1	LDC2	+	Q15287	3
1293	Secretory carrier-associated membrane protein 2	SCAMP2	+	O15127	3
1294	Signal transducer and activator of transcription 1-alpha/beta	STAT1	+	P42224	3
1295	Small nuclear ribonucleoprotein G-like protein	PBSCG	+	A8MWD9	3
1296	Small nuclear ribonucleoprotein Sm D1	SNRPD1	+	P62314	3
1297	Thymosin beta-10	PTMB10	+	P63313	3
1298	TRM112-like protein	AD-001	+	Q9UI30	3
1299	Ubiquitin-like 4A	UBL4A	+	Q5HY81	3
1300	Uncharacterized methyltransferase WBSCR22	HUSSY-03	+	O43709	3
1301	UPF0293 protein C16orf42	C16orf42	+	Q9UJK0	3
1302	UPF0760 protein C2orf29	C2orf29	+	Q9UKZ1	3
1303	Vacuolar proton pump subunit E 1	ATP6E	+	P36543	3
1304	WD repeat-containing protein 59	FP977	+	Q6PJI9	3
1305	Zinc finger CCHC domain-containing protein 4	ZCCHC4	+	Q9H5U6	3
1306	Zinc finger protein 622	ZNF622	+	Q969S3	3
1307	11-zinc finger protein	CTCF	+	P49711	2
1308	14-3-3 protein beta/alpha	YWHAB	+	P31946	2
1309	26S proteasome non-ATPase regulatory subunit 4	MCB1	+	P55036	2
1310	27 kDa Golgi SNARE protein	GOSR2	+	E7EQ34	2
1311	3-beta-hydroxysteroid-Delta(8),Delta(7)-isomerase	EBP	+	Q15125	2
1312	40 kDa SR-repressor protein	FUSIP1	+	O75494	2
1313	5-phosphoribosylglycinamide transformylase	GART	+	P22102	2
1314	60S acidic ribosomal protein P0	RPLP0	+	P05388	2
1315	60S ribosomal protein L36a-like	RPL36AL	+	Q969Q0	2
1316	60S ribosomal protein L39	RPL39	+	P62891	2
1317	60S ribosomal protein L40	UBA52	+	P62987	2
1318	Actin, alpha cardiac muscle 1	ACTC	+	P68032	2
1319	Adhesion protein with leucine-rich repeats and immunoglobulin domains related to perlecan	MXRA5	+	Q9NR99	2
1320	ADP-ribosylation factor 4	ARF2	+	P18085	2
1321	ADP-ribosylation factor-like protein 10C	ARL10C	+	Q9NVJ2	2
1322	Alcohol dehydrogenase class 4 mu/sigma chain	ADH7	+	P40394-2	2
1323	Ally of AML-1 and LEF-1	ALY	+	Q86V81	2
1324	Androgen receptor trapped clone 27 protein	HSPC024	+	Q9UBK9	2

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1325	Antigen MLAA-22	BCOX1	+	F6XS94	2
1326	Antigen NY-CO-4	EEF1D	+	E9PRY8	2
1327	Antigen NY-CO-7	CHIP	+	Q9UNE7	2
1328	Anti-silencing function protein 1 homolog B	ASF1B	+	Q9NVP2	2
1329	Ataxia telangiectasia mutated	ATM	+	Q13315	2
1330	ATP-dependent RNA helicase eIF4A-2	DDX2B	+	Q14240-2	2
1331	Beta-III spectrin	KIAA0302	+	O15020	2
1332	Binding to curved DNA	BTCD	+	O60870	2
1333	Brain-type aldolase	ALDC	+	P09972	2
1334	Cactin	C19orf29	+	Q8WUQ7-2	2
1335	Cadherin family member 15	CDHF15	+	Q8TDW7	2
1336	Caldesmon	CAD	+	Q05682	2
1337	Cancer/testis antigen 58	NALP4	+	Q96MN2	2
1338	Cardiomyopathy-associated protein 2	CMYA2	+	Q5VU43-3	2
1339	cDNA FLJ58897, moderately similar to Dpy-30-like protein	hCG_2039808	+	B4DIS3	2
1340	cDNA FLJ59559, highly similar to Glutaryl-CoA dehydrogenase, mitochondrial (EC 1.3.99.7)	DC2	+	D6RH22	2
1341	cDNA FLJ60124, highly similar to Mitochondrial dicarboxylate carrier	MRPL12	+	B4DLN1	2
1342	Cell cycle regulatory protein	CHC1	+	P18754-2	2
1343	Cellular nucleic acid-binding protein	CNBP	+	P62633	2
1344	Chromobox protein homolog 3	CBX3	+	Q13185	2
1345	Cleavage and polyadenylation specificity factor 73 kDa subunit	CPSF3	+	Q9UKF6	2
1346	Coiled-coil domain-containing protein 12	CCDC12	+	Q8WUD4	2
1347	Component of oligomeric Golgi complex 3	COG3	+	Q96JB2	2
1348	Conserved edge-expressed protein	C7orf20	+	Q7L5D6	2
1349	CpG island protein	F8A	+	P23610	2
1350	CTD-binding SR-like protein RA4	KIAA1172	+	O95104	2
1351	Cyclin-T1	CCNT1	+	O60563	2
1352	Cytosolic malate dehydrogenase	MDH1	+	F5H098	2
1353	Cytosolic prostaglandin E2 synthase	P23	+	Q15185	2
1354	D-tyrosyl-tRNA(Tyr) deacylase 1	C20orf88	+	Q8TEA8	2
1355	Dynein light chain Tctex-type 1	DYNLT1	+	P63172	2
1356	EF-hand calcium-binding domain-containing protein 5	EFCAB5	+	A4FU69	2
1357	Egl nine homolog 1	C1orf12	+	Q9GZT9	2
1358	ESCRT-I complex subunit TSG101	TSG101	+	Q99816	2
1359	Eukaryotic translation initiation factor 4 gamma 1	EIF4F	+	E9PFM1	2
1360	Eukaryotic translation initiation factor 4E nuclear import factor 1	EIF4ENIF1	+	Q9NRA8	2

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1361	Exosome complex exonuclease RRP40	CGI-102	+	Q9NQT5	2
1362	Fragile X mental retardation 1 protein	FMR1	+	Q06787	2
1363	FTP-3	FTP3	+	P55795	2
1364	Glutathione reductase, mitochondrial	GLUR	+	P00390	2
1365	Guanine nucleotide-binding protein G(I)/G(S)/G(T) subunit beta-1	GNB1	+	P62873	2
1366	H/ACA ribonucleoprotein complex subunit 2	HSPC286	+	Q9NX24	2
1367	Heterogeneous nuclear ribonucleoprotein H	HNRNPH1	+	G8JLB6	2
1368	Heterogeneous nuclear ribonucleoprotein U	HNRNPU	+	Q00839	2
1369	High mobility group nucleosome-binding domain-containing protein 1	HMG14	+	P05114	2
1370	Histone H2A type 1	H2AFC	+	P0C0S8	2
1371	Histone H2B type 1-J	H2BFR	+	P06899	2
1372	Histone H2B	H2BFT	+	B4DR52	2
1373	Histone RNA hairpin-binding protein	HBP	+	F8W8D3	2
1374	Importin alpha Q1	KPNA4	+	O00629	2
1375	Interferon-induced protein 53	IFI53	+	P23381	2
1376	Kinesin-like protein 5	KIF23	+	Q02241	2
1377	Kinesin-like protein KIF1B	KIAA0591	+	O60333-3	2
1378	Kinesin-like protein KIF1C	KIAA0706	+	O43896	2
1379	Macrophage myristoylated alanine-rich C kinase substrate	MARCKSL1	+	P49006	2
1380	Madh6	MADH6	+	O15198	2
1381	Methyltransferase-like protein 13	CGI-01	+	Q8N6R0	2
1382	Microfibrillar-associated protein 1	MFAP1	+	P55081	2
1383	Microtubule-associated proteins 1A/1B light chain 3 beta 2	MAP1LC3B2	+	A6NCE7	2
1384	MORF4 family-associated protein 1	MRFAP1	+	Q9Y605	2
1385	N-acetyltransferase 5	NAA20	+	P61599	2
1386	NAP-1-related protein	NAP1L1	+	P55209	2
1387	Neuroendocrine-specific protein-like 2	ASYIP	+	O95197-3	2
1388	Nuclear protein localization protein 4 homolog	KIAA1499	+	Q8TAT6-2	2
1389	Nucleoplasmin-3	NPM3	+	O75607	2
1390	Oncogene c-mel	MEL	+	P61006	2
1391	p27Kip1-releasing factor from RhoA	C11orf59	+	Q6IAA8	2
1392	PAI1 RNA-binding protein 1	CGI-55	+	Q8NC51	2
1393	Phosphofurin acidic cluster sorting protein 1	KIAA1175	+	Q6VY07-2	2
1394	Phosphorylase b kinase gamma catalytic chain, testis/liver isoform	PHKG2	+	P15735	2
1395	Polynucleotide kinase Clp1	CLP1	+	E9PL17	2
1396	Prefoldin subunit 6	HKE2	+	O15212	2

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1397	Pre-mRNA-splicing factor SF3b 10 kDa subunit	SF3B10	+	Q9BWJ5	2
1398	Protein FAM83D	C20orf129	+	Q9H4H8	2
1399	Protein Njmu-R1	C17orf75	+	Q9HAS0	2
1400	Protein phosphatase 1C catalytic subunit	PPP1CC	+	P36873-2	2
1401	Protein RMD5 homolog A	RMND5A	+	Q9H871	2
1402	Putative uncharacterized protein CCNK	CCNK	+	C9JI13	2
1403	Ran-binding protein 10	KIAA1464	+	Q6VN20	2
1404	Ras-related protein Rab-10	RAB10	+	P61026	2
1405	Replication factor A protein 2	REPA2	+	P15927-3	2
1406	Secretory carrier-associated membrane protein 4	SCAMP4	+	H7BZ59	2
1407	Serine/threonine-protein phosphatase 4 regulatory subunit 1	MEG1	+	Q8TF05	2
1408	SLAIN motif-containing protein 2	KIAA1458	+	Q9P270	2
1409	Sm protein E	SNRPE	+	P62304	2
1410	Talin-2	KIAA0320	+	Q9Y4G6	2
1411	Transformer-2 protein homolog A	TRA2A	+	Q13595	2
1412	Tubulin 5 beta	TUBB4	+	P04350	2
1413	U6 snRNA-associated Sm-like protein LSm5	LSM5	+	Q9Y4Y9	2
1414	UDP--Glc:glycoprotein glucosyltransferase	GT	+	Q9NYU2	2
1415	Uncharacterized protein C19orf43	C19orf43	+	Q9BQ61	2
1416	WASH complex subunit 7	KIAA1033	+	Q2M389	2
1417	Zinc finger protein 828	C13orf8	+	Q96JM3	2
1418	28 kDa cis-Golgi SNARE p28	GOSR1	+	O95249	1
1419	60S ribosomal protein L11	RPL11	+	P62913	1
1420	Actin, cytoplasmic 2	ACTB	+	P63261	1
1421	Actin-related protein 2/3 complex subunit 3	ARC21	+	O15145	1
1422	Actin-RPV	ACTR1A	+	P61163	1
1423	Adapter-related protein complex 1 subunit beta-1	ADTB1	+	Q10567	1
1424	Aldehyde dehydrogenase 1 family, member A1	ALDH1A1	+	Q5SYQ7	1
1425	ANKRD26-like family A member 1	A26A1	+	Q6S8J7	1
1426	Apoptosis regulator Bcl-X	BCL2L	+	Q07817	1
1427	Arsenical pump-driving ATPase	ARSA	+	O43681	1
1428	Asparagine synthetase [glutamine-hydrolyzing]	ASNS	+	P08243	1
1429	AT1 receptor-associated protein	AGTRAP	+	Q6RW13	1
1430	ATP-citrate (pro-S-)-lyase	ACLY	+	E7ENH9	1
1431	BCL2-associated athanogene 2	BAG2	+	A2A296	1
1432	Beta-1,4-galactosyltransferase 1	B4GALT1	+	P15291	1
1433	C1 factor	HCF1	+	P51610	1

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1434	cDNA FLJ58476, highly similar to Poly(rC)-binding protein 2	PCBP3	+	Q15366-3	1
1435	cDNA FLJ58569, highly similar to Nucleosome assembly protein 1-like 1	hCG_2015037	+	B7Z9C2	1
1436	CF-1 50 kDa subunit	CSTF1	+	Q05048	1
1437	Coated vesicle-associated kinase of 104 kDa	CVAK104	+	Q6P3W7	1
1438	Core histone macro-H2A.1	H2AFY	+	O75367	1
1439	CTCL tumor antigen se70-2	C13orf10	+	Q5T8P6	1
1440	C-t-PAK2	PAK2	+	Q13177	1
1441	Diaphorase-1	CYB5R3	+	P00387-3	1
1442	Dimethylallyltranstransferase	FDPS	+	P14324	1
1443	DNA-binding protein NEFA	NEFA	+	P80303	1
1444	Double-stranded RNA-binding protein 76	DRBF	+	Q12906	1
1445	Dual specificity protein kinase TTK	MPS1	+	P33981	1
1446	eIF-2B GDP-GTP exchange factor subunit delta	EIF2B4	+	E7ERK9	1
1447	EIF4E-binding protein	C14orf120	+	Q8NEJ9	1
1448	Enhancer of zeste homolog 2	EZH2	+	Q15910-2	1
1449	Functional spliceosome-associated protein 79	C22orf19	+	Q13769	1
1450	Genetic suppressor element 1	GSE1	+	Q14687	1
1451	Glycine- and tyrosine-rich RNA-binding protein	HNRPQ	+	O60506	1
1452	Glycosylation-inhibiting factor	GLIF	+	P14174	1
1453	H/ACA ribonucleoprotein complex subunit 3	NOLA3	+	Q9NPE3	1
1454	Heat shock 70 kDa protein 6	HSP70B	+	P17066	1
1455	Hepatocellular carcinoma-associated antigen 59	C9orf78	+	Q9NZ63	1
1456	Heterogeneous nuclear ribonucleoprotein A1-like 2	HNRNPA1L	+	Q32P51	1
1457	Heterogeneous nuclear ribonucleoprotein K	HNRNPK	+	P61978	1
1458	High mobility group AT-hook protein 1	HMGA1	+	H7BYM6	1
1459	Histone H2A type 1-C	H2AFL	+	Q93077	1
1460	Histone H3	HIST2H3PS2	+	Q5TEC6	1
1461	Huntingtin yeast partner K	C15orf63	+	Q9NX55	1
1462	Kinesin-like protein KIF18B	KIF18B	+	Q86Y91-4	1
1463	Kinesin-like protein KIF6	C6orf102	+	Q6ZMV9	1
1464	Lamina-associated polypeptide 2, isoforms beta/gamma	LAP2	+	P42167	1
1465	Lark homolog	RBM4	+	Q9BWF3	1
1466	Leucine-rich repeat protein SHOC-2	KIAA0862	+	Q9UQ13	1
1467	LIM and senescent cell antigen-like-containing domain protein 2	LIMS2	+	P48059-2	1
1468	L-lactate dehydrogenase A-like 6B	LDHAL6	+	Q9BYZ2	1
1469	Major histocompatibility complex class II DP beta 1	DADB-22N23.4-003	+	A2BEU2	1

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1470	Mitotic-spindle organizing protein associated with a ring of gamma-tubulin 1	C13orf37	+	Q08AG7	1
1471	Mitsugumin-23	TMEM109	+	Q9BVC6	1
1472	Monocarboxylate transporter 1	MCT1	+	P53985	1
1473	Pantophysin	SYPL	+	Q16563	1
1474	Partner of Y14 and mago	PYM	+	Q9BRP8	1
1475	Phosphoribosyl pyrophosphate synthase I	PRPS1	+	P60891	1
1476	Pituitary tumor-transforming gene 1 protein-interacting protein	C21orf1	+	P53801	1
1477	Plexin-4	PLXN4	+	P51805	1
1478	Programmed cell death protein 2-like	PDCD2L	+	Q9BRP1	1
1479	Protein cornichon homolog 4	CNIH4	+	Q9P003	1
1480	Protein DB83	DB83	+	P57088	1
1481	Protein KRI1 homolog	KRI1	+	Q8N9T8	1
1482	Protein mago nashi homolog 2	MAGOH2	+	Q96A72	1
1483	Protein transport protein Sec24A	SEC24A	+	O95486	1
1484	Pumilio homolog 1	KIAA0099	+	Q14671	1
1485	Putative heat shock protein HSP 90-beta 4	HSP90AB4P	+	Q58FF6	1
1486	Putative Speedy protein E3	SPDYE3	+	A6NKU9	1
1487	Putative uncharacterized protein FMR1	FMR1	+	Q06787-7	1
1488	Putative uncharacterized protein HNRPLL	HNRPLL	+	C9IYN3	1
1489	Putative uncharacterized protein RANBP1	RANBP1	+	C9JDM3	1
1490	Ran-binding protein 1	RANBP1	+	P43487	1
1491	Ras-related protein Rab-4A	RAB4	+	P20338	1
1492	Ribonuclease P/MRP protein subunit POP5	AD-008	+	Q969H6	1
1493	Ribose-phosphate pyrophosphokinase	PRPS1	+	B4DNL6	1
1494	Sorting nexin-5	SNX5	+	Q9Y5X3	1
1495	STAF28	TAF10	+	Q12962	1
1496	Transmembrane protein 165	TMEM165	+	Q9HC07	1
1497	Tubulin alpha chain-like 3	TUBAL3	+	A6NHL2	1
1498	Tubulin beta-2 chain	TUBB2C	+	P68371	1
1499	Ubiquitin carboxyl-terminal hydrolase L5	RP11-101E13.2-006	+	Q5LJA9	1
1500	Ubiquitin-fold modifier 1	BM-002	+	P61960	1
1501	Vacuolar protein sorting-associated protein 45	VPS45	+	Q9NRW7	1
1502	Ankyrin repeat domain-containing protein 17	ANKRD17	-	O75179	10
1503	Chromokinesin-A	KIF4	-	O95239	8
1504	Dolichyl-diphosphooligosaccharide--protein glycosyltransferase 63 kDa subunit	RPN2	-	P04844	8

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1505	E3 ligase for inhibin receptor	HECTD1	-	Q9ULT8	8
1506	Septin-11	11-Sep	-	D6RER5	8
1507	Alpha-2-macroglobulin receptor	A2MR	-	Q07954	7
1508	Epithelial cell-transforming sequence 2 oncogene	ECT2	-	Q9H8V3-3	7
1509	133 kDa nucleoporin	NUP133	-	Q8WUM0	6
1510	CFR-1	CFR1	-	Q92896-2	6
1511	Cysteine and glycine-rich protein 2	CSRP2	-	F8VW96	6
1512	Endophilin I	PACSIN3	-	Q9UKS6	6
1513	Endoplasmic reticulum resident protein 46	TLP46	-	Q8NBS9	6
1514	Ensconsin	MAP7	-	Q14244	6
1515	Metavinculin	VCL	-	P18206	6
1516	Nuclear localized protein 1	FKSG26	-	Q9BQ70	6
1517	Protein KU002155	SPTY2D1	-	Q68D10	6
1518	RNA-binding motif protein 12	HRIHFB2091	-	Q9NTZ6	6
1519	Surfeit locus protein 6	SURF6	-	O75683	6
1520	Thymocyte nuclear protein 1	HSPC144	-	Q9P016	6
1521	61E3.4	ATX	-	I3L0C1	5
1522	Activator-recruited cofactor 150 kDa component	ARC150	-	O60244	5
1523	Alkylated DNA repair protein alkB homolog 5	ABH5	-	Q6P6C2-2	5
1524	Antigen NY-CO-1	SDCCAG1	-	O60524	5
1525	Apoptosis regulator BAX	BAX	-	Q07812-2	5
1526	ATP synthase subunit beta, mitochondrial	ATP5B	-	P06576	5
1527	B5	ITM1	-	P46977	5
1528	Calmodulin-regulated spectrin-associated protein 1	CAMSAP1	-	Q5T5Y3-3	5
1529	CDKN2A-interacting protein	CARF	-	Q9NXV6	5
1530	Centromere/kinetochore protein zw10 homolog	ZW10	-	O43264	5
1531	Coiled-coil domain-containing protein 9	CCDC9	-	Q9Y3X0	5
1532	COP9 signalosome complex subunit 5	COPS5	-	Q92905	5
1533	Down-regulated in human cancers protein	DRHC	-	P42694	5
1534	HPK/GCK-like kinase HGK	HGK	-	G5E948	5
1535	Integrator complex subunit 3	C1orf193	-	Q68E01	5
1536	Intron-binding protein aquarius	AQR	-	O60306	5
1537	MAP7 domain-containing protein 3	MAP7D3	-	Q8IWC1	5
1538	MHC class I region proline-rich protein CAT53	CAT53	-	Q96QC0	5
1539	Myc-associated zinc finger protein	MAZ	-	G5E927	5
1540	Nucleolar protein 17 homolog	NOP17	-	Q9NWS0	5
1541	Putative splicing factor YT521	KIAA1966	-	Q96MU7	5
1542	Renal carcinoma antigen NY-REN-37	ZC3H14	-	Q6PJT7	5

Continuing Appendix 1. List of proteins identified by MS/MS in global protein subcellular distribution analysis on a global proteomic scale

1543	tRNA (adenine-N(1)-)-methyltransferase non-catalytic subunit TRM6	CGI-09	-	Q9UJA5	5
1544	[Histone-H3]-lysine-36 demethylase 1A	CXXC8	-	Q9Y2K7	4
1545	100 kDa coated vesicle protein C	ADTAB	-	O94973-2	4
1546	153 kDa nucleoporin	NUP153	-	F6QR24	4
1547	17-beta-hydroxysteroid dehydrogenase 4	EDH17B4	-	F5HE57	4
1548	5-azacytidine-induced protein 1	AZI1	-	Q9UPN4	4
1549	A121	EIF1	-	P41567	4
1550	ABP-280-like protein	ABPL	-	Q14315	4
1551	Acid finger protein	RNF95	-	Q12899	4
1552	Activator-recruited cofactor 100 kDa component	ARC100	-	O75448	4
1553	Adenine nucleotide translocator 2	ANT2	-	P05141	4
1554	Alba-like protein C9orf23	C9orf23	-	Q8N5L8	4
1555	ALL1-fused gene from chromosome 4p12 protein	AF4P12	-	F5GX82	4
1556	Anaphase-promoting complex subunit 8	ANAPC8	-	Q9UJX2	4
1557	ARF GTPase-activating protein GIT1	GIT1	-	Q9Y2X7-3	4
1558	Arginine/serine-rich coiled-coil protein 2	RSRC2	-	Q7L4I2	4
1559	Armadillo repeat-containing protein 8	ARMC8	-	Q8IUR7	4
1560	Aryl hydrocarbon receptor-associated protein 3	ARA3	-	Q9Y6Y0	4
1561	Ashwin	C2orf49	-	Q9BVC5	4
1562	ATP synthase subunit alpha, mitochondrial	ATP5A	-	P25705	4
1563	ATP-dependent helicase SMARCA1	SMARCA1	-	P28370	4
1564	Biorientation of chromosomes in cell division protein 1-like	BOD1L	-	Q8NFC6	4
1565	Block of proliferation 1 protein	BOP1	-	Q14137	4
1566	Box C/D snoRNA protein 1	BCD1	-	Q9NWK9	4
1567	Brix domain-containing protein 1	BXDC1	-	Q9H7B2	4
1568	C128	POLR3B	-	Q9NW08	4
1569	Caspase-like protein ESPL1	ESP1	-	Q14674	4
1570	Cell division cycle protein 20 homolog	CDC20	-	Q12834	4
1571	Cell division protein kinase 2	CDK2	-	G3V5T9	4
1572	Component of oligomeric Golgi complex 4	COG4	-	Q9H9E3	4
1573	Cytoplasmic dynein 2 heavy chain	DHC1B	-	Q8NCM8-2	4
1574	Cytosolic NADP-isocitrate dehydrogenase	IDH1	-	O75874	4
1575	Damage-specific DNA-binding protein 2	DDB2	-	Q92466	4
1576	DLG4	DLG4	-	Q15334	4
1577	DNA polymerase epsilon catalytic subunit A	POLE	-	E7ETS8	4
1578	DNA-directed RNA polymerase II 23 kDa polypeptide	POLR2E	-	P19388	4
1579	DNA-directed RNA polymerase III subunit D	BN51	-	P05423	4

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1580	Heat shock 70 kDa protein 4L	APG1	-	E7ES43	4
1581	Hyaluronan mediated motility receptor	HMMR	-	O75330-3	4
1582	Just another zinc finger protein	JAZ	-	Q9UL40-2	4
1583	Kinetochore-associated protein 1	KIAA0166	-	P50748	4
1584	Lamin-B1	LMN2	-	P20700	4
1585	MET18 homolog	MMS19	-	Q96T76	4
1586	Mitochondrial import receptor subunit TOM34	TOMM34	-	Q15785	4
1587	Myosin phosphatase-targeting subunit 1	MBS	-	F5H1B6	4
1588	p105-Rb	RB1	-	P06400	4
1589	p190-B	ARHGAP5	-	Q13017	4
1590	Phosphohydroxythreonine aminotransferase	PSA	-	Q9Y617	4
1591	Prefoldin subunit 2	HSPC231	-	Q9UHV9	4
1592	Putative uncharacterized protein REPIN1	REPIN1	-	C9J3L7	4
1593	Pyridoxal-dependent decarboxylase domain-containing protein 1	KIAA0251	-	H3BND4	4
1594	Septin and tuftelin-interacting protein 1	HSPC006	-	Q9UBB9	4
1595	Signal recognition particle 19 kDa protein	SRP19	-	P09132	4
1596	Swd2	UNQ9342/PRO34047	-	Q6UXN9	4
1597	Synaptic glycoprotein SC2	GPSN2	-	Q9NZ01	4
1598	Uncharacterized protein KIAA1797	KIAA1797	-	Q5VW36	4
1599	Zinc finger CCCH domain-containing protein 8	ZC3H8	-	Q8N5P1	4
1600	Zinc finger protein 91 homolog	FKSG11	-	Q96JP5	4
1601	20 kDa nuclear cap-binding protein	CBP20	-	P52298	3
1602	85 kDa lysosomal membrane sialoglycoprotein	CD36L2	-	Q14108	3
1603	Abeta-degrading protease	IDE	-	P14735	3
1604	AC40	POLR1C	-	O15160	3
1605	Activity-dependent neuroprotective protein	ADNP	-	Q9H2P0	3
1606	Acute-phase response factor	APRF	-	P40763	3
1607	ADP-ribosylation factor GTPase-activating protein 2	ARFGAP2	-	Q8N6H7	3
1608	ADP-ribosylation factor-like protein 1	ARL1	-	F8VYN9	3
1609	Aldehyde dehydrogenase family 7 member A1	ALDH7A1	-	P49419	3
1610	Alpha-2-macroglobulin receptor-associated protein	A2MRAP	-	P30533	3
1611	Anaphase-promoting complex subunit 2	ANAPC2	-	Q9UJX6	3
1612	Annexin A1	ANX1	-	P04083	3
1613	Antigen NY-CO-16	SDCCAG16	-	Q9BVJ6	3
1614	Arginine and glutamate-rich protein 1	ARGLU1	-	Q9NWB6	3
1615	ATP-dependent RNA helicase DDX55	DDX55	-	Q8NHQ9	3
1616	Band 4.1-like protein 2	EPB41L2	-	O43491	3

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1617	BPG-dependent PGAM 1	CDABP0006	-	P18669	3
1618	Cancer susceptibility candidate gene 3 protein	CASC3	-	O15234	3
1619	CAP-Gly domain-containing linker protein 1	CLIP1	-	P30622	3
1620	Cavin-1	FKSG13	-	Q6NZI2	3
1621	CDC2-related protein kinase 5	CDC2L	-	Q14004	3
1622	Cell cycle regulator Mat89Bb homolog	C12orf11	-	Q9NVM9	3
1623	Cell division cycle protein 73 homolog	C1orf28	-	Q6P1J9	3
1624	Cell proliferation-inducing gene 26 protein	CGI-31	-	Q9Y320	3
1625	Centrosomal protein of 120 kDa	CCDC100	-	Q8N960	3
1626	Centrosomal protein of 55 kDa	C10orf3	-	Q53EZ4	3
1627	CG-1 antigen	CG1	-	Q86UP2	3
1628	Coated vesicle-associated kinase of 90 kDa	CVAK90	-	Q96KG9	3
1629	Coiled-coil domain-containing protein 131	CCDC131	-	O60293	3
1630	Coiled-coil domain-containing protein 137	CCDC137	-	I3L0U5	3
1631	COMM domain-containing protein 9	COMMD9	-	Q9P000	3
1632	Component of oligomeric Golgi complex 1	COG1	-	Q8WTW3	3
1633	CREB-binding protein	CBP	-	Q92793	3
1634	Cyclin-dependent kinase inhibitor 2B-related protein	P15RS	-	Q96P16	3
1635	DEAH box protein 37	DDX37	-	Q8IY37	3
1636	DNA-directed RNA polymerase II 33 kDa polypeptide	A-152E5.7	-	P19387	3
1637	DNA-directed RNA polymerase II subunit H	POLR2H	-	P52434	3
1638	DNA-directed RNA polymerase III subunit C	POLR3C	-	Q9BUI4	3
1639	Echinoderm microtubule associated protein like 3, isoform CRA_e	EML3	-	B7WPE2	3
1640	Elongator complex protein 2	ELP2	-	E7EP23	3
1641	Elongin 110 kDa subunit	MSTP059	-	Q14241	3
1642	Endoribonuclease Dicer	DICER	-	Q9UPY3	3
1643	Erythrocyte band 7 integral membrane protein	BND7	-	P27105	3
1644	Female-lethal(2)D homolog	KIAA0105	-	Q15007	3
1645	G patch domain-containing protein 4	GPATC4	-	Q5T3I0-3	3
1646	HBEBP2-binding protein C	C14orf151	-	Q27J81	3
1647	HEAT repeat-containing protein 1	BAP28	-	Q9H583	3
1648	Hepatocellular carcinoma-susceptibility protein 3	HCCA3	-	Q969U7	3
1649	Histone lysine demethylase PHF8	KIAA1111	-	Q9UPP1	3
1650	HpaII tiny fragments locus 9c protein	HTF9C	-	F2Z2W7	3
1651	hTREX84	HPR1	-	Q96FV9	3
1652	Human cervical cancer suppressor gene 4 protein	HCCS4	-	Q9NYL2	3
1653	Integrator complex subunit 5	INTS5	-	Q6P9B9	3
1654	Integrator complex subunit 7	C1orf73	-	Q9NVH2	3

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1655	Interferon-induced protein with tetratricopeptide repeats 5	IFIT5	-	Q13325	3
1656	Intramembrane protease 1	H13	-	Q8TCT9	3
1657	Kinesin-like protein 1	EG5	-	P52732	3
1658	Membrane-associated progesterone receptor component 1	HPR6.6	-	O00264	3
1659	Methenyltetrahydrofolate synthase domain-containing protein	MTHFSD	-	E9PAM1	3
1660	Microsomal glutathione S-transferase 1	GST12	-	P10620	3
1661	MIR-interacting saposin-like protein	CNPY2	-	F8W031	3
1662	Modulator of estrogen-induced transcription	MET	-	Q9NWH9	3
1663	Myosin-IXb	MYO9B	-	Q13459	3
1664	Nuclear protein NHN1	NHN1	-	E7ERS3	3
1665	Nuclear transcription factor, X box-binding protein 1	NFX1	-	Q12986	3
1666	Nucleolar MIF4G domain-containing protein 1	C7orf3	-	Q5C9Z4	3
1667	Nucleoporin SEH1	SEC13L	-	Q96EE3-1	3
1668	OMPdecase	OK/SW-cl.21	-	P11172	3
1669	Oxysterol-binding protein-related protein 3	KIAA0704	-	Q9H4L5	3
1670	p150 target of rapamycin (TOR)-scaffold protein	KIAA1303	-	Q8N122	3
1671	p160 ROCK-1	ROCK1	-	Q13464	3
1672	Progesterone-induced-blocking factor 1	C13orf24	-	Q8WXW3	3
1673	Proteasomal ATPase-associated factor 1	PAAF1	-	Q9BRP4	3
1674	Protein C20orf11	C20orf11	-	Q9NWU2	3
1675	Protein H105e3	H105E3	-	Q15738	3
1676	Protein transport protein Sec61 subunit beta	SEC61B	-	P60468	3
1677	Protein TSSC4	TSSC4	-	Q9Y5U2	3
1678	Proto-oncogene c-RAF	RAF	-	P04049	3
1679	Purine-rich element-binding protein B	PURB	-	Q96QR8	3
1680	Putative uncharacterized protein ABI1	ABI1	-	A6NFN2	3
1681	Rab3 GTPase-activating protein 150 kDa subunit	KIAA0839	-	Q9H2M9	3
1682	Ras-related protein Rab-6A	RAB6	-	P20340	3
1683	Rho GDP-dissociation inhibitor 1	ARHGDIA	-	P52565	3
1684	Ribonuclease P protein subunit p40	RNASEP1	-	O75818	3
1685	RNA-binding motif protein 7	RBM7	-	Q9Y580	3
1686	RNA-binding motif, single-stranded-interacting protein 2	RBMS2	-	Q15434	3
1687	Scaffold attachment factor B2	KIAA0138	-	Q14151	3
1688	Slit homolog 2 protein	SLIL3	-	O94813	3
1689	SM22-alpha homolog	CDABP0035	-	P37802	3
1690	Spermatid perinuclear RNA-binding protein	SPNR	-	Q96SI9	3

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1691	Synaptobrevin homolog YKT6	YKT6	-	O15498	3
1692	Testis-specific Y-encoded-like protein 1	TSPYL	-	Q9H0U9	3
1693	Tropomodulin-3	TMOD3	-	Q9NYL9	3
1694	Uncharacterized protein C8orf41	C8orf41	-	Q6NXR4	3
1695	UV excision repair protein RAD23 homolog B	RAD23B	-	P54727	3
1696	Vasodilator-stimulated phosphoprotein	VASP	-	P50552	3
1697	WD repeat-containing protein 33	WDC146	-	Q9C0J8	3
1698	Zinc finger CCCH domain-containing protein 7A	HSPC055	-	Q8IWR0	3
1699	17-beta-hydroxysteroid dehydrogenase 11	DHRS8	-	Q8NBQ5	2
1700	17-beta-hydroxysteroid dehydrogenase 12	HSD17B12	-	Q53GQ0	2
1701	19 kDa FK506-binding protein	FKBP11	-	Q9NYL4	2
1702	20 kDa CGG-binding protein	CGGBP	-	Q9UFW8	2
1703	20-alpha-hydroxysteroid dehydrogenase	AKR1C1	-	Q04828	2
1704	26S proteasome non-ATPase regulatory subunit 10	PSMD10	-	O75832	2
1705	350/400 kDa PCAF-associated factor	PAF400	-	Q9Y4A5	2
1706	372 kDa Golgi complex-associated protein	GOLGB1	-	E7EP74	2
1707	38 kDa FK506-binding protein	FKBP38	-	Q14318-2	2
1708	3-methylcrotonyl-CoA carboxylase 1	MCCA	-	Q96RQ3	2
1709	4F2 light chain	CD98LC	-	Q01650	2
1710	50 kDa nucleoporin	NPAP60L	-	Q9UKX7	2
1711	60S ribosomal protein L7-like 1	RPL7L1	-	Q6DKI1	2
1712	67 kDa calelectrin	ANX6	-	P08133	2
1713	72 kDa Tat-interacting protein	HT2A	-	Q13049	2
1714	7-dehydrocholesterol reductase	D7SR	-	Q9UBM7	2
1715	96 kDa nucleoporin	ADAR2	-	P52948	2
1716	Acidic leucine-rich nuclear phosphoprotein 32 family member B	ANP32B	-	Q92688	2
1717	Actin-related protein 2/3 complex subunit 5	ARC16	-	O15511-2	2
1718	Activator-recruited cofactor 240 kDa component	ARC240	-	Q93074-2	2
1719	Acyl-coenzyme A:cholesterol acyltransferase 1	ACACT	-	P35610	2
1720	Adaptor protein TKS5	FISH	-	Q5TCZ1	2
1721	Adipocyte plasma membrane-associated protein	APMAP	-	Q9HDC9	2
1722	ADP-ribosylation factor 1 GTPase-activating protein	ARF1GAP	-	Q8N6T3-2	2
1723	Alanyl aminopeptidase	ANPEP	-	P15144	2
1724	Alpha-SGT	SGT	-	O43765	2
1725	Anaphase-promoting complex subunit 5	ANAPC5	-	Q9UJX4	2
1726	Anaphase-promoting complex subunit 6	ANAPC6	-	Q13042	2
1727	APOBEC1-like	APOBEC1L	-	Q9NRW3	2
1728	ATP-dependent DNA helicase Q4	RECQ4	-	O94761	2

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1729	ATP-dependent helicase 1	KIAA1122	-	Q9H4L7-2	2
1730	ATP-dependent helicase CHD6	CHD5	-	Q8TD26	2
1731	ATP-dependent helicase ERCC6	CSB	-	Q03468	2
1732	ATP-dependent helicase GTF2F2	GTF2F2	-	P13984	2
1733	Aurora- and IPL1-like midbody-associated protein 1	AIK2	-	Q96GD4	2
1734	AVO3 homolog	KIAA1999	-	Q6R327-3	2
1735	Axonemal beta dynein heavy chain 9	DNAH17L	-	Q9NYC9	2
1736	Baboon M7 virus receptor	ASCT2	-	Q15758	2
1737	Bardet-Biedl syndrome 14 protein	BBS14	-	O15078	2
1738	Basic transcription factor 2 44 kDa subunit	BTF2P44	-	Q13888	2
1739	Beta-1 metal-binding globulin	PRO1400	-	P02787	2
1740	Brain-expressed RING finger protein	BERP	-	O75382	2
1741	BRCA1-A complex subunit RAP80	RAP80	-	F8VQY2	2
1742	Brefeldin A-inhibited guanine nucleotide-exchange protein 2	ARFGEF2	-	Q9Y6D5	2
1743	BRG1-associated factor 170	BAF170	-	F8VXC8	2
1744	CAF1-like protein	CALIF	-	Q9UFF9	2
1745	Calgranulin-B	CAGB	-	P06702	2
1746	Carbon catabolite repressor protein 4 homolog	CCR4	-	Q9ULM6	2
1747	Casein kinase I isoform delta	CSNK1D	-	P48730	2
1748	Catenin gamma	CTNNG	-	F5GWP8	2
1749	cDNA FLJ34439 fis, clone HLUNG2001146, highly similar to Splicing factor, arginine/serine-rich 12	SFRS12	-	Q8WXA9-2	2
1750	cDNA FLJ53622, highly similar to Hematological and neurological expressed1-like protein	C16orf34	-	B4DLH4	2
1751	cDNA FLJ55884, highly similar to RNA-binding protein 16	CCAP7	-	B7Z888	2
1752	cDNA FLJ59739, highly similar to Protein transport protein Sec61 subunit alpha isoform 1	SEC61A	-	B4DR61	2
1753	cDNA FLJ60834, highly similar to Engulfment and cell motility protein 2 (CED-12 homolog A)	CED12A	-	B4DRL5	2
1754	Cell cycle autoantigen SG2NA	GS2NA	-	Q13033-2	2
1755	Cell proliferation-inducing gene 5 protein	EMP	-	E7ESC7	2
1756	Cellugyrin	SYNGR2	-	O43760	2
1757	Cellular transcription factor ILF-1	FOXK2	-	Q01167	2
1758	Chloride channel protein 3	CLCN3	-	P51790-2	2
1759	Ciliary rootlet coiled-coil protein	CROCC	-	Q5TZA2	2
1760	Cirhin	CIRH1A	-	H3BSH7	2
1761	Cleavage and polyadenylation-specific factor 3-like protein	CPSF3L	-	Q5TA45	2
1762	Coat protein GPP34	GOLPH3	-	Q9H4A6	2

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1763	Cofactor required for Sp1 transcriptional activation subunit 8	CRSP34	-	Q6P2C8	2
1764	Coiled-coil domain-containing protein 50	C3orf6	-	Q8IVM0-2	2
1765	Collapsin response mediator protein 5	CRMP5	-	Q9BPU6	2
1766	Component of oligomeric Golgi complex 2	COG2	-	Q14746	2
1767	Constitutive coactivator of peroxisome proliferator-activated receptor gamma	CCPG	-	F5GY05	2
1768	Constitutive coactivator of PPAR-gamma-like protein 2	CXorf17	-	F8W881	2
1769	COP9 signalosome complex subunit 7a	COPS7A	-	Q9UBW8	2
1770	CpG-binding protein	CFP1	-	Q9P0U4-2	2
1771	CTP synthase 1	CTPS	-	P17812	2
1772	Cullin-4B	CUL4B	-	Q13620	2
1773	Cutaneous T-cell lymphoma-associated antigen se33-1	NP220	-	Q14966-4	2
1774	CWF19-like protein 1	CWF19L1	-	Q69YN2	2
1775	Cyclin-A2	CCN1	-	P20248	2
1776	Cyclin-E-binding protein 1	CEB1	-	Q9UII4	2
1777	Daxx	BING2	-	Q9UER7	2
1778	Delta transcription factor	INO80S	-	P25490	2
1779	Delta-aminolevulinic acid dehydratase	ALAD	-	P13716	2
1780	Deubiquitinating enzyme 16	MSTP039	-	Q9Y5T5	2
1781	DNA polymerase delta subunit 3	KIAA0039	-	Q15054	2
1782	DNA topoisomerase 2-binding protein 1	KIAA0259	-	Q92547	2
1783	DNA-binding protein/plasminogen activator inhibitor 1 regulator	HIP116A	-	Q14527	2
1784	DNA-directed RNA polymerase III 80 kDa polypeptide	KIAA1452	-	Q9NVU0	2
1785	DNA-directed RNA polymerase III subunit F	POLR3F	-	Q9H1D9	2
1786	DnaJ homolog subfamily B member 6	DNAJB6	-	O75190	2
1787	DnaJ homolog subfamily C member 13	DNAJC13	-	O75165	2
1788	E3 ubiquitin-protein ligase RING1	RING1	-	Q06587	2
1789	Endocrine regulatory protein	HRIHFB2436	-	Q5VUA4	2
1790	Endoplasmic reticulum lipid raft-associated protein 1	C10orf69	-	O75477	2
1791	Endoplasmic reticulum-Golgi intermediate compartment protein 1	ERGIC1	-	Q969X5	2
1792	Ephrin type-A receptor 2	ECK	-	P29317	2
1793	Epidermal growth factor receptor	EGFR	-	P00533	2
1794	Eukaryotic translation initiation factor 2-alpha kinase 4	EIF2AK4	-	Q9P2K8	2
1795	Exocyst complex 84 kDa subunit	EXOC8	-	Q8IYI6	2
1796	Exocyst complex component 1	BM-012	-	Q9NV70	2
1797	Exocyst complex component 4	EXOC4	-	Q96A65	2
1798	Formin-binding protein 30	FBP30	-	Q8N3X1-2	2

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1799	G2 and S phase-expressed protein 1	GTSE1	-	Q9NYZ3	2
1800	Gamma-soluble NSF attachment protein	NAPG	-	Q99747	2
1801	Gamma-tubulin complex component 6	GCP6	-	Q96RT7	2
1802	GC-rich sequence DNA-binding factor	C2orf3	-	P16383	2
1803	GDP-mannose pyrophosphorylase A	GMPPA	-	Q96IJ6-2	2
1804	Glioma tumor suppressor candidate region gene 2 protein	GLTSCR2	-	Q9NZM5	2
1805	Glycogen synthase kinase-3 beta	GSK3B	-	P49841-2	2
1806	Glycoprotein GP36b	C5orf8	-	Q12907	2
1807	GPN-loop GTPase 1	GPN1	-	Q9HCN4	2
1808	GTPase-activating protein RAB7	TBC1D15	-	Q8TC07	2
1809	GTP-binding protein 9	GTPBP9	-	Q9NTK5	2
1810	Guanidinoacetate N-methyltransferase	GAMT	-	Q14353	2
1811	Hairy cell leukemia protein 1	HUSSY-29	-	O95478	2
1812	HAUS augmin-like complex subunit 6	DGT6	-	Q7Z4H7	2
1813	HEAT repeat-containing protein 3	HEATR3	-	Q7Z4Q2	2
1814	Heat shock protein 75 kDa, mitochondrial	HSP75	-	Q12931	2
1815	HepA-related protein	HARP	-	Q9NZC9	2
1816	Hepatocellular carcinoma-associated antigen 56	HCA56	-	P41214	2
1817	Histone acetyltransferase type B catalytic subunit	HAT1	-	O14929	2
1818	hSGT2	KIAA1230	-	Q86W92	2
1819	Huntingtin	HD	-	P42858	2
1820	Importin-8	IPO8	-	O15397	2
1821	Inositol polyphosphate 5-phosphatase OCRL-1	INPP5F	-	Q01968	2
1822	Integrin alpha-V	ITGAV	-	P06756	2
1823	Intercellular adhesion molecule 1	ICAM1	-	P05362	2
1824	Interferon-related developmental regulator 1	IFRD1	-	O00458	2
1825	Lung cancer metastasis-related protein 1	LCMR1	-	A0JLT2	2
1826	Lysophospholipase	PLB	-	Q6P1J6	2
1827	Lysyl oxidase homolog 1	LOXL	-	Q08397	2
1828	MacGAP	ARHGAP18	-	Q8N392	2
1829	Malic enzyme 1	ME1	-	P48163	2
1830	Matriptase-3	TMPRSS7	-	Q7RTY8	2
1831	Mediator complex subunit 12-like protein	KIAA1635	-	Q86YW9	2
1832	Mediator complex subunit 16	DRIP92	-	Q9Y2X0	2
1833	Mediator complex subunit 20	MED20	-	Q9H944	2
1834	Mediator complex subunit 22	MED22	-	Q15528	2
1835	Metastasis-associated protein MTA3	KIAA1266	-	Q9BTC8	2
1836	Methionine aminopeptidase 1	KIAA0094	-	P53582	2

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1837	mRNA export factor	MRNP41	-	P78406	2
1838	Muskelin	MKLN1	-	Q9UL63	2
1839	Myosin heavy chain 14	FP17425	-	G8JLL9	2
1840	NADPH--cytochrome P450 reductase	CYPOR	-	P16435	2
1841	NHS-like protein 1	C6orf63	-	Q5SYE7	2
1842	Nicotinamide phosphoribosyltransferase	NAMPT	-	P43490	2
1843	NOC2-like protein	NOC2L	-	Q9Y3T9	2
1844	Nuclear distribution protein C homolog	NUDC	-	Q9Y266	2
1845	Opa-interacting protein 1	OIP1	-	Q15654	2
1846	Otoancorin	OTOA	-	Q7RTW8	2
1847	Outer mitochondrial membrane protein porin 2	VDAC2	-	P45880-1	2
1848	p164 ROCK-2	KIAA0619	-	O75116	2
1849	p21-activated kinase 4	KIAA1142	-	O96013	2
1850	Plasma membrane calcium ATPase isoform 1	ATP2B1	-	P20020	2
1851	Polyposis locus protein 1	C5orf18	-	Q00765	2
1852	PP2A B subunit isoform B56-gamma	KIAA0044	-	F5GWP3	2
1853	PP2A subunit A isoform PR65-beta	PPP2R1B	-	P30154-2	2
1854	Pre-mRNA-splicing factor SRP46	SFRS2B	-	Q9BRL6	2
1855	PRKC apoptosis WT1 regulator protein	PAR4	-	Q96IZ0	2
1856	Probable cation-transporting ATPase 13A1	ATP13A	-	Q9HD20	2
1857	Prosaposin	PSAP	-	B1AVU8	2
1858	Protein ALAESM	C1orf122	-	Q6ZSJ8	2
1859	Protein FAM195A	C16orf14	-	F5H0Z5	2
1860	Protein Hook homolog 3	HOOK3	-	Q86VS8	2
1861	Protein kinase C and casein kinase substrate in neurons protein 2	PACSIN2	-	Q9UNF0	2
1862	Protein NNX3	C19orf2	-	O94763	2
1863	Protein phosphatase T	PPP5	-	P53041	2
1864	Protein-glutamine gamma-glutamyltransferase 2	TGM2	-	P21980	2
1865	Puromycin-sensitive aminopeptidase	NPEPPS	-	P55786	2
1866	Putative uncharacterized protein CEP78	CEP78	-	Q5JTW2-2	2
1867	Putative uncharacterized protein SEPT10	hCG_27895	-	B5ME97	2
1868	Pyrroline-5-carboxylate reductase 3	PYCRL	-	Q53H96	2
1869	R3H and coiled-coil domain-containing protein 1	R3HCC1	-	Q9Y3T6	2
1870	R3H domain-containing protein 1	KIAA0029	-	E9PBB4	2
1871	RAS protein activator like 2	RASAL2	-	F8W755	2
1872	Ras-related protein Ral-A	RAL	-	P11233	2
1873	Ribonuclease P protein subunit p20	POP7	-	O75817	2
1874	Ribosomal RNA-processing protein 15	CGI-115	-	Q9Y3B9	2

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1875	RISC-loading complex subunit TARBP2	TARBP2	-	Q15633	2
1876	RNA-binding motif protein 34	KIAA0117	-	P42696	2
1877	RNA-binding protein Musashi homolog 2	MSI2	-	Q96DH6	2
1878	RNA-binding protein RBP	SF4	-	Q8IWZ8	2
1879	Round spermatid basic protein 1-like protein	RSBN1L	-	Q6PCB5	2
1880	Schlafen family member 11	SLFN11	-	Q7Z7L1	2
1881	Sentrin/SUMO-specific protease SENP3	SENP3	-	Q9H4L4	2
1882	Serine/threonine-protein phosphatase 6 catalytic subunit	PPP6	-	O00743-3	2
1883	SH3 domain-binding protein 1	SH3BP1	-	Q9Y3L3	2
1884	Signal sequence receptor subunit delta	SSR4	-	P51571	2
1885	Sodium/potassium-dependent ATPase subunit beta-3	ATP1B3	-	P54709	2
1886	Sorting nexin-2	SNX2	-	O60749	2
1887	Spartin	KIAA0610	-	Q8N0X7	2
1888	Structural maintenance of chromosomes flexible hinge domain-containing protein 1	KIAA0650	-	A6NHR9	2
1889	Synaptic inositol-1,4,5-trisphosphate 5-phosphatase 2	KIAA0348	-	O15056	2
1890	Tau-tubulin kinase	TTBK2	-	Q8IWY7	2
1891	T-cell protein-tyrosine phosphatase	PTPN2	-	P17706	2
1892	Telomeric DNA-binding protein	TERF2	-	Q15554	2
1893	Terminal uridylyltransferase 7	HS2	-	Q5VYS8-2	2
1894	TNPO2 variant protein	TNPO2 variant protein	-	Q4LE60	2
1895	Transaldolase	TAL	-	P37837	2
1896	Translin-associated factor X	TRAX	-	Q99598	2
1897	Transmembrane 9 superfamily member 4	KIAA0255	-	Q92544	2
1898	Tuberin	TSC2	-	P49815	2
1899	U11/U12 small nuclear ribonucleoprotein 31 kDa protein	ZCRB1	-	Q8TBF4	2
1900	U6 snRNA-associated Sm-like protein LSm7	LSM7	-	Q9UK45	2
1901	Uncharacterized protein C14orf48	C14orf48	-	Q8NCU1	2
1902	Uncharacterized protein C3orf17	C3orf17	-	Q6NW34	2
1903	Uncharacterized protein KIAA0802	KIAA0802	-	Q9Y4B5	2
1904	UPF0428 protein CXorf56	CXorf56	-	Q9H5V9	2
1905	UPF0557 protein C10orf119	C10orf119	-	Q9BTE3	2
1906	UPF0688 protein C1orf174	C1orf174	-	Q8IYL3	2
1907	Vacuolar protein sorting-associated protein 13C	KIAA1421	-	Q709C8	2
1908	Vacuolar proton pump subunit D	ATP6M	-	Q9Y5K8	2
1909	WD repeat-containing protein 64	WDR64	-	B1ANS9	2

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1910	WD repeat-containing protein mio	MIOS	-	Q9NXC5	2
1911	WW domain-containing adapter protein with coiled-coil	KIAA1844	-	Q9BTA9	2
1912	Zinc finger CCCH domain-containing protein 13	KIAA0853	-	Q5T200	2
1913	Zinc finger protein 99	C19orf9	-	A8MXY4	2
1914	Zyxin	ZYX	-	Q15942	2
1915	CD99 antigen	CD99	-	P14209	1
1916	130 kDa cis-Golgi matrix protein	GOLGA2	-	Q08379	1
1917	150 kDa oxygen-regulated protein	GRP170	-	Q9Y4L1	1
1918	15-oxoprostaglandin 13-reductase	LTB4DH	-	Q14914	1
1919	160 kDa neurofilament protein	NEF3	-	P07197	1
1920	1F5 antigen	CD59	-	E9PR17	1
1921	1-phosphatidylinositol-5-phosphate 4-kinase 2-beta	PIP4K2B	-	P78356	1
1922	28S ribosomal protein S17, mitochondrial	HSPC011	-	I3L0E3	1
1923	2-phospho-D-glycerate hydro-lyase	ENO2	-	P09104	1
1924	3-hydroxyisobutyrate dehydrogenase-like protein	GLYR1	-	Q49A26	1
1925	40 kDa peptidyl-prolyl cis-trans isomerase	CYP40	-	Q08752	1
1926	40S ribosomal protein S19-binding protein 1	AROS	-	Q86WX3	1
1927	45 kDa NF-AT-interacting protein	NFATC2IP	-	Q8NCF5	1
1928	60 kDa BRG-1/Brm-associated factor subunit B	BAF60B	-	Q92925	1
1929	60S acidic ribosomal protein P0-like	RPLP0P6	-	Q8NHW5	1
1930	70 kDa mitochondrial autoantigen of primary biliary cirrhosis	DLAT	-	P10515	1
1931	70 kDa ribosomal protein S6 kinase 2	RPS6KB2	-	Q9UBS0	1
1932	75 kDa glucose-regulated protein	GRP75	-	P38646	1
1933	78 kDa gastrin-binding protein	HADH	-	P40939	1
1934	85 kDa nucleoporin	NUP75	-	Q9BW27	1
1935	90 kDa ribosomal protein S6 kinase 2	MAPKAPK1C	-	F2Z2J1	1
1936	Acetolactate synthase-like protein	AHAS	-	A1L0T0	1
1937	Actin-related protein 1B	ACTR1B	-	P42025	1
1938	Activator-recruited cofactor 36 kDa component	ARC36	-	Q9NPJ6	1
1939	Activator-recruited cofactor 77 kDa component	ARC77	-	Q9NVC6	1
1940	Acyl-CoA thioesterase 7	ACOT7	-	O00154	1
1941	Acyl-CoA-binding protein	DBI	-	P07108	1
1942	Adapter-related protein complex 1 sigma-1B subunit	AP1S2	-	P56377	1
1943	Adaptor-associated kinase 1	AAK1	-	Q2M2I8	1
1944	Adenylate cyclase-inhibiting G alpha protein	GNAI2	-	P04899-4	1
1945	Adoplin-2	HSPC160	-	Q53FV1	1
1946	ADP-ribosylation factor 5	ARF5	-	P84085	1

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1947	AF4/FMR2 family member 4	AF5Q31	-	Q9UHB7	1
1948	Aging-associated gene 4 protein	AAG4	-	P10909-2	1
1949	AGX-1	SPAG2	-	Q16222	1
1950	Aicardi-Goutieres syndrome 3 protein	AYP1	-	E9PN81	1
1951	Akirin-2	AKIRIN2	-	Q53H80	1
1952	Alpha T-catenin	CTNNA3	-	Q9UI47	1
1953	Alpha-1,3-mannosyltransferase ALG2	ALG2	-	Q9H553	1
1954	Alpha-CP2	PCBP2	-	Q15366-2	1
1955	Alpha-globin	HBA1	-	P69905	1
1956	Alpha-NAC-like	NACA2	-	Q9H009	1
1957	Alpha-PAK	PAK1	-	Q13153-2	1
1958	Alpha-tubulin 1	TUBA1	-	P68366	1
1959	Amidophosphoribosyltransferase	GPAT	-	Q06203	1
1960	Amino acid transporter A2	ATA2	-	Q96QD8	1
1961	Aminopeptidase P3	XPNPEP3	-	Q9NQH7	1
1962	Amplaxin	CTTN	-	Q14247-2	1
1963	Amplified in breast cancer protein 1	ABC1	-	Q6AI08	1
1964	Amplified in liver cancer protein 1	ALC1	-	Q86WJ1	1
1965	Amyotrophic lateral sclerosis 4 protein	ALS4	-	Q7Z333-4	1
1966	AN1-type zinc finger protein 5	ZA20D2	-	O76080	1
1967	Anchorin CII	ANX5	-	P08758	1
1968	ANKRD26-like family C member 1B	A26C1B	-	A5A3E0	1
1969	Ankyrin repeat domain-containing protein 30A	ANKRD30A	-	Q9BXX3	1
1970	Ankyrin repeat domain-containing protein 5	ANKRD5	-	Q9NU02	1
1971	Another new gene 2 protein	ANG2	-	Q9UID3	1
1972	Anti-silencing function protein 1 homolog A	ASF1A	-	Q9Y294	1
1973	AP1 subunit gamma-binding protein 1	AP1GBP1	-	Q9UMZ2	1
1974	APG5-like	APG5L	-	Q9H1Y0	1
1975	Apolipoprotein C3	APOC3	-	P02656	1
1976	ARF GTPase-activating protein GIT2	GIT2	-	Q14161	1
1977	Arf-GAP domain and FG repeats-containing protein 2	AGFG2	-	O95081	1
1978	Arf-GAP with GTPase, ANK repeat and PH domain-containing protein 3	AGAP3	-	E7EUN2	1
1979	Arginine-rich protein	ARMET	-	P55145	1
1980	Aromatic esterase 2	PON2	-	G3V1K3	1
1981	Arrestin beta-1	ARR1	-	P49407	1
1982	Aspartate aminotransferase, cytoplasmic	GOT1	-	P17174	1
1983	Aster-associated protein	ASAP	-	Q49MG5	1
1984	ATM and Rad3-related-interacting protein	AGS1	-	Q8WXE1	1

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1985	ATP-binding cassette sub-family F member 3	ABCF3	-	Q9NUQ8	1
1986	ATP-dependent RNA helicase DDX19B	DBP5	-	H3BQK0	1
1987	ATP-dependent RNA helicase DDX50	DDX50	-	Q9BQ39	1
1988	AU-rich element RNA-binding factor	HNRPDL	-	O14979	1
1989	AXOR35	GPCR105	-	Q9H3N8	1
1990	Basic transcription factor 2 34 kDa subunit	GTF2H3	-	Q13889	1
1991	Basic transcription factor 2 62 kDa subunit	BTF2	-	P32780	1
1992	Basic transcription factor 3-like 4	BTF3L4	-	Q96K17	1
1993	Beta-type platelet-derived growth factor receptor	PDGFRB	-	P09619	1
1994	Bobby sox homolog	BBX	-	Q8WY36	1
1995	Brain protein 16-like	BRP16L	-	P0CB43	1
1996	BRAP2	BRAP	-	Q7Z569	1
1997	Brefeldin A-inhibited guanine nucleotide-exchange protein 1	ARFGEF1	-	Q9Y6D6	1
1998	BRG1-associated factor 57	BAF57	-	Q969G3	1
1999	BRMS2	C15orf12	-	Q9NV31	1
2000	Butyrate response factor 2	BRF2	-	P47974	1
2001	Butyrate-induced protein 1	BIND1	-	H3BS72	1
2002	C3 and PZP-like alpha-2-macroglobulin domain-containing protein 6	CPAMD6	-	P20742	1
2003	Carboxypeptidase D	CPD	-	O75976	1
2004	Cdc42-interacting protein 4	CIP4	-	Q15642	1
2005	cDNA FLJ51245, highly similar to Actin-related protein 2/3 complex subunit 3	ARPC3	-	B4DM63	1
2006	cDNA FLJ52061, highly similar to Translocon-associated protein subunit gamma	SSR3	-	B4E2P2	1
2007	cDNA FLJ52800, highly similar to Alpha-centractin		-	B4DXP9	1
2008	cDNA FLJ53621, highly similar to Adapter-relatedprotein complex 1 sigma- 1B subunit		-	B7Z3M9	1
2009	cDNA FLJ54150, highly similar to Heterogeneous nuclear ribonucleoprotein D0		-	Q14103-2	1
2010	cDNA FLJ54536, highly similar to Mitochondrial 28S ribosomal protein S27	KIAA0264	-	B4DRT2	1
2011	cDNA FLJ55447, highly similar to ATP-citrate synthase (EC 2.3.3.8)		-	P53396-2	1
2012	cDNA FLJ56036, highly similar to Homo sapiens kinesin family member C3 (KIFC3), mRNA	KIFC3	-	B7Z484	1
2013	cDNA FLJ56946, highly similar to Cysteine-rich protein 2	CRIP2	-	B7Z6C0	1
2014	cDNA FLJ57714, highly similar to Poly(A)-binding protein 2		-	G3V4T2	1
2015	cDNA FLJ58012, moderately similar to Eukaryotic initiation factor 4A-I (EC 3.6.1.-)		-	B4DKP9	1

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2016	cDNA FLJ60932, highly similar to T-complex protein 1 subunit zeta-2		-	B4DX20	1
2017	Cell cycle regulatory protein p95	NBN	-	O60934	1
2018	Cell division autoantigen 1	CDA1	-	Q9H2G4	1
2019	Cell division protein kinase 6	CDK6	-	Q00534	1
2020	Cell growth-inhibiting gene 4 protein	GIG4	-	P51153	1
2021	Centromere protein E	CENPE	-	Q02224	1
2022	Cerebral cavernous malformations 3 protein	CCM3	-	Q9BUL8	1
2023	Charged multivesicular body protein 4b	C20orf178	-	Q9H444	1
2024	CHETK-alpha	CHK	-	P35790	1
2025	Chloride channel, nucleotide sensitive 1A	CLCI	-	P54105	1
2026	Clathrin interactor 1	CLINT1	-	Q14677	1
2027	Coiled-coil domain-containing protein 101	CCDC101	-	Q96ES7	1
2028	Coiled-coil domain-containing protein 151	CCDC151	-	A5D8V7	1
2029	Coiled-coil domain-containing protein 21	CCDC21	-	Q6P2H3	1
2030	COMM domain-containing protein 3	BUP	-	Q9UBI1	1
2031	Complement component C4B (Childo blood group)	C4B-1	-	A2BHY4	1
2032	Concentrative nucleoside transporter 1	CNT1	-	O00337	1
2033	CROC-1	CROC1	-	G3V2F7	1
2034	Cyclin-dependent kinase-like 4	CDKL4	-	Q2NME9	1
2035	Cyclophilin J	PPIL3	-	H7BZ14	1
2036	CYPLI	CYP51	-	Q16850	1
2037	Cysteine-rich interactor of PDZ three	CRIPT	-	Q9P021	1
2038	Cytosolic Fe-S cluster assembly factor NUBP2	NUBP2	-	H3BNS4	1
2039	DAP-like kinase	DAPK3	-	O43293	1
2040	DBI-1	DBI1	-	Q9UL03	1
2041	DEAH box protein 34	DDX34	-	Q14147	1
2042	Deubiquitinating enzyme 8	KIAA0055	-	P40818	1
2043	Differentiation-dependent protein A4	A4	-	Q04941	1
2044	DiGeorge syndrome critical region 8	C22orf12	-	Q8WYQ5	1
2045	Disco-interacting protein 2 homolog B	DIP2B	-	Q9P265	1
2046	Divergent upstream protein	DUC1	-	P20585	1
2047	DNA polymerase delta subunit 2	POLD2	-	P49005	1
2048	Dolichyl-diphosphooligosaccharide--protein glycosyltransferase subunit STT3B	SIMP	-	Q8TCJ2	1
2049	DSH homolog 2	DVL2	-	O14641	1
2050	Dual specificity mitogen-activated protein kinase kinase 1	MAP2K1	-	Q02750	1
2051	Dual specificity protein phosphatase 11	DUSP11	-	O75319	1
2052	DUBA-5	CGI-77	-	Q8N6M0	1

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2053	Early endosome antigen 1	EEA1	-	Q15075	1
2054	Echinoderm microtubule-associated protein-like 1	EMAP1	-	O00423-3	1
2055	Endoplasmic reticulum resident protein 44	ERP44	-	Q9BS26	1
2056	Endothelial zinc finger protein 2	EZF2	-	Q8N0Y2	1
2057	Enhancer of split groucho-like protein 3	KIAA1547	-	H0YL70	1
2058	EPH homology kinase 2	EHK2	-	Q9UF33	1
2059	Epidermal growth factor receptor kinase substrate 8-like protein 3	EPS8L3	-	Q8TE67-3	1
2060	Epidermal Langerhans cell protein LCP1	C14orf92	-	O94842	1
2061	Epidermal-type fatty acid-binding protein	FABP5	-	Q01469	1
2062	Erasin	KIAA0242	-	Q92575	1
2063	ERC protein 2	ERC2	-	O15083	1
2064	ESCRT-I complex subunit VPS28	VPS28	-	Q9UK41-2	1
2065	Ethanolaminephosphotransferase 1	EPT1	-	Q9C0D9	1
2066	Eukaryotic translation initiation factor 2C 1	AGO1	-	Q9UL18	1
2067	Extracellular matrix protein FRAS1	FRAS1	-	E9PHH6	1
2068	Eyes absent homolog 3	EYA3	-	Q99504	1
2069	FAS-associated factor 2	ETEA	-	Q96CS3	1
2070	F-box and leucine-rich repeat protein 12	FBL12	-	Q9NXK8	1
2071	F-box only protein 3	FBX3	-	Q9UK99	1
2072	Fermitin family homolog 2	FERMT2	-	Q96AC1-3	1
2073	Fibroblast growth factor receptor 3	FGFR3	-	P22607-2	1
2074	FK506-binding protein-associated protein	FAP48	-	Q92990	1
2075	Flavin-containing amine oxidase domain-containing protein 1	AOF1	-	Q8NB78	1
2076	F-spondin	KIAA0762	-	Q9HCB6	1
2077	G protein beta subunit-like	GBL	-	Q9BVC4	1
2078	G(i) alpha-3	GNAI3	-	P08754	1
2079	Gametogenetin-binding protein 2	GGNBP2	-	Q9H3C7	1
2080	GC-box-binding zinc finger protein 1	GZP1	-	Q9Y2X9	1
2081	GCUNC-45	SMAP1	-	Q9H3U1	1
2082	GDP-mannose pyrophosphorylase B	GMPPB	-	Q9Y5P6-2	1
2083	GLE1-like protein	GLE1	-	Q53GS7	1
2084	Glucocorticoid receptor	GRL	-	P04150-3	1
2085	Glucose phosphomutase 2	MSTP006	-	Q96G03	1
2086	Glutamine amidotransferase	GMPS	-	P49915	1
2087	Glycogen synthase kinase-3 alpha	GSK3A	-	P49840	1
2088	Glypican-6	GPC6	-	Q9Y625	1
2089	Golgi complex-associated protein of 170 kDa	GOLGA3	-	Q08378	1

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2090	Golgi integral membrane protein 4	GIMPC	-	O00461	1
2091	Gonadotropin-inducible ovary transcription repressor 2	GIOT2	-	P15621	1
2092	GTP-binding protein HSR1	GNL1	-	P36915	1
2093	Guanine nucleotide-binding protein G(y) subunit alpha	GA11	-	P29992	1
2094	H/ACA ribonucleoprotein complex non-core subunit NAF1	NAF1	-	Q96HR8	1
2095	HAUS augmin-like complex subunit 5	HAUS5	-	O94927	1
2096	HBV X protein up-regulated gene 4 protein	KIAA1507	-	Q8TCY9	1
2097	Heat shock 90 kDa protein 1 alpha-like 2	HSP90AA4P	-	Q58FG1	1
2098	Heat shock protein 90-beta b	HSP90AB2P	-	Q58FF8	1
2099	Histidine--tRNA ligase	HARS2	-	P49590	1
2100	Histone deacetylase 6	HDAC6	-	Q9UBN7	1
2101	Histone H2A.x	H2AFX	-	P16104	1
2102	Histone H2A.Z	H2AFZ	-	P0C0S5	1
2103	hTREX45	THOC3	-	Q96J01	1
2104	Importin subunit alpha-7	IPOA7	-	F5GYL8	1
2105	Inhibitor of growth protein 1	ING1	-	Q9UK53	1
2106	Inosine phosphorylase	NP	-	P00491	1
2107	Insulin receptor substrate 2	IRS2	-	Q9Y4H2	1
2108	Inter-alpha-trypsin inhibitor complex component II	IGHEP2	-	P19823	1
2109	Interleukin-25	C19orf10	-	Q969H8	1
2110	Intersex-like protein	IXL	-	Q9NX70	1
2111	IQ calmodulin-binding motif-containing protein 1	IQCB1	-	Q15051	1
2112	Kinesin heavy chain isoform 5C	KIAA0531	-	O60282	1
2113	Kinesin-like protein KIF3C	KIF3C	-	O14782	1
2114	La ribonucleoprotein domain family member 1B	LARP1B	-	Q659C4	1
2115	Leucine-rich repeat-containing protein 41	LRRC41	-	Q15345	1
2116	L-iditol 2-dehydrogenase	SORD	-	Q00796	1
2117	LIM domain-containing protein 2	LIMD2	-	Q9BT23	1
2118	Limkain-b1	KIAA0430	-	Q9Y4F3	1
2119	L-isoaspartyl protein carboxyl methyltransferase	PCMT1	-	H7BY58	1
2120	M phase phosphoprotein 10	MPHOSPH10	-	O00566	1
2121	MAGE tumor antigen CCF	MAGED1	-	Q9Y5V3-2	1
2122	Membrane-associated progesterone receptor component 2	DG6	-	O15173	1
2123	Microsomal signal peptidase 12 kDa subunit	HSPC033	-	Q9Y6A9	1
2124	MLL septin-like fusion protein MSF-A	KIAA0991	-	Q9UHD8	1
2125	MRG-binding protein	C20orf20	-	Q9NV56	1
2126	Nectin-like protein 5	PVR	-	P15151	1

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2127	Neuroblastoma-derived sulfhydryl oxidase	QSCN6L1	-	Q6ZRP7	1
2128	Nicalin	NCLN	-	Q969V3	1
2129	NOP seven-associated protein 1	NSA1	-	Q6RFH5	1
2130	Nori-2	C20orf64	-	Q96S44	1
2131	Nuclear FMRP-interacting protein 1	NUFIP1	-	Q9UHK0	1
2132	Nuclear nucleic acid-binding protein C1D	C1D	-	Q13901	1
2133	Nuclear receptor-interacting factor 4	NRIF4	-	Q9NRM2	1
2134	Nucleolar protein 11	L14	-	Q9H8H0	1
2135	Nucleolar protein 12	NOL12	-	Q9UGY1	1
2136	Obscurin-like protein 1	KIAA0657	-	O75147	1
2137	Olfactomedin-like protein 2B	OLFML2B	-	F2Z3N3	1
2138	Origin recognition complex subunit 5	ORC5	-	O43913	1
2139	Ovarian cancer-related protein	OCR	-	Q9Y657	1
2140	Oxysterol-binding protein	hCG_1780933	-	Q96SU4-6	1
2141	p53 and DNA damage-regulated protein 1	C20orf126	-	Q9NUG6	1
2142	Pancreatic differentiation protein 2	PAF1	-	Q8N7H5	1
2143	Papillary renal cell carcinoma translocation-associated gene protein	PRCC	-	Q92733	1
2144	Peptidyl-prolyl cis-trans isomerase H	CYP20	-	O43447	1
2145	PEST proteolytic signal-containing nuclear protein	PCNP	-	Q8WW12	1
2146	Phosducin-like protein	PDCL	-	Q13371	1
2147	Plasma membrane calcium ATPase isoform 3	ATP2B3	-	Q16720	1
2148	Prefoldin subunit 4	PFD4	-	E9PQY2	1
2149	Programmed cell death protein 2	PDCD2	-	Q16342	1
2150	Prostate leucine zipper variant 1	PrLZ	-	F5H0B0	1
2151	Proteasome (Prosome, macropain) 26S subunit, non-ATPase, 4	hCG_1741224	-	Q5VWC4	1
2152	Protein mago nashi homolog	MAGOH	-	P61326	1
2153	Protein phosphatase X	PPP4	-	P60510	1
2154	Protein rogdi homolog	ROGDI	-	Q9GZN7	1
2155	Protein taube nuss	TAF8	-	Q7Z7C8-4	1
2156	Protein transport protein Sec24D	KIAA0755	-	O94855-2	1
2157	Protein YIF1B	PP4519	-	Q5BJH7	1
2158	Protein YIPF2	YIPF2	-	Q9BWQ6	1
2159	Proximal sequence element-binding transcription factor subunit alpha	SNAP190	-	Q5SXM2	1
2160	Pseudouridylate synthase 7 homolog-like protein	PUS7L	-	Q9H0K6	1
2161	Pumilio homolog 1 (Drosophila)	hCG_19946	-	Q5T1Z8	1
2162	Putative PIP5K1A and PSMD4-like protein	PIPSL	-	A2A3N6	1

Continuing Appendix 1. List of proteins identified by MS/MS in global protein subcellular distribution analysis on a global proteomic scale

2163	Putative uncharacterized protein C20orf43	C20orf43	-	A8MSH5	1
2164	Putative uncharacterized protein DIAPH3	DIAPH3	-	C9JDG0	1
2165	Putative uncharacterized protein GPC3	GPC3	-	C9JLE3	1
2166	Putative uncharacterized protein HCFC1	HCFC1	-	A6NEM2	1
2167	Putative uncharacterized protein MED25	MED25	-	Q71SY5-3	1
2168	Putative uncharacterized protein OBSCN	OBSCN	-	H3BPX2	1
2169	Putative uncharacterized protein RBBP7	RBBP7	-	C9JAJ9	1
2170	Putative uncharacterized protein SNRPB	SNRPB	-	A8MT02	1
2171	Putative uncharacterized protein SUMO3	SUMO3	-	A8MU27	1
2172	Ras suppressor protein 1	RSP1	-	Q15404	1
2173	Ras-related protein Rab-8B	RAB8B	-	Q92930	1
2174	RB1-inducible coiled-coil protein 1	KIAA0203	-	Q8TDY2	1
2175	Replication factor A protein 3	REPA3	-	P35244	1
2176	RER1 protein	RER1	-	Q9P0H9	1
2177	Retinoic acid suppression protein A	KOX17	-	P17028	1
2178	Rho GTPase-activating protein 17	ARHGAP17	-	Q68EM7	1
2179	Rho GTPase-activating protein 19	ARHGAP19	-	Q14CB8-2	1
2180	Ribosomal RNA processing protein 1 homolog B	KIAA0179	-	Q14684	1
2181	RING finger protein 126	RNF126	-	Q9BV68	1
2182	RING finger protein 86	KIAA0517	-	Q9C040	1
2183	RNA-binding motif protein 30	RBM30	-	Q9BQ04	1
2184	Round spermatid basic protein 1	RSBN1	-	Q5VWQ0	1
2185	S phase cyclin A-associated protein in the endoplasmic reticulum	KIAA1454	-	H3BNR7	1
2186	S1 RNA-binding domain-containing protein 1	SRBD1	-	Q8N5C6	1
2187	SEC16A protein	SEC16A	-	A4QN19	1
2188	SEC24B protein	SEC24B	-	B7ZKM8	1
2189	Septin-6	KIAA0128	-	F5H1J5	1
2190	Septin-8	KIAA0202	-	Q92599	1
2191	Serine arginine-rich pre-mRNA splicing factor SR-A1	SCAF1	-	Q9H7N4	1
2192	Serine/threonine-protein phosphatase 2A catalytic subunit beta isoform	PPP2CB	-	P62714	1
2193	Serine/threonine-protein phosphatase PP1-beta catalytic subunit	PPP1CB	-	P62140	1
2194	Serum-spreading factor	VTN	-	P04004	1
2195	SNARE-associated protein Snapin	SNAP25BP	-	O95295	1
2196	Solute carrier family 30 member 7	SLC30A7	-	Q8NEW0	1
2197	Sorting nexin-4	SNX4	-	O95219	1
2198	Spectrin, beta, erythrocytic (Includes spherocytosis, clinical type I) variant	SPTB	-	Q59FP5	1

Continuing Appendix 1. List of proteins identified by MS/MS in global protein subcellular distribution analysis on a global proteomic scale

2199	Spindle and kinetochore-associated protein 1	C18orf24	-	Q96BD8	1
2200	Splicing factor, arginine/serine-rich 8	SFRS8	-	F5H6B8	1
2201	Staufen, RNA binding protein, homolog 1 (Drosophila)	RP3-470L14.2-007	-	Q5JW30	1
2202	SUMO-activating enzyme subunit 1	AOS1	-	Q9UBE0	1
2203	Superoxide dismutase [Cu-Zn]	SOD1	-	P00441	1
2204	Synaptosomal-associated protein 23	SNAP23	-	O00161	1
2205	Syndecan-1	SDC	-	P18827	1
2206	TCP1-chaperonin cofactor A	TBCA	-	E5RJD8	1
2207	Telomerase Cajal body protein 1	TCAB1	-	Q9BUR4	1
2208	Terminal uridylyltransferase 4	KIAA0191	-	Q5TAX3	1
2209	Transcription factor IIB	C16orf80	-	Q9Y6A4	1
2210	Transducin beta-like protein 2	TBL2	-	Q9Y4P3	1
2211	Translocation protein SEC63 homolog	SEC63	-	Q9UGP8	1
2212	Transmembrane and coiled-coil domain-containing protein 5A	TMCO5	-	Q8N6Q1	1
2213	Transmembrane protein 179B	TMEM179B	-	Q7Z7N9	1
2214	Transmembrane protein 192	TMEM192	-	Q8IY95	1
2215	Transmembrane protein 2	KIAA1412	-	Q9UHN6	1
2216	Transmembrane protein 205	TMEM205	-	Q6UW68	1
2217	Transmembrane protein 87A	PSEC0094	-	Q8NBN3	1
2218	Trinucleotide repeat-containing gene 6B protein	KIAA1093	-	Q9UPQ9	1
2219	Tropomyosin 3	hCG_1997142	-	P06753-2	1
2220	TTD non-photosensitive 1 protein	C7orf11	-	Q8TAP9	1
2221	Tubulin beta-3 chain	TUBB3	-	Q13509	1
2222	Uba6-specific E2 conjugating enzyme 1	HOYS7	-	Q9H832	1
2223	Ubiquitin carboxyl-terminal hydrolase L5	RP11-101E13.2-007	-	Q5LJA5	1
2224	Uncharacterized protein C22orf30	C22orf30	-	Q5THK1	1
2225	Uncharacterized protein C7orf26	C7orf26	-	Q96N11	1
2226	Uncharacterized protein C9orf142	C9orf142	-	Q9BUH6	1
2227	Uncharacterized protein FLJ45252		-	I3L097	1
2228	Vacuolar protein sorting-associated protein 26A	VPS26	-	O75436	1
2229	WASH complex subunit strumpellin	KIAA0196	-	Q12768	1
2230	WD repeat-containing protein 37	KIAA0982	-	Q9Y2I8	1
2231	Zinc finger CCCH-type antiviral protein 1-like	C7orf39	-	Q96H79	1
2232	Zinc finger protein 260	ZFP260	-	Q3ZCT1	1

2233	Zinc finger protein 323, isoform CRA_b	hCG_1770259	-	Q6P178	1
2234	Zinc finger protein 404	ZNF404	-	Q494X3	1
2235	Zinc finger protein 49	ZNF49	-	Q86UE3	1
2236	Zinc finger protein 502	ZNF502	-	Q8TBZ5	1
2237	Zinc finger protein 512	KIAA1805	-	Q96ME7	1
2238	Zinc finger protein 545	KIAA1948	-	F5H4L8	1
2239	Zinc finger protein 689	ZNF689	-	Q96CS4	1
2240	Zinc finger protein 878	ZNF878	-	C9JN71	1

Appendix 2. List of proteins identified by MS/MS in the comparative global proteomic analyses of secretomes from HUH7 cells overexpressing PCSK9 and its variants

Majority Protein Uniprot ID	Gene Names	Protein Names	Unique Peptides
A0AVT1	UBA6	ubiquitin-like modifier activating enzyme 6	4
A0MZ66	KIAA1598	KIAA1598	2
A5A3E0	POTEE/POTEF	POTE ankyrin domain family, member F	1
A6NDG6	PGP	phosphoglycolate phosphatase	7
A6NE09	RPSAP58	ribosomal protein SA pseudogene 58	13
A6NEM2	HCFC1	host cell factor C1	4
A6NFX8	NUDT5	nudix (nucleoside diphosphate linked moiety X)-type motif 5	7
A6NG51	SPTAN1	Putative uncharacterized protein SPTAN1	54
A6NHF2	BTD	biotinidase	2
A6NJG9	PSME1	proteasome (prosome, macropain) activator subunit 1 (PA28 alpha)	5
A7MAP0	CORO1C	coronin, actin binding protein, 1C	17
A7YIJ8	RDX	radixin	22
A8KAH5	IST1	increased sodium tolerance 1 homolog (yeast)	3
A8MSH5	C20orf43	Putative uncharacterized protein C20orf43	1
A8MUH7	PDZK1P2	PDZ domain containing 1 pseudogene 2	7
A8MV58	DBN1	drebrin 1	9
A8MWH4	MT1F	Metallothionein	3
A8MXP9	MATR3	matrin 3	6
A8MZ22	NAP1L4	Nucleosome assembly protein 1-like 4b	6
B0QY89	EIF3L	eukaryotic translation initiation factor 3, subunit L	10
B0QYA3	EIF3D	Eukaryotic translation initiation factor 3, subunit D	1
B0QYN7	UBE2I	ubiquitin-conjugating enzyme E2I	4
B0V043	DAAP-21F2.2-001	Valyl-tRNA synthetase	9
B0YIW6	ARCN1	archain 1	13
B1AHD1	NHP2L1	NHP2 non-histone chromosome protein 2-like 1 (S. cerevisiae)	4
B1AHL2	FBLN1*	fibulin 1	4
B1AKJ5	NRD1	nardilysin (N-arginine dibasic convertase)	6
B1AKR6	DYNLRB1	dynein, light chain, roadblock-type 1	4
B1AVU8	PSAP	prosaposin	14
B1PS43	MYH11	myosin, heavy chain 11, smooth muscle	1

B3KRG5	RBMX*	RNA binding motif protein, X-linked	1
B3KSH1	EIF3F	eukaryotic translation initiation factor 3, subunit F	5
B3KUE5	PLTP	phospholipid transfer protein	1
B3KXW5	AP1G1	adaptor-related protein complex 1, gamma 1 subunit	6
B4DEG8	TARS	threonyl-tRNA synthetase	17
B4DFL2	IDH2	isocitrate dehydrogenase 2 (NADP+), mitochondrial	1
B4DKT0	GORASP2	golgi reassembly stacking protein 2, 55kDa	4
B4DLH4	HN1L	hematological and neurological expressed 1-like	7
B4DNJ6	STRAP	serine/threonine kinase receptor associated protein	10
B4DPK8	HNRNPL*	heterogeneous nuclear ribonucleoprotein L	1
B4DPQ7	PROC	protein C (inactivator of coagulation factors Va and VIIIa)	3
B4DR87	PLOD1	procollagen-lysine, 2-oxoglutarate 5-dioxygenase 1	4
B4DRY3	MOB1B	MOB kinase activator 1B	1
B4DT31	FUBP1	far upstream element (FUSE) binding protein 1	13
B4DT67	SPATS2L	spermatogenesis associated, serine-rich 2-like	2
B4DW92	NR2C2AP	nuclear receptor 2C2-associated protein	2
B4E1Z4	BF	cDNA FLJ55673, highly similar to Complement factor B (EC 3.4.21.47)	8
B5MBZ0	EML4	echinoderm microtubule associated protein like 4	4
B5MC82	DDT	D-dopachrome tautomerase	4
B5MDF5	RAN	RAN, member RAS oncogene family	14
B5ME19	EIF3CL	eukaryotic translation initiation factor 3, subunit C-like	19
B5ME97	hCG_27895	septin 10	2
B7TY16	ACTN1	actinin, alpha 1	16
B7Z596	TPM1	tropomyosin 1 (alpha)	4
B7Z6Z4	MYL6	myosin, light chain 6, alkali, smooth muscle and non-muscle	6
B7Z7Q0	PITPNB	phosphatidylinositol transfer protein, beta	5
B7ZKJ8	ITIH4	inter-alpha-trypsin inhibitor heavy chain family, member 4	2
B7ZKM8	SEC24B	SEC24 family member B	1
B8ZZN6	SUMO1	small ubiquitin-like modifier 1	7
C9J3F9	PPAN	peter pan homolog (Drosophila)	2
C9J436	PRRC1	Putative uncharacterized protein PRRC1	3
C9J6D7	KIAA0664	Putative uncharacterized protein KIAA0664	9
C9J6P4	ZC3HAV1	zinc finger CCCH-type, antiviral 1	1
C9J7E5	TNPO3	transportin 3	4
C9J8T6	COX17	COX17 cytochrome c oxidase copper chaperone	1
C9JAF2	IGF2	insulin-like growth factor 2	10
C9JC84	FGG	fibrinogen gamma chain	16
C9JCC6	DRAP1	DR1-associated protein 1 (negative cofactor 2 alpha)	2

C9JE22	H2AFJ	Histone H2A	1
C9JIF9	APEH	acylaminoacyl-peptide hydrolase	9
C9JLE3	GPC3	glypican 3	4
C9JPG0	SCRN1	secernin 1	5
C9JVQ0	SNRPG	small nuclear ribonucleoprotein polypeptide G	3
C9JWM7	ARPC4	actin related protein 2/3 complex, subunit 4, 20kDa	7
D6R956	UCHL1*	ubiquitin carboxyl-terminal esterase L1 (ubiquitin thiolesterase)	1
D6R9I9	ABCE1*	ATP-binding cassette, sub-family E (OABP), member 1	1
D6RB78	PDLIM5	PDZ and LIM domain 5	3
D6RER5	SEP11	septin 11	8
D6REX3	SEC31A	SEC31 homolog A (S. cerevisiae)	9
E5RGV0	HNRNPH1*	heterogeneous nuclear ribonucleoprotein H1 (H)	1
E5RIM7	ATOX1	antioxidant 1 copper chaperone	4
E5RJR5	SKP1	S-phase kinase-associated protein 1	5
E7EMY5	C17orf25	Glyoxalase domain-containing protein 4	10
E7ENU4	ADAR	136 kDa double-stranded RNA-binding protein	2
E7EP23	ELP2	elongator acetyltransferase complex subunit 2	1
E7EPK6	RPS24	ribosomal protein S24	4
E7EQ40	CAPZB	Capping protein (Actin filament) muscle Z-line, beta	11
E7EQT4	ACIN1	apoptotic chromatin condensation inducer 1	3
E7EQY1	FAM136A	family with sequence similarity 136, member A	4
E7ERS3	ZC3H18	zinc finger CCCH-type containing 18	3
E7ES43	APG1	Heat shock 70 kDa protein 4L	2
E7ESY9	PK1	Pyruvate kinase 1	1
E7ETH0	CFI	complement factor I	16
E7EU23	GDI2	Guanosine diphosphate dissociation inhibitor 2	21
E7EUC7	UGP2	UDP-glucose pyrophosphorylase 2	13
E7EUJ1	LIPC	lipase, hepatic	1
E7EVA0	MAP4	microtubule-associated protein 4	26
E7EW61	PALB	ATTR	2
E7EWS7	UBE2L3	ubiquitin-conjugating enzyme E2L 3	8
E7EWZ9	GMFB	Glia maturation factor beta	5
E7EX17	EIF4B	eukaryotic translation initiation factor 4B	19
E7EX29	YWHAZ	tyrosine 3-monooxygenase/tryptophan 5-monooxygenase activation protein, zeta	17
E9PAV3	NACA	nascent polypeptide-associated complex alpha subunit	6
E9PB61	ALYREF	Aly/REF export factor	5
E9PBF1	UCHL3	Ubiquitin carboxyl-terminal hydrolase isozyme L3	5

E9PBP6	MTTP	microsomal triglyceride transfer protein	4
E9PC67	ABC50	ATP-binding cassette 50	2
E9PCA1	CCT5*	chaperonin containing TCP1, subunit 5 (epsilon)	1
E9PCT1	SRRM1	serine/arginine repetitive matrix 1	2
E9PD68	CRMP1	collapsin response mediator protein 1	1
E9PDB1	SPTBN4	spectrin, beta, non-erythrocytic 4	1
E9PEG8	USP19	ubiquitin specific peptidase 19	1
E9PEQ4	DNM2	dynamin 2	6
E9PF41	ACTR2	ARP2 actin-related protein 2 homolog (yeast)	8
E9PF86	PGM3	phosphoglucomutase 3	11
E9PFM1	EIF4G1	eukaryotic translation initiation factor 4 gamma, 1	8
E9PFU1	PDCD6IP	programmed cell death 6 interacting protein	23
E9PGC5	PTPRK	protein tyrosine phosphatase, receptor type, K	5
E9PGN7	SERPING1	serpin peptidase inhibitor, clade G (C1 inhibitor), member 1	14
E9PGZ1	CALD1	caldesmon 1	11
E9PHH6	FRAS1	Extracellular matrix protein FRAS1	3
E9PHK9	TCOF1	Treacher Collins syndrome protein	9
E9PK25	CFL1	cofilin 1 (non-muscle)	14
E9PK86	SERPINH1*	serpin peptidase inhibitor, clade H (heat shock protein 47), member 1, (collagen binding protein 1)	1
E9PLM6	MDK	midkine (neurite growth-promoting factor 2)	8
E9PP21	CSRP1	cysteine and glycine-rich protein 1	3
E9PQI8	SART1*	squamous cell carcinoma antigen recognized by T cells	1
E9PQY2	PFDN4	prefoldin subunit 4	3
E9PR47	AAMDC	adipogenesis associated, Mth938 domain containing	4
E9PRY8	EEF1D	eukaryotic translation elongation factor 1 delta (guanine nucleotide exchange protein)	2
E9PS38	NEDD8-MDP1	NEDD8-MDP1 readthrough	5
F5GYI6	CDH1	Cadherin-1	3
F5H098	MDH1	malate dehydrogenase 1, NAD (soluble)	20
F5H0Q5	AHSG	Alpha-2-HS-glycoprotein	12
F5H0X5	hCG_29955	SMARCA4 isoform 2	2
F5H1B6	MBS	Myosin phosphatase-targeting subunit 1	3
F5H1E9	BM28	DNA replication licensing factor MCM2	25
F5H1Y4	GOPC	golgi-associated PDZ and coiled-coil motif containing	2
F5H299	COL1A2	Alpha-2 type I collagen	4
F5H2F4	MTHFD1	methylenetetrahydrofolate dehydrogenase (NADP+ dependent) 1, methenyltetrahydrofolate cyclohydrolase, formyltetrahydrofolate synthetase	5

F5H2S7	DCTN2	50 kDa dynein-associated polypeptide	7
F5H301	TJP2	tight junction protein 2	5
F5H423	ARF3	ADP-ribosylation factor 3	3
F5H4L0	TUBGCP2	tubulin, gamma complex associated protein 2	3
F5H5E4	BCAT1	branched chain amino-acid transaminase 1, cytosolic	3
F5H6K1	DMD	Dystrophin	3
F5H6Q1	OTUB1	OTU deubiquitinase, ubiquitin aldehyde binding 1	6
F5H8B1	PCYT2	phosphate cytidylyltransferase 2, ethanolamine	7
F6S8M0	GNS	glucosamine (N-acetyl)-6-sulfatase	3
F6XZQ7	GSTM2	glutathione S-transferase mu 2 (muscle)	2
F8VQ10	BAT1	56 kDa U2AF65-associated protein	19
F8VTL3	MYH10	myosin, heavy chain 10, non-muscle	3
F8VZ69	PA2G4*	proliferation-associated 2G4, 38kDa	1
F8W1H5	RAB5C	RAB5C, member RAS oncogene family	4
F8W543	NAP1L1*	nucleosome assembly protein 1-like 1	1
F8W696	APOA1*	apolipoprotein A-I	1
F8W6F6	ATSV	Axonal transporter of synaptic vesicles	1
F8W727	RPL32	ribosomal protein L32	5
F8W8D1	DLP1	Dnm1p/Vps1p-like protein	3
F8W8S0	DNCI2	Cytoplasmic dynein 1 intermediate chain 2	4
F8W930	IGF2BP2	insulin-like growth factor 2 mRNA binding protein 2	1
F8W9U3	ABHD14B*	abhydrolase domain containing 14B	1
F8WCL5	NEB	nebulin	3
G3V180	DPP3	dipeptidyl-peptidase 3	27
G3V1L9	TJP1	tight junction protein 1	1
G3V1S2	CUL2	cullin 2	5
G3V1Z5	BAT3	HLA-B-associated transcript 3	2
G3V3A4	SNW1	SNW domain containing 1	2
G3V3D1	NPC2	Niemann-Pick disease, type C2	3
G3V5I3	SERPINA3	serpin peptidase inhibitor, clade A (alpha-1 antiproteinase, antitrypsin), member 3	2
G3V5T9	CDK2	cyclin-dependent kinase 2	1
G3V5Z7	PSMA6	proteasome (prosome, macropain) subunit, alpha type, 6	14
G3XAI2	LAMB1	laminin, beta 1	19
G5E977	NAPRT	nicotinate phosphoribosyltransferase	5
G5E9M5	ILF3	interleukin enhancer binding factor 3, 90kDa	16
G5E9Q6	PFN2*	profilin 2	2
G5E9Y2	XPNPEP1	X-prolyl aminopeptidase (aminopeptidase P) 1, soluble	2

G8JLA8	BIGH3	Kerato-epithelin	14
G8JLB6	HNRNPH1*	heterogeneous nuclear ribonucleoprotein H1 (H)	3
H0Y3D5	FGFR4	Fibroblast growth factor receptor 4	2
H0Y3H1	TOLLIP	Toll-interacting protein	3
H0Y5B4	RPL36A	ribosomal protein L36a	3
H0Y7A7	CALM1 (includes others)	calmodulin 1 (phosphorylase kinase, delta)	7
H0YB34	HRSP12	heat-responsive protein 12	2
H0YG33	HSPA1	Heat shock 70 kDa protein 1/2	14
H0YG49	SRSF2	serine/arginine-rich splicing factor 2	10
H0YGH4	A2M	Alpha-2-macroglobulin	48
H0YGW6	TIA1	Nucleolysin TIA-1 isoform p15	1
H0YK46	RPS17	40S ribosomal protein S17	5
H0YMV8	RPS27L	ribosomal protein S27-like	3
H0YNJ6	GMPR2	guanosine monophosphate reductase 2	2
H3BMD8	ARPP19	cAMP-regulated phosphoprotein, 19kDa	2
H3BP20	HEXA	hexosaminidase A (alpha polypeptide)	3
H3BPE7	FUS	FUS RNA binding protein	2
H3BQK0	DDX19B	DEAD (Asp-Glu-Ala-Asp) box polypeptide 19B	6
H3BQK9	MACF1	microtubule-actin crosslinking factor 1	3
H3BUK8	RPL13*	ribosomal protein L13	1
H7BY58	PCMT1	protein-L-isoaspartate (D-aspartate) O-methyltransferase	6
H7BY64	PRAP1	proline-rich acidic protein 1	3
H7BY78	HPRT	Hypoxanthine-guanine phosphoribosyltransferase	3
H7BZJ3	PDIA3*	protein disulfide isomerase family A, member 3	1
H7BZR0	XRCC1	DNA repair protein XRCC1	2
H7C1E4	AP1S1	adaptor-related protein complex 1, sigma 1 subunit	1
H7C2I1	PRMT1	protein arginine methyltransferase 1	10
H9KV28	DIAPH1	diaphanous-related formin 1	7
I1E4Y6	GIGYF2	GRB10 interacting GYF protein 2	1
I3L080	HDGFRP2	hepatoma-derived growth factor-related protein 2	5
I3L0C9	FAM21A	WASH complex subunit FAM21A	1
I3L504	EIF5A	eukaryotic translation initiation factor 5A	9
O00115	DNASE2	deoxyribonuclease II, lysosomal	2
O00139	KIF2A	kinesin heavy chain member 2A	3
O00151	PDLIM1	PDZ and LIM domain 1	10
O00170	AIP	aryl hydrocarbon receptor interacting protein	11
O00231	PSMD11	proteasome (prosome, macropain) 26S subunit, non-ATPase, 11	8

O00267	SUPT5H	suppressor of Ty 5 homolog (S. cerevisiae)	2
O00299	CLIC1	chloride intracellular channel 1	13
O00300	TNFRSF11B	tumor necrosis factor receptor superfamily, member 11b	3
O00391	QSOX1	quiescin Q6 sulfhydryl oxidase 1	8
O00410	IPO5	importin 5	18
O00425	IGF2BP3	insulin-like growth factor 2 mRNA binding protein 3	2
O00461	GOLIM4	golgi integral membrane protein 4	1
O00468	AGRN	agrin	9
O00479	HMGN4	high mobility group nucleosomal binding domain 4	2
O00499	BIN1	bridging integrator 1	3
O00505	KPNA3	karyopherin alpha 3 (importin alpha 4)	3
O00571	DDX3X	DEAD (Asp-Glu-Ala-Asp) box helicase 3, X-linked	5
O00625	PIR	pirin (iron-binding nuclear protein)	4
O00629	KPNA4	karyopherin alpha 4 (importin alpha 3)	1
O00743	PPP6C	protein phosphatase 6, catalytic subunit	3
O14579	COPE	coatomer protein complex, subunit epsilon	8
O14618	CCS	copper chaperone for superoxide dismutase	7
O14672	ADAM10	ADAM metallopeptidase domain 10	2
O14732	IMPA2	inositol(myo)-1(or 4)-monophosphatase 2	3
O14737	PDCD5	programmed cell death 5	10
O14744	PRMT5	protein arginine methyltransferase 5	8
O14745	SLC9A3R1	solute carrier family 9, subfamily A (NHE3, cation proton antiporter 3), member 3 regulator 1	14
O14776	TCERG1	transcription elongation regulator 1	4
O14818	PSMA7	proteasome (prosome, macropain) subunit, alpha type, 7	12
O14929	HAT1	histone acetyltransferase 1	5
O14950	MYL12B	myosin, light chain 12B, regulatory	2
O14979	HNRNPDL	heterogeneous nuclear ribonucleoprotein D-like	7
O14980	XPO1	exportin 1	11
O15067	PFAS	phosphoribosylformylglycinamidine synthase	20
O15078	CEP290	centrosomal protein 290kDa	3
O15143	ARPC1B	actin related protein 2/3 complex, subunit 1B, 41kDa	4
O15144	ARPC2	actin related protein 2/3 complex, subunit 2, 34kDa	8
O15145	ARPC3	actin related protein 2/3 complex, subunit 3, 21kDa	4
O15212	PFDN6	prefoldin subunit 6	6
O15230	LAMA5	laminin, alpha 5	2
O15232	MATN3	matrilin 3	3
O15347	HMGB3	high mobility group box 3	9

O15355	PPM1G	protein phosphatase, Mg2+/Mn2+ dependent, 1G	11
O15371	EIF3D	eukaryotic translation initiation factor 3, subunit D	2
O15372	EIF3H	eukaryotic translation initiation factor 3, subunit H	5
O15498	YKT6	YKT6 v-SNARE homolog (S. cerevisiae)	3
O15511	ARPC5	actin related protein 2/3 complex, subunit 5, 16kDa	2
O43143	DHX15	DEAH (Asp-Glu-Ala-His) box helicase 15	13
O43148	RNMT	RNA (guanine-7-) methyltransferase	3
O43172	PRPF4	pre-mRNA processing factor 4	3
O43175	PHGDH	phosphoglycerate dehydrogenase	18
O43242	PSMD3	proteasome (prosome, macropain) 26S subunit, non-ATPase, 3	8
O43252	PAPSS1	3'-phosphoadenosine 5'-phosphosulfate synthase 1	1
O43290	SART1*	squamous cell carcinoma antigen recognized by T cells	2
O43324	EEF1E1	eukaryotic translation elongation factor 1 epsilon 1	2
O43390	HNRNPR	heterogeneous nuclear ribonucleoprotein R	8
O43396	TXNL1	thioredoxin-like 1	2
O43399	TPD52L2	tumor protein D52-like 2	4
O43405	COCH	cochlin	18
O43447	PPIH	peptidylprolyl isomerase H (cyclophilin H)	2
O43488	AKR7A2	aldo-keto reductase family 7, member A2 (aflatoxin aldehyde reductase)	4
O43491	EPB41L2	erythrocyte membrane protein band 4.1-like 2	9
O43493	TGOLN2	trans-golgi network protein 2	3
O43505	B4GAT1	beta-1,4-glucuronyltransferase 1	15
O43583	DENR	density-regulated protein	4
O43592	XPOT	exportin, tRNA	3
O43617	TRAPPC3	trafficking protein particle complex 3	2
O43684	BUB3	BUB3 mitotic checkpoint protein	5
O43707	ACTN4	actinin, alpha 4	41
O43765	SGTA	small glutamine-rich tetratricopeptide repeat (TPR)-containing, alpha	9
O43776	NARS	asparaginyl-tRNA synthetase	8
O43809	NUDT21	nudix (nucleoside diphosphate linked moiety X)-type motif 21	6
O43852	CALU	calumenin	5
O43865	AHCYL1	adenosylhomocysteinase-like 1	1
O43896	KIF1C	kinesin family member 1C	1
O43903	GAS2	growth arrest-specific 2	2
O60218	AKR1B10	aldo-keto reductase family 1, member B10 (aldose reductase)	5
O60256	PRPSAP2	phosphoribosyl pyrophosphate synthetase-associated protein 2	2
O60333	KIF1B	kinesin family member 1B	1
O60343	TBC1D4	TBC1 domain family, member 4	2

O60493	SNX3	sorting nexin 3	4
O60506	SYNCRIP*	synaptotagmin binding, cytoplasmic RNA interacting protein	1
O60568	PLOD3	procollagen-lysine, 2-oxoglutarate 5-dioxygenase 3	10
O60664	PLIN3	perilipin 3	6
O60701	UGDH	UDP-glucose 6-dehydrogenase	16
O60749	SNX2	sorting nexin 2	3
O60763	USO1	USO1 vesicle transport factor	7
O60841	EIF5B	eukaryotic translation initiation factor 5B	5
O60869	EDF1	endothelial differentiation-related factor 1	8
O60888	CUTA	cutA divalent cation tolerance homolog (E. coli)	2
O60911	CTSV	cathepsin V	3
O60925	PFDN1	prefoldin subunit 1	4
O75083	WDR1	WD repeat domain 1	20
O75116	ROCK2	Rho-associated, coiled-coil containing protein kinase 2	1
O75155	CAND2	cullin-associated and neddylation-dissociated 2 (putative)	1
O75175	CNOT3	CCR4-NOT transcription complex, subunit 3	2
O75191	XYLB	xylulokinase homolog (H. influenzae)	6
O75223	GGCT	gamma-glutamylcyclotransferase	6
O75312	ZPR1	ZPR1 zinc finger	2
O75347	TBCA	tubulin folding cofactor A	16
O75348	ATP6V1G1	ATPase, H+ transporting, lysosomal 13kDa, V1 subunit G1	3
O75369	FLNB	filamin B, beta	33
O75390	CS	citrate synthase	3
O75396	SEC22B	SEC22 vesicle trafficking protein homolog B (S. cerevisiae) (gene/pseudogene)	2
O75436	VPS26A	vacuolar protein sorting 26 homolog A (S. pombe)	1
O75475	PSIP1	PC4 and SFRS1 interacting protein 1	3
O75533	SF3B1	splicing factor 3b, subunit 1, 155kDa	4
O75608	LYPLA1	lysophospholipase I	2
O75643	SNRNP200	small nuclear ribonucleoprotein 200kDa (U5)	8
O75787	ATP6AP2	ATPase, H+ transporting, lysosomal accessory protein 2	2
O75821	EIF3G	eukaryotic translation initiation factor 3, subunit G	7
O75822	EIF3J	eukaryotic translation initiation factor 3, subunit J	6
O75874	IDH1	isocitrate dehydrogenase 1 (NADP+), soluble	33
O75937	DNAJC8	DnaJ (Hsp40) homolog, subfamily C, member 8	7
O76003	GLRX3	glutaredoxin 3	2
O76021	RSL1D1	ribosomal L1 domain containing 1	2
O76061	STC2	stanniocalcin 2	5

O76094	SRP72	signal recognition particle 72kDa	2
O94760	DDAH1	dimethylarginine dimethylaminohydrolase 1	6
O94808	GFPT2	glutamine-fructose-6-phosphate transaminase 2	1
O94855	SEC24D	SEC24 family member D	1
O94903	PROSC	proline synthetase co-transcribed homolog (bacterial)	1
O94907	DKK1	dickkopf WNT signaling pathway inhibitor 1	1
O94985	CLSTN1	calsyntenin 1	16
O94992	HEXIM1	hexamethylene bis-acetamide inducible 1	2
O95163	IKBKAP	inhibitor of kappa light polypeptide gene enhancer in B-cells, kinase complex-associated protein	4
O95336	PGLS	6-phosphogluconolactonase	7
O95373	IPO7	importin 7	4
O95433	AHSA1	AHA1, activator of heat shock 90kDa protein ATPase homolog 1 (yeast)	8
O95445	APOM	apolipoprotein M	6
O95721	SNAP29	synaptosomal-associated protein, 29kDa	2
O95777	LSM8	LSM8 homolog, U6 small nuclear RNA associated (S. cerevisiae)	2
O95782	AP2A1	adaptor-related protein complex 2, alpha 1 subunit	4
O95831	AIFM1	apoptosis-inducing factor, mitochondrion-associated, 1	6
O95861	BPNT1	3'(2'), 5'-bisphosphate nucleotidase 1	10
O95865	DDAH2	dimethylarginine dimethylaminohydrolase 2	7
O96019	ACTL6A	actin-like 6A	5
P00338	LDHA	lactate dehydrogenase A	23
P00352	ALDH1A1	aldehyde dehydrogenase 1 family, member A1	34
P00390	GSR	glutathione reductase	12
P00439	PAH	phenylalanine hydroxylase	2
P00441	SOD1	superoxide dismutase 1, soluble	5
P00450	CP	ceruloplasmin (ferroxidase)	5
P00491	PNP	purine nucleoside phosphorylase	7
P00505	GOT2	glutamic-oxaloacetic transaminase 2, mitochondrial	2
P00558	PGK1	phosphoglycerate kinase 1	28
P00734	F2	coagulation factor II (thrombin)	12
P00738	HP	haptoglobin	8
P00742	F10	coagulation factor X	6
P00747	PLG	plasminogen	10
P00748	F12	coagulation factor XII (Hageman factor)	7
P00918	CA2	carbonic anhydrase II	6
P00995	SPINK1	serine peptidase inhibitor, Kazal type 1	7
P01008	SERPINC1	serpin peptidase inhibitor, clade C (antithrombin), member 1	10

P01009	SERPINA1	serpin peptidase inhibitor, clade A (alpha-1 antiproteinase, antitrypsin), member 1	29
P01019	AGT	angiotensinogen (serpin peptidase inhibitor, clade A, member 8)	10
P01024	C3	complement component 3	90
P01031	C5	complement component 5	35
P01033	TIMP1	TIMP metallopeptidase inhibitor 1	1
P01034	CST3	cystatin C	8
P01042	KNG1	kininogen 1	24
P01308	INS	insulin	2
P02452	COL1A1	collagen, type I, alpha 1	3
P02458	COL2A1	collagen, type II, alpha 1	22
P02545	LMNA	lamin A/C	13
P02647	APOA1*	apolipoprotein A-I	6
P02649	APOE	apolipoprotein E	46
P02652	APOA2	apolipoprotein A-II	7
P02655	APOC2	apolipoprotein C-II	5
P02656	APOC3	apolipoprotein C-III	3
P02671	FGA	fibrinogen alpha chain	38
P02675	FGB	fibrinogen beta chain	20
P02749	APOH	apolipoprotein H (beta-2-glycoprotein I)	13
P02750	LRG1	leucine-rich alpha-2-glycoprotein 1	5
P02751	FN1	fibronectin 1	62
P02753	RBP4	retinol binding protein 4, plasma	14
P02760	AMBP	alpha-1-microglobulin/bikunin precursor	21
P02763	ORM1	orosomucoid 1	4
P02771	AFP	alpha-fetoprotein	55
P02774	GC	group-specific component (vitamin D binding protein)	25
P02787	TF	transferrin	91
P02790	HPX	hemopexin	6
P02795	MT2A	metallothionein 2A	1
P03950	ANG	angiogenin, ribonuclease, RNase A family, 5	4
P04004	VTN	vitronectin	16
P04075	ALDOA	aldolase A, fructose-bisphosphate	29
P04114	APOB	apolipoprotein B	151
P04217	A1BG	alpha-1-B glycoprotein	9
P04350	TUBB4A	tubulin, beta 4A class IVa	1
P04406	GAPDH	glyceraldehyde-3-phosphate dehydrogenase	25
P04424	ASL	argininosuccinate lyase	5

P04792	HSPB1	heat shock 27kDa protein 1	6
P05067	APP	amyloid beta (A4) precursor protein	8
P05089	ARG1	arginase 1	4
P05114	HMGN1	high mobility group nucleosome binding domain 1	5
P05121	SERPINE1	serpin peptidase inhibitor, clade E (nexin, plasminogen activator inhibitor type 1), member 1	8
P05154	SERPINA5	serpin peptidase inhibitor, clade A (alpha-1 antiproteinase, antitrypsin), member 5	15
P05198	EIF2S1	eukaryotic translation initiation factor 2, subunit 1 alpha, 35kDa	7
P05204	HMGN2	high mobility group nucleosomal binding domain 2	5
P05387	RPLP2	ribosomal protein, large, P2	3
P05388	RPLP0	ribosomal protein, large, P0	12
P05455	SSB	Sjogren syndrome antigen B (autoantigen La)	19
P05546	SERPIND1	serpin peptidase inhibitor, clade D (heparin cofactor), member 1	12
P05783	KRT18	keratin 18, type I	24
P05997	COL5A2	collagen, type V, alpha 2	18
P06132	UROD	uroporphyrinogen decarboxylase	4
P06280	GLA	galactosidase, alpha	5
P06454	PTMA	prothymosin, alpha	9
P06493	CDK1	cyclin-dependent kinase 1	4
P06576	ATP5B	ATP synthase, H+ transporting, mitochondrial F1 complex, beta polypeptide	4
P06681	C2	complement component 2	2
P06730	EIF4E	eukaryotic translation initiation factor 4E	2
P06733	ENO1	enolase 1, (alpha)	50
P06737	PYGL	phosphorylase, glycogen, liver	11
P06744	GPI	glucose-6-phosphate isomerase	27
P06748	NPM1*	nucleophosmin (nucleolar phosphoprotein B23, numatrin)	5
P06748	NPM1*	nucleophosmin (nucleolar phosphoprotein B23, numatrin)	2
P06753	TPM3*	tropomyosin 3	17
P06753	TPM3*	tropomyosin 3	1
P07108	DBI	diazepam binding inhibitor (GABA receptor modulator, acyl-CoA binding protein)	11
P07148	FABP1	fatty acid binding protein 1, liver	11
P07205	PGK2	phosphoglycerate kinase 2	1
P07225	PROS1	protein S (alpha)	12
P07237	P4HB	prolyl 4-hydroxylase, beta polypeptide	19
P07305	H1F0	H1 histone family, member 0	2
P07307	ASGR2	asialoglycoprotein receptor 2	2

P07339	CTSD	cathepsin D	7
P07355	ANXA2	annexin A2	10
P07384	CAPN1	calpain 1, (mu/I) large subunit	2
P07437	TUBB	tubulin, beta class I	3
P07686	HEXB	hexosaminidase B (beta polypeptide)	2
P07711	CTSL	cathepsin L	1
P07737	PFN1	profilin 1	17
P07741	APRT	adenine phosphoribosyltransferase	4
P07814	EPRS	glutamyl-prolyl-tRNA synthetase	31
P07858	CTSB	cathepsin B	3
P07900	HSP90AA1	heat shock protein 90kDa alpha (cytosolic), class A member 1	39
P07910	HNRNPC	heterogeneous nuclear ribonucleoprotein C (C1/C2)	20
P07954	FH	fumarate hydratase	4
P07996	THBS1	thrombospondin 1	2
P08185	SERPINA6	serpin peptidase inhibitor, clade A (alpha-1 antiproteinase, antitrypsin), member 6	5
P08238	HSP90AB1	heat shock protein 90kDa alpha (cytosolic), class B member 1	34
P08243	ASNS	asparagine synthetase (glutamine-hydrolyzing)	11
P08579	SNRPB2	small nuclear ribonucleoprotein polypeptide B	3
P08603	CFH	complement factor H	31
P08621	SNRNP70	small nuclear ribonucleoprotein 70kDa (U1)	8
P08670	VIM	vimentin	15
P08697	SERPINF2	serpin peptidase inhibitor, clade F (alpha-2 antiplasmin, pigment epithelium derived factor), member 2	12
P08758	ANXA5	annexin A5	5
P09012	SNRPA	small nuclear ribonucleoprotein polypeptide A	4
P09104	ENO2	enolase 2 (gamma, neuronal)	1
P09132	SRP19	signal recognition particle 19kDa	1
P09211	GSTP1	glutathione S-transferase pi 1	9
P09327	VIL1	villin 1	20
P09341	CXCL1	chemokine (C-X-C motif) ligand 1 (melanoma growth stimulating activity, alpha)	1
P09382	LGALS1	lectin, galactoside-binding, soluble, 1	5
P09417	QDPR	quinoid dihydropteridine reductase	10
P09429	HMGB1	high mobility group box 1	18
P09486	SPARC	secreted protein, acidic, cysteine-rich (osteonectin)	2
P09496	CLTA	clathrin, light chain A	7
P09622	DLD	dihydrolipoamide dehydrogenase	4
P09651	HNRNPA1	heterogeneous nuclear ribonucleoprotein A1	19

P09661	SNRPA1	small nuclear ribonucleoprotein polypeptide A'	2
P09871	C1S	complement component 1, s subcomponent	3
P09874	PARP1	poly (ADP-ribose) polymerase 1	24
P09936	UCHL1*	ubiquitin carboxyl-terminal esterase L1 (ubiquitin thiolesterase)	5
P09960	LTA4H	leukotriene A4 hydrolase	8
P09972	ALDOC	aldolase C, fructose-bisphosphate	9
P0C0S5	H2AFZ	H2A histone family, member Z	1
P0CB38	PABPC4L	poly(A) binding protein, cytoplasmic 4-like	1
P10155	TROVE2	TROVE domain family, member 2	2
P10412	HIST1H1E	histone cluster 1, H1e	5
P10586	PTPRF	protein tyrosine phosphatase, receptor type, F	10
P10599	TXN	thioredoxin	9
P10619	CTSA	cathepsin A	3
P10644	PRKAR1A	protein kinase, cAMP-dependent, regulatory, type I, alpha	2
P10646	TFPI	tissue factor pathway inhibitor (lipoprotein-associated coagulation inhibitor)	3
P10768	ESD	esterase D	4
P10809	HSPD1	heat shock 60kDa protein 1 (chaperonin)	19
P10909	CLU	clusterin	31
P11021	HSPA5	heat shock 70kDa protein 5 (glucose-regulated protein, 78kDa)	29
P11047	LAMC1	laminin, gamma 1 (formerly LAMB2)	10
P11142	HSPA8	heat shock 70kDa protein 8	32
P11172	UMPS	uridine monophosphate synthetase	4
P11216	PYGB	phosphorylase, glycogen; brain	4
P11233	RALA	v-ral simian leukemia viral oncogene homolog A (ras related)	4
P11387	TOP1	topoisomerase (DNA) I	4
P11413	G6PD	glucose-6-phosphate dehydrogenase	20
P11717	IGF2R	insulin-like growth factor 2 receptor	2
P11766	ADH5	alcohol dehydrogenase 5 (class III), chi polypeptide	17
P11940	PABPC1	poly(A) binding protein, cytoplasmic 1	7
P12004	PCNA	proliferating cell nuclear antigen	14
P12081	HARS	histidyl-tRNA synthetase	12
P12109	COL6A1	collagen, type VI, alpha 1	4
P12259	F5	coagulation factor V (proaccelerin, labile factor)	6
P12268	IMPDH2	IMP (inosine 5'-monophosphate) dehydrogenase 2	10
P12270	TPR	translocated promoter region, nuclear basket protein	7
P12277	CKB	creatine kinase, brain	13
P12955	PEPD	peptidase D	8

P12956	XRCC6	X-ray repair complementing defective repair in Chinese hamster cells 6	19
P13010	XRCC5	X-ray repair complementing defective repair in Chinese hamster cells 5 (double-strand-break rejoining)	15
P13284	IFI30	interferon, gamma-inducible protein 30	2
P13489	RNH1	ribonuclease/angiogenin inhibitor 1	9
P13611	VCAN	versican	2
P13639	EEF2	eukaryotic translation elongation factor 2	54
P13667	PDIA4	protein disulfide isomerase family A, member 4	16
P13693	TPT1	tumor protein, translationally-controlled 1	9
P13797	PLS3	plastin 3	16
P13861	PRKAR2A	protein kinase, cAMP-dependent, regulatory, type II, alpha	4
P14174	MIF	macrophage migration inhibitory factor (glycosylation-inhibiting factor)	4
P14209	CD99	CD99 molecule	1
P14314	PRKCSH	protein kinase C substrate 80K-H	11
P14324	FDPS	farnesyl diphosphate synthase	9
P14543	NID1	nidogen 1	37
P14550	AKR1A1	aldo-keto reductase family 1, member A1 (aldehyde reductase)	9
P14618	PKM	pyruvate kinase, muscle	29
P14625	HSP90B1	heat shock protein 90kDa beta (Grp94), member 1	21
P14735	IDE	insulin-degrading enzyme	5
P14854	COX6B1	cytochrome c oxidase subunit VIb polypeptide 1 (ubiquitous)	2
P14866	HNRNPL*	heterogeneous nuclear ribonucleoprotein L	13
P14868	DARS	aspartyl-tRNA synthetase	13
P15104	GLUL	glutamate-ammonia ligase	7
P15121	AKR1B1	aldo-keto reductase family 1, member B1 (aldose reductase)	3
P15151	PVR	poliovirus receptor	3
P15169	CPN1	carboxypeptidase N, polypeptide 1	3
P15170	GSPT1	G1 to S phase transition 1	7
P15311	EZR	ezrin	27
P15531	NME1	NME/NM23 nucleoside diphosphate kinase 1	1
P15559	NQO1	NAD(P)H dehydrogenase, quinone 1	8
P15880	RPS2	ribosomal protein S2	11
P15907	ST6GAL1	ST6 beta-galactosamide alpha-2,6-sialyltranferase 1	9
P15927	RPA2	replication protein A2, 32kDa	5
P16152	CBR1	carbonyl reductase 1	8
P16401	HIST1H1B	histone cluster 1, H1b	7
P16455	MGMT	O-6-methylguanine-DNA methyltransferase	1
P16870	CPE	carboxypeptidase E	3

P16949	STMN1	stathmin 1	25
P17066	HSPA6	heat shock 70kDa protein 6 (HSP70B')	1
P17096	HMGA1	high mobility group AT-hook 1	3
P17174	GOT1	glutamic-oxaloacetic transaminase 1, soluble	16
P17612	PRKACA	protein kinase, cAMP-dependent, catalytic, alpha	5
P17812	CTPS1	CTP synthase 1	6
P17844	DDX5	DEAD (Asp-Glu-Ala-Asp) box helicase 5	8
P17858	PFKL	phosphofructokinase, liver	3
P17931	LGALS3	lectin, galactoside-binding, soluble, 3	4
P17936	IGFBP3	insulin-like growth factor binding protein 3	3
P17980	PSMC3	proteasome (prosome, macropain) 26S subunit, ATPase, 3	6
P17987	TCP1	t-complex 1	20
P18065	IGFBP2	insulin-like growth factor binding protein 2, 36kDa	1
P18077	RPL35A	ribosomal protein L35a	7
P18085	ARF4	ADP-ribosylation factor 4	3
P18124	RPL7	ribosomal protein L7	6
P18206	VCL	vinculin	16
P18621	RPL17	ribosomal protein L17	3
P18669	PGAM1	phosphoglycerate mutase 1 (brain)	15
P19022	CDH2	cadherin 2, type 1, N-cadherin (neuronal)	6
P19338	NCL	nucleolin	53
P19623	SRM	spermidine synthase	7
P19652	ORM2	orosomucoid 2	3
P19823	ITIH2	inter-alpha-trypsin inhibitor heavy chain 2	31
P19827	ITIH1	inter-alpha-trypsin inhibitor heavy chain 1	3
P19883	FST	follistatin	3
P20042	EIF2S2	eukaryotic translation initiation factor 2, subunit 2 beta, 38kDa	5
P20290	BTF3	basic transcription factor 3	2
P20618	PSMB1	proteasome (prosome, macropain) subunit, beta type, 1	9
P20700	LMNB1	lamin B1	7
P20810	CAST	calpastatin	9
P20827	EFNA1	ephrin-A1	4
P20962	PTMS	parathymosin	8
P21283	ATP6V1C1	ATPase, H+ transporting, lysosomal 42kDa, V1 subunit C1	3
P21333	FLNA	filamin A, alpha	25
P21399	ACO1	aconitase 1, soluble	5
P22059	OSBP	oxysterol binding protein	10

P22102	GART	phosphoribosylglycinamide formyltransferase, phosphoribosylglycinamide synthetase, phosphoribosylaminoimidazole synthetase	7
P22234	PAICS	phosphoribosylaminoimidazole carboxylase, phosphoribosylaminoimidazole succinocarboxamide synthetase	14
P22307	SCP2	sterol carrier protein 2	2
P22314	UBA1	ubiquitin-like modifier activating enzyme 1	26
P22392	NME2	NME/NM23 nucleoside diphosphate kinase 2	1
P22492	HIST1H1T	histone cluster 1, H1t	1
P22626	HNRNPA2B1*	heterogeneous nuclear ribonucleoprotein A2/B1	4
P22626	HNRNPA2B1*	heterogeneous nuclear ribonucleoprotein A2/B1	1
P23142	FBLN1*	fibulin 1	2
P23193	TCEA1	transcription elongation factor A (SII), 1	11
P23246	SFPQ	splicing factor proline/glutamine-rich	28
P23284	PPIB	peptidylprolyl isomerase B (cyclophilin B)	15
P23381	WARS	tryptophanyl-tRNA synthetase	8
P23396	RPS3	ribosomal protein S3	16
P23434	GCSH	glycine cleavage system protein H (aminomethyl carrier)	1
P23468	PTPRD	protein tyrosine phosphatase, receptor type, D	1
P23526	AHCY	adenosylhomocysteinase	29
P23919	DTYMK	deoxythymidylate kinase (thymidylate kinase)	1
P23921	RRM1	ribonucleotide reductase M1	5
P24534	EEF1B2	eukaryotic translation elongation factor 1 beta 2	9
P24666	ACP1	acid phosphatase 1, soluble	4
P25205	MCM3	minichromosome maintenance complex component 3	18
P25325	MPST	mercaptopyruvate sulfurtransferase	3
P25398	RPS12	ribosomal protein S12	10
P25685	DNAJB1	DnaJ (Hsp40) homolog, subfamily B, member 1	2
P25786	PSMA1	proteasome (prosome, macropain) subunit, alpha type, 1	14
P25787	PSMA2	proteasome (prosome, macropain) subunit, alpha type, 2	6
P25788	PSMA3	proteasome (prosome, macropain) subunit, alpha type, 3	14
P25789	PSMA4	proteasome (prosome, macropain) subunit, alpha type, 4	10
P25815	S100P	S100 calcium binding protein P	6
P26038	MSN	moesin	1
P26196	DDX6	DEAD (Asp-Glu-Ala-Asp) box helicase 6	3
P26368	U2AF2	U2 small nuclear RNA auxiliary factor 2	7
P26373	RPL13*	ribosomal protein L13	5
P26583	HMGB2	high mobility group box 2	14
P26599	PTBP1	polypyrimidine tract binding protein 1	15

P26641	EEF1G	eukaryotic translation elongation factor 1 gamma	20
P27348	YWHAQ	tyrosine 3-monooxygenase/tryptophan 5-monooxygenase activation protein, theta	13
P27361	MAPK3	mitogen-activated protein kinase 3	1
P27635	RPL10	ribosomal protein L10	2
P27694	RPA1	replication protein A1, 70kDa	7
P27695	APEX1	APEX nuclease (multifunctional DNA repair enzyme) 1	11
P27708	CAD	carbamoyl-phosphate synthetase 2, aspartate transcarbamylase, and dihydroorotase	2
P27797	CALR	calreticulin	4
P28066	PSMA5	proteasome (prosome, macropain) subunit, alpha type, 5	10
P28070	PSMB4	proteasome (prosome, macropain) subunit, beta type, 4	5
P28072	PSMB6	proteasome (prosome, macropain) subunit, beta type, 6	5
P28074	PSMB5	proteasome (prosome, macropain) subunit, beta type, 5	11
P28482	MAPK1	mitogen-activated protein kinase 1	9
P28799	GRN	granulin	10
P28838	LAP3	leucine aminopeptidase 3	16
P29218	IMPA1	inositol(myo)-1(or 4)-monophosphatase 1	3
P29279	CTGF	connective tissue growth factor	6
P29401	TKT	transketolase	29
P29622	SERPINA4	serpin peptidase inhibitor, clade A (alpha-1 antiproteinase, antitrypsin), member 4	4
P29966	MARCKS	myristoylated alanine-rich protein kinase C substrate	4
P30040	ERP29	endoplasmic reticulum protein 29	6
P30041	PRDX6	peroxiredoxin 6	12
P30043	BLVRB	biliverdin reductase B	4
P30044	PRDX5	peroxiredoxin 5	4
P30048	PRDX3	peroxiredoxin 3	6
P30050	RPL12	ribosomal protein L12	8
P30085	CMPK1	cytidine monophosphate (UMP-CMP) kinase 1, cytosolic	9
P30086	PEBP1	phosphatidylethanolamine binding protein 1	12
P30101	PDIA3*	protein disulfide isomerase family A, member 3	10
P30153	PPP2R1A	protein phosphatase 2, regulatory subunit A, alpha	13
P30419	NMT1	N-myristoyltransferase 1	9
P30520	ADSS	adenylosuccinate synthase	11
P30566	ADSL	adenylosuccinate lyase	8
P30622	CLIP1	CAP-GLY domain containing linker protein 1	2
P30626	SRI	sorcin	3
P30740	SERPINB1	serpin peptidase inhibitor, clade B (ovalbumin), member 1	11

P30990	NTS	neurotensin	6
P31150	GDI1	GDP dissociation inhibitor 1	3
P31153	MAT2A	methionine adenosyltransferase II, alpha	6
P31431	SDC4	syndecan 4	2
P31689	DNAJA1	DnaJ (Hsp40) homolog, subfamily A, member 1	3
P31937	HIBADH	3-hydroxyisobutyrate dehydrogenase	1
P31939	ATIC	5-aminoimidazole-4-carboxamide ribonucleotide formyltransferase/IMP cyclohydrolase	21
P31942	HNRNPH3	heterogeneous nuclear ribonucleoprotein H3 (2H9)	2
P31946	YWHAB*	tyrosine 3-monooxygenase/tryptophan 5-monooxygenase activation protein, beta	1
P31946	YWHAB*	tyrosine 3-monooxygenase/tryptophan 5-monooxygenase activation protein, beta	1
P31947	SFN	stratifin	1
P31948	STIP1	stress-induced phosphoprotein 1	34
P31949	S100A11	S100 calcium binding protein A11	5
P32119	PRDX2	peroxiredoxin 2	11
P33176	KIF5B	kinesin family member 5B	20
P33316	DUT	deoxyuridine triphosphatase	5
P33908	MAN1A1	mannosidase, alpha, class 1A, member 1	20
P33991	MCM4	minichromosome maintenance complex component 4	10
P33992	MCM5	minichromosome maintenance complex component 5	11
P33993	MCM7	minichromosome maintenance complex component 7	14
P34096	RNASE4	ribonuclease, RNase A family, 4	9
P34896	SHMT1	serine hydroxymethyltransferase 1 (soluble)	3
P34897	SHMT2	serine hydroxymethyltransferase 2 (mitochondrial)	7
P34913	EPHX2	epoxide hydrolase 2, cytoplasmic	5
P34932	HSPA4	heat shock 70kDa protein 4	31
P35080	PFN2*	profilin 2	1
P35221	CTNNA1	catenin (cadherin-associated protein), alpha 1, 102kDa	3
P35222	CTNNB1	catenin (cadherin-associated protein), beta 1, 88kDa	2
P35237	SERPINB6	serpin peptidase inhibitor, clade B (ovalbumin), member 6	4
P35244	RPA3	replication protein A3, 14kDa	2
P35268	RPL22	ribosomal protein L22	3
P35270	SPR	sepiapterin reductase (7,8-dihydrobiopterin:NADP+ oxidoreductase)	2
P35318	ADM	adrenomedullin	1
P35443	THBS4	thrombospondin 4	6
P35520	CBS/LOC102724560	cystathionine-beta-synthase	9

P35579	MYH9	myosin, heavy chain 9, non-muscle	7
P35606	COPB2	coatomer protein complex, subunit beta 2 (beta prime)	9
P35659	DEK	DEK proto-oncogene	4
P35754	GLRX	glutaredoxin (thioltransferase)	3
P35790	CHKA	choline kinase alpha	2
P35813	PPM1A	protein phosphatase, Mg2+/Mn2+ dependent, 1A	3
P35998	PSMC2	proteasome (prosome, macropain) 26S subunit, ATPase, 2	9
P36405	ARL3	ADP-ribosylation factor-like 3	1
P36543	ATP6V1E1	ATPase, H+ transporting, lysosomal 31kDa, V1 subunit E1	2
P36551	CPOX	coproporphyrinogen oxidase	2
P36578	RPL4	ribosomal protein L4	14
P36639	NUDT1	nudix (nucleoside diphosphate linked moiety X)-type motif 1	1
P36871	PGM1	phosphoglucomutase 1	13
P36873	PPP1CC	protein phosphatase 1, catalytic subunit, gamma isozyme	2
P36955	SERPINF1	serpin peptidase inhibitor, clade F (alpha-2 antiplasmin, pigment epithelium derived factor), member 1	15
P37108	SRP14	signal recognition particle 14kDa (homologous Alu RNA binding protein)	7
P37802	TAGLN2	transgelin 2	12
P37837	TALDO1	transaldolase 1	27
P38159	RBMX*	RNA binding motif protein, X-linked	8
P38606	ATP6V1A	ATPase, H+ transporting, lysosomal 70kDa, V1 subunit A	6
P38646	HSPA9	heat shock 70kDa protein 9 (mortalin)	14
P38919	EIF4A3	eukaryotic translation initiation factor 4A3	12
P39019	RPS19	ribosomal protein S19	14
P39023	RPL3	ribosomal protein L3	9
P39687	ANP32A	acidic (leucine-rich) nuclear phosphoprotein 32 family, member A	7
P39748	FEN1	flap structure-specific endonuclease 1	12
P40121	CAPG	capping protein (actin filament), gelsolin-like	7
P40123	CAP2	CAP, adenylate cyclase-associated protein, 2 (yeast)	2
P40222	TXLNA	taxilin alpha	3
P40227	CCT6A	chaperonin containing TCP1, subunit 6A (zeta 1)	16
P40429	RPL13A	ribosomal protein L13a	3
P40926	MDH2	malate dehydrogenase 2, NAD (mitochondrial)	16
P41091	EIF2S3	eukaryotic translation initiation factor 2, subunit 3 gamma, 52kDa	5
P41227	NAA10	N(alpha)-acetyltransferase 10, NatA catalytic subunit	4
P41236	PPP1R2	protein phosphatase 1, regulatory (inhibitor) subunit 2	2
P41250	GARS	glycyl-tRNA synthetase	14
P41252	IARS	isoleucyl-tRNA synthetase	9

P42025	ACTR1B	ARP1 actin-related protein 1 homolog B, centractin beta (yeast)	3
P42126	ECI1	enoyl-CoA delta isomerase 1	2
P42167	TMPO	thymopoietin	6
P42285	SKIV2L2	superkiller viralicidic activity 2-like 2 (S. cerevisiae)	4
P42345	MTOR	mechanistic target of rapamycin (serine/threonine kinase)	2
P42765	ACAA2	acetyl-CoA acyltransferase 2	1
P42766	RPL35	ribosomal protein L35	8
P42830	CXCL5	chemokine (C-X-C motif) ligand 5	3
P42858	HTT	huntingtin	3
P43034	PAFAH1B1	platelet-activating factor acetylhydrolase 1b, regulatory subunit 1 (45kDa)	6
P43487	RANBP1	RAN binding protein 1	13
P43490	NAMPT	nicotinamide phosphoribosyltransferase	7
P43686	PSMC4	proteasome (prosome, macropain) 26S subunit, ATPase, 4	3
P45877	PPIC	peptidylprolyl isomerase C (cyclophilin C)	2
P45973	CBX5	chromobox homolog 5	9
P45974	USP5	ubiquitin specific peptidase 5 (isopeptidase T)	11
P46013	MKI67	marker of proliferation Ki-67	2
P46060	RANGAP1	Ran GTPase activating protein 1	7
P46063	RECQL	RecQ helicase-like	3
P46108	CRK	v-crk avian sarcoma virus CT10 oncogene homolog	3
P46109	CRKL	v-crk avian sarcoma virus CT10 oncogene homolog-like	3
P46459	NSF	N-ethylmaleimide-sensitive factor	4
P46776	RPL27A	ribosomal protein L27a	3
P46777	RPL5	ribosomal protein L5	23
P46778	RPL21	ribosomal protein L21	5
P46779	RPL28	ribosomal protein L28	9
P46781	RPS9	ribosomal protein S9	12
P46782	RPS5	ribosomal protein S5	8
P46783	RPS10	ribosomal protein S10	7
P46926	GNPDA1	glucosamine-6-phosphate deaminase 1	5
P46940	IQGAP1	IQ motif containing GTPase activating protein 1	5
P47755	CAPZA2	capping protein (actin filament) muscle Z-line, alpha 2	1
P47813	EIF1AX	eukaryotic translation initiation factor 1A, X-linked	6
P47897	QARS	glutaminyl-tRNA synthetase	10
P47914	RPL29	ribosomal protein L29	4
P48147	PREP	prolyl endopeptidase	8
P48163	ME1	malic enzyme 1, NADP(+)-dependent, cytosolic	9
P48506	GCLC	glutamate-cysteine ligase, catalytic subunit	3

P48634	PRRC2A	proline-rich coiled-coil 2A	2
P48637	GSS	glutathione synthetase	17
P48643	CCT5*	chaperonin containing TCP1, subunit 5 (epsilon)	2
P48681	NES	nestin	5
P48723	HSPA13	heat shock protein 70kDa family, member 13	4
P48740	MASP1	mannan-binding lectin serine peptidase 1 (C4/C2 activating component of Ra-reactive factor)	5
P48741	HSPA7	heat shock 70kDa protein 7 (HSP70B)	1
P49006	MARCKSL1	MARCKS-like 1	3
P49189	ALDH9A1	aldehyde dehydrogenase 9 family, member A1	2
P49207	RPL34	ribosomal protein L34	9
P49321	NASP	nuclear autoantigenic sperm protein (histone-binding)	14
P49327	FASN	fatty acid synthase	92
P49354	FNTA	farnesyltransferase, CAAX box, alpha	3
P49366	DHPS	deoxyhypusine synthase	1
P49368	CCT3	chaperonin containing TCP1, subunit 3 (gamma)	29
P49419	ALDH7A1	aldehyde dehydrogenase 7 family, member A1	3
P49458	SRP9	signal recognition particle 9kDa	10
P49588	AARS	alanyl-tRNA synthetase	38
P49589	CARS	cysteinyl-tRNA synthetase	4
P49720	PSMB3	proteasome (prosome, macropain) subunit, beta type, 3	7
P49721	PSMB2	proteasome (prosome, macropain) subunit, beta type, 2	9
P49770	EIF2B2	eukaryotic translation initiation factor 2B, subunit 2 beta, 39kDa	1
P49773	HINT1	histidine triad nucleotide binding protein 1	7
P49792	RANBP2	RAN binding protein 2	3
P49908	SEPP1	selenoprotein P, plasma, 1	2
P49915	GMPS	guanine monphosphate synthase	3
P50120	RBP2	retinol binding protein 2, cellular	2
P50135	HNMT	histamine N-methyltransferase	8
P50440	GATM	glycine amidinotransferase (L-arginine:glycine amidinotransferase)	4
P50453	SERPINB9	serpin peptidase inhibitor, clade B (ovalbumin), member 9	5
P50454	SERPINH1*	serpin peptidase inhibitor, clade H (heat shock protein 47), member 1, (collagen binding protein 1)	1
P50502	ST13	suppression of tumorigenicity 13 (colon carcinoma) (Hsp70 interacting protein)	13
P50552	VASP	vasodilator-stimulated phosphoprotein	4
P50579	METAP2	methionyl aminopeptidase 2	3
P50895	BCAM	basal cell adhesion molecule (Lutheran blood group)	18
P50914	RPL14	ribosomal protein L14	11

P50990	CCT8	chaperonin containing TCP1, subunit 8 (theta)	27
P50991	CCT4	chaperonin containing TCP1, subunit 4 (delta)	21
P51149	RAB7A	RAB7A, member RAS oncogene family	8
P51397	DAP	death-associated protein	5
P51452	DUSP3	dual specificity phosphatase 3	4
P51665	PSMD7	proteasome (prosome, macropain) 26S subunit, non-ATPase, 7	4
P51812	RPS6KA3	ribosomal protein S6 kinase, 90kDa, polypeptide 3	13
P51857	AKR1D1	aldo-keto reductase family 1, member D1	1
P51858	HDGF	hepatoma-derived growth factor	14
P51884	LUM	lumican	1
P51991	HNRNPA3*	heterogeneous nuclear ribonucleoprotein A3	2
P51991	HNRNPA3*	heterogeneous nuclear ribonucleoprotein A3	1
P52209	PGD	phosphogluconate dehydrogenase	17
P52272	HNRNPM	heterogeneous nuclear ribonucleoprotein M	23
P52292	KPNA2	karyopherin alpha 2 (RAG cohort 1, importin alpha 1)	7
P52294	KPNA1	karyopherin alpha 1 (importin alpha 5)	2
P52565	ARHGDIA	Rho GDP dissociation inhibitor (GDI) alpha	12
P52597	HNRNPF	heterogeneous nuclear ribonucleoprotein F	3
P52657	GTF2A2	general transcription factor IIA, 2, 12kDa	1
P52788	SMS	spermine synthase	4
P52888	THOP1	thimet oligopeptidase 1	11
P52895	AKR1C1/AKR1C2*	aldo-keto reductase family 1, member C2	2
P52907	CAPZA1	capping protein (actin filament) muscle Z-line, alpha 1	5
P52926	HMGA2	high mobility group AT-hook 2	5
P53041	PPP5C	protein phosphatase 5, catalytic subunit	11
P53396	ACLY	ATP citrate lyase	35
P53582	METAP1	methionyl aminopeptidase 1	1
P53602	MVD	mevalonate (diphospho) decarboxylase	10
P53611	RABGGTB	Rab geranylgeranyltransferase, beta subunit	1
P53618	COPB1	coatomer protein complex, subunit beta 1	6
P53621	COPA	coatomer protein complex, subunit alpha	26
P53634	CTSC	cathepsin C	6
P53992	SEC24C	SEC24 family member C	3
P53999	SUB1	SUB1 homolog (S. cerevisiae)	7
P54105	CLNS1A	chloride channel, nucleotide-sensitive, 1A	1
P54136	RARS	arginyl-tRNA synthetase	14
P54577	YARS	tyrosyl-tRNA synthetase	13
P54578	USP14	ubiquitin specific peptidase 14 (tRNA-guanine transglycosylase)	11

P54652	HSPA2	heat shock 70kDa protein 2	1
P54725	RAD23A	RAD23 homolog A (S. cerevisiae)	1
P54727	RAD23B	RAD23 homolog B (S. cerevisiae)	2
P54819	AK2	adenylate kinase 2	8
P54920	NAPA	N-ethylmaleimide-sensitive factor attachment protein, alpha	2
P55010	EIF5	eukaryotic translation initiation factor 5	7
P55060	CSE1L	CSE1 chromosome segregation 1-like (yeast)	17
P55072	VCP	valosin containing protein	49
P55081	MFAP1	microfibrillar-associated protein 1	1
P55196	MLLT4	myeloid/lymphoid or mixed-lineage leukemia (trithorax homolog, Drosophila); translocated to, 4	4
P55209	NAP1L1*	nucleosome assembly protein 1-like 1	1
P55263	ADK	adenosine kinase	1
P55327	TPD52	tumor protein D52	4
P55735	SEC13	SEC13 homolog (S. cerevisiae)	4
P55786	NPEPPS	aminopeptidase puromycin sensitive	26
P55884	EIF3B	eukaryotic translation initiation factor 3, subunit B	14
P56192	MARS	methionyl-tRNA synthetase	8
P56537	EIF6	eukaryotic translation initiation factor 6	4
P58546	MTPN	myotrophin	3
P60174	TPI1	triosephosphate isomerase 1	19
P60228	EIF3E	eukaryotic translation initiation factor 3, subunit E	6
P60510	PPP4C	protein phosphatase 4, catalytic subunit	3
P60842	EIF4A1	eukaryotic translation initiation factor 4A1	10
P60866	RPS20	ribosomal protein S20	8
P60891	PRPS1	phosphoribosyl pyrophosphate synthetase 1	5
P60953	CDC42	cell division cycle 42	3
P60981	DSTN	destrin (actin depolymerizing factor)	5
P61006	RAB8A	RAB8A, member RAS oncogene family	1
P61019	RAB2A	RAB2A, member RAS oncogene family	1
P61077	UBE2D3	ubiquitin-conjugating enzyme E2D 3	2
P61081	UBE2M	ubiquitin-conjugating enzyme E2M	9
P61086	UBE2K	ubiquitin-conjugating enzyme E2K	5
P61088	UBE2N	ubiquitin-conjugating enzyme E2N	8
P61106	RAB14	RAB14, member RAS oncogene family	3
P61158	ACTR3	ARP3 actin-related protein 3 homolog (yeast)	7
P61221	ABCE1*	ATP-binding cassette, sub-family E (OABP), member 1	2
P61224	RAP1B	RAP1B, member of RAS oncogene family	2

P61247	RPS3A	ribosomal protein S3A	20
P61254	RPL26	ribosomal protein L26	1
P61289	PSME3	proteasome (prosome, macropain) activator subunit 3 (PA28 gamma; Ki)	3
P61313	RPL15	ribosomal protein L15	5
P61326	MAGOH	mago-nashi homolog, proliferation-associated (Drosophila)	1
P61353	RPL27	ribosomal protein L27	5
P61513	RPL37A	ribosomal protein L37a	2
P61586	RHOA	ras homolog family member A	4
P61604	HSPE1	heat shock 10kDa protein 1	14
P61626	LYZ	lysozyme	8
P61758	VBP1	von Hippel-Lindau binding protein 1	6
P61769	B2M	beta-2-microglobulin	5
P61927	RPL37	ribosomal protein L37	4
P61956	SUMO2	small ubiquitin-like modifier 2	5
P61960	UFM1	ubiquitin-fold modifier 1	2
P61970	NUTF2	nuclear transport factor 2	4
P61978	HNRNPK	heterogeneous nuclear ribonucleoprotein K	22
P61981	YWHAG	tyrosine 3-monooxygenase/tryptophan 5-monooxygenase activation protein, gamma	13
P62081	RPS7	ribosomal protein S7	5
P62136	PPP1CA	protein phosphatase 1, catalytic subunit, alpha isozyme	4
P62191	PSMC1	proteasome (prosome, macropain) 26S subunit, ATPase, 1	9
P62195	PSMC5	proteasome (prosome, macropain) 26S subunit, ATPase, 5	5
P62241	RPS8	ribosomal protein S8	11
P62244	RPS15A	ribosomal protein S15a	6
P62249	RPS16	ribosomal protein S16	12
P62253	UBE2G1	ubiquitin-conjugating enzyme E2G 1	2
P62258	YWHAE	tyrosine 3-monooxygenase/tryptophan 5-monooxygenase activation protein, epsilon	18
P62263	RPS14	ribosomal protein S14	7
P62266	RPS23	ribosomal protein S23	5
P62269	RPS18	ribosomal protein S18	8
P62273	RPS29	ribosomal protein S29	3
P62277	RPS13	ribosomal protein S13	6
P62280	RPS11	ribosomal protein S11	17
P62304	SNRPE	small nuclear ribonucleoprotein polypeptide E	2
P62306	SNRPF	small nuclear ribonucleoprotein polypeptide F	3
P62310	LSM3	LSM3 homolog, U6 small nuclear RNA associated (S. cerevisiae)	3

P62312	LSM6	LSM6 homolog, U6 small nuclear RNA associated (S. cerevisiae)	5
P62314	SNRPD1	small nuclear ribonucleoprotein D1 polypeptide 16kDa	1
P62316	SNRPD2	small nuclear ribonucleoprotein D2 polypeptide 16.5kDa	3
P62318	SNRPD3	small nuclear ribonucleoprotein D3 polypeptide 18kDa	3
P62328	TMSB10/TMSB4X*	thymosin beta 10	3
P62333	PSMC6	proteasome (prosome, macropain) 26S subunit, ATPase, 6	4
P62424	RPL7A	ribosomal protein L7a	6
P62495	ETF1	eukaryotic translation termination factor 1	3
P62633	CNBP	CCHC-type zinc finger, nucleic acid binding protein	11
P62701	RPS4X	ribosomal protein S4, X-linked	9
P62750	RPL23A	ribosomal protein L23a	4
P62753	RPS6	ribosomal protein S6	16
P62760	VSNL1	visinin-like 1	2
P62805	H4/A	Histone H4	10
P62820	RAB1A	RAB1A, member RAS oncogene family	1
P62829	RPL23	ribosomal protein L23	4
P62841	RPS15	ribosomal protein S15	2
P62851	RPS25	ribosomal protein S25	6
P62854	RPS26	ribosomal protein S26	4
P62857	RPS28	ribosomal protein S28	10
P62873	GNB1	guanine nucleotide binding protein (G protein), beta polypeptide 1	3
P62877	RBX1	ring-box 1, E3 ubiquitin protein ligase	2
P62879	GNB2	guanine nucleotide binding protein (G protein), beta polypeptide 2	1
P62888	RPL30	ribosomal protein L30	7
P62899	RPL31	ribosomal protein L31	2
P62906	RPL10A	ribosomal protein L10a	13
P62913	RPL11	ribosomal protein L11	7
P62917	RPL8	ribosomal protein L8	6
P62937	PPIA	peptidylprolyl isomerase A (cyclophilin A)	20
P62942	FKBP1A	FK506 binding protein 1A, 12kDa	6
P62979	RPS27A	ribosomal protein S27a	3
P62987	UBA52	ubiquitin A-52 residue ribosomal protein fusion product 1	1
P62993	GRB2	growth factor receptor-bound protein 2	8
P62995	TRA2B	transformer 2 beta homolog (Drosophila)	2
P63000	RAC1	ras-related C3 botulinum toxin substrate 1 (rho family, small GTP binding protein Rac1)	9
P63010	AP2B1	adaptor-related protein complex 2, beta 1 subunit	1
P63151	PPP2R2A	protein phosphatase 2, regulatory subunit B, alpha	2

P63167	DYNLL1	dynein, light chain, LC8-type 1	2
P63173	RPL38	ribosomal protein L38	1
P63218	GNG5	guanine nucleotide binding protein (G protein), gamma 5	2
P63220	RPS21	ribosomal protein S21	9
P63244	GNB2L1	guanine nucleotide binding protein (G protein), beta polypeptide 2-like 1	15
P63272	SUPT4H1	suppressor of Ty 4 homolog 1 (S. cerevisiae)	2
P63313	TMSB10/TMSB4X*	thymosin beta 10	5
P67775	PPP2CA	protein phosphatase 2, catalytic subunit, alpha isozyme	5
P67809	YBX1	Y box binding protein 1	14
P67936	TPM4	tropomyosin 4	8
P68032	ACTC1	actin, alpha, cardiac muscle 1	1
P68104	EEF1A1	eukaryotic translation elongation factor 1 alpha 1	25
P68366	TUBA4A	tubulin, alpha 4a	1
P68371	TUBB4B	tubulin, beta 4B class IVb	2
P68402	PAFAH1B2	platelet-activating factor acetylhydrolase 1b, catalytic subunit 2 (30kDa)	2
P68431	H3FA	Histone H3.1	8
P69905	HBA1	Alpha-globin	1
P78324	SIRPA	signal-regulatory protein alpha	1
P78330	PSPH	phosphoserine phosphatase	11
P78371	CCT2	chaperonin containing TCP1, subunit 2 (beta)	22
P78417	GSTO1	glutathione S-transferase omega 1	10
P78504	JAG1	jagged 1	6
P78509	RELN	reelin	4
P78527	PRKDC	protein kinase, DNA-activated, catalytic polypeptide	41
P78556	CCL20	chemokine (C-C motif) ligand 20	3
P80297	MT1X	metallothionein 1X	1
P80303	NUCB2	nucleobindin 2	1
P82979	SARNP	SAP domain containing ribonucleoprotein	11
P83731	RPL24	ribosomal protein L24	8
P83916	CBX1	chromobox homolog 1	3
P84085	ARF5	ADP-ribosylation factor 5	1
P84090	ERH	enhancer of rudimentary homolog (Drosophila)	9
P84098	RPL19	ribosomal protein L19	8
P84101	SERF2	small EDRK-rich factor 2	5
P84103	SRSF3	serine/arginine-rich splicing factor 3	3
P98160	HSPG2	heparan sulfate proteoglycan 2	11
P98175	RBM10	RNA binding motif protein 10	2

P98179	RBM3	RNA binding motif (RNP1, RRM) protein 3	4
P99999	CYCS	cytochrome c, somatic	7
Q00341	HDLBP	high density lipoprotein binding protein	14
Q00535	CDK5	cyclin-dependent kinase 5	1
Q00610	CLTC	clathrin, heavy chain (Hc)	39
Q00688	FKBP3	FK506 binding protein 3, 25kDa	16
Q00796	SORD	sorbitol dehydrogenase	11
Q00839	HNRNPU	heterogeneous nuclear ribonucleoprotein U (scaffold attachment factor A)	16
Q01082	SPTBN1*	spectrin, beta, non-erythrocytic 1	5
Q01082	SPTBN1*	spectrin, beta, non-erythrocytic 1	1
Q01085	TIAL1	TIA1 cytotoxic granule-associated RNA binding protein-like 1	1
Q01105	SET	SET nuclear proto-oncogene	10
Q01415	GALK2	galactokinase 2	4
Q01469	FABP5	fatty acid binding protein 5 (psoriasis-associated)	16
Q01518	CAP1	CAP, adenylate cyclase-associated protein 1 (yeast)	22
Q01581	HMGCS1	3-hydroxy-3-methylglutaryl-CoA synthase 1 (soluble)	19
Q01844	EWSR1	EWS RNA-binding protein 1	3
Q01970	PLCB3	phospholipase C, beta 3 (phosphatidylinositol-specific)	2
Q02539	HIST1H1A	histone cluster 1, H1a	1
Q02543	RPL18A	ribosomal protein L18a	4
Q02790	FKBP4	FK506 binding protein 4, 59kDa	18
Q02818	NUCB1	nucleobindin 1	21
Q02878	RPL6	ribosomal protein L6	6
Q03154	ACY1	aminoacylase 1	3
Q03252	LMNB2	lamin B2	8
Q04323	UBXN1	UBX domain protein 1	5
Q04446	GBE1	glucan (1,4-alpha-), branching enzyme 1	2
Q04760	GLO1	glyoxalase I	10
Q04828	AKR1C1/AKR1C2*	aldo-keto reductase family 1, member C2	4
Q04837	SSBP1	single-stranded DNA binding protein 1, mitochondrial	1
Q04917	YWHAH	tyrosine 3-monooxygenase/tryptophan 5-monooxygenase activation protein, eta	9
Q05048	CSTF1	cleavage stimulation factor, 3' pre-RNA, subunit 1, 50kDa	3
Q05707	COL14A1	collagen, type XIV, alpha 1	3
Q06124	PTPN11	protein tyrosine phosphatase, non-receptor type 11	4
Q06210	GFPT1	glutamine--fructose-6-phosphate transaminase 1	7
Q06265	EXOSC9	exosome component 9	1

Q06481	APLP2	amyloid beta (A4) precursor-like protein 2	4
Q06520	SULT2A1	sulfotransferase family, cytosolic, 2A, dehydroepiandrosterone (DHEA)-preferring, member 1	3
Q06830	PRDX1	peroxiredoxin 1	20
Q06HC4	BNC2	basonuclin 2	2
Q07020	RPL18	ribosomal protein L18	6
Q07666	KHDRBS1	KH domain containing, RNA binding, signal transduction associated 1	12
Q07812	BAX	BCL2-associated X protein	4
Q07955	SRSF1	serine/arginine-rich splicing factor 1	13
Q08043	ACTN3	actinin, alpha 3 (gene/pseudogene)	1
Q08170	SRSF4	serine/arginine-rich splicing factor 4	1
Q08211	DHX9	DEAH (Asp-Glu-Ala-His) box helicase 9	19
Q08257	CRYZ	crystallin, zeta (quinone reductase)	12
Q08380	LGALS3BP	lectin, galactoside-binding, soluble, 3 binding protein	7
Q08752	PPID	peptidylprolyl isomerase D	6
Q08830	FGL1	fibrinogen-like 1	8
Q08945	SSRP1	structure specific recognition protein 1	12
Q09028	RBBP4	retinoblastoma binding protein 4	4
Q09666	AHNAK	AHNAK nucleoprotein	9
Q10471	GALNT2	polypeptide N-acetylgalactosaminyltransferase 2	3
Q10567	AP1B1	adaptor-related protein complex 1, beta 1 subunit	3
Q10570	CPSF1	cleavage and polyadenylation specific factor 1, 160kDa	4
Q10713	PMPCA	peptidase (mitochondrial processing) alpha	1
Q12792	TWF1	twinfilin actin-binding protein 1	4
Q12805	EFEMP1	EGF containing fibulin-like extracellular matrix protein 1	11
Q12874	SF3A3	splicing factor 3a, subunit 3, 60kDa	8
Q12888	TP53BP1	tumor protein p53 binding protein 1	3
Q12904	AIMP1	aminoacyl tRNA synthetase complex-interacting multifunctional protein 1	7
Q12905	ILF2	interleukin enhancer binding factor 2	5
Q12907	LMAN2	lectin, mannose-binding 2	5
Q12931	TRAP1	TNF receptor-associated protein 1	5
Q13011	ECH1	enoyl CoA hydratase 1, peroxisomal	5
Q13017	ARHGAP5	Rho GTPase activating protein 5	8
Q13033	STRN3	striatin, calmodulin binding protein 3	2
Q13045	FLII	flightless I homolog (Drosophila)	3
Q13057	COASY	CoA synthase	1
Q13093	PLA2G7	phospholipase A2, group VII (platelet-activating factor acetylhydrolase, plasma)	2

Q13098	GPS1	G protein pathway suppressor 1	6
Q13126	MTAP	methylthioadenosine phosphorylase	4
Q13151	HNRNPA0	heterogeneous nuclear ribonucleoprotein A0	4
Q13155	AIMP2	aminoacyl tRNA synthetase complex-interacting multifunctional protein 2	2
Q13162	PRDX4	peroxiredoxin 4	3
Q13177	PAK2	p21 protein (Cdc42/Rac)-activated kinase 2	10
Q13185	CBX3	chromobox homolog 3	8
Q13200	PSMD2	proteasome (prosome, macropain) 26S subunit, non-ATPase, 2	7
Q13247	SRSF6	serine/arginine-rich splicing factor 6	2
Q13263	TRIM28	tripartite motif containing 28	19
Q13283	G3BP1	GTPase activating protein (SH3 domain) binding protein 1	8
Q13310	PABPC4	poly(A) binding protein, cytoplasmic 4 (inducible form)	6
Q13347	EIF3I	eukaryotic translation initiation factor 3, subunit I	10
Q13363	CTBP1	C-terminal binding protein 1	3
Q13404	UBE2V1	ubiquitin-conjugating enzyme E2 variant 1	5
Q13435	SF3B2	splicing factor 3b, subunit 2, 145kDa	7
Q13442	PDAP1	PDGFA associated protein 1	9
Q13443	ADAM9	ADAM metallopeptidase domain 9	3
Q13464	ROCK1	Rho-associated, coiled-coil containing protein kinase 1	3
Q13509	TUBB3	tubulin, beta 3 class III	2
Q13526	PIN1	peptidylprolyl cis/trans isomerase, NIMA-interacting 1	5
Q13541	EIF4EBP1	eukaryotic translation initiation factor 4E binding protein 1	3
Q13542	EIF4EBP2	eukaryotic translation initiation factor 4E binding protein 2	1
Q13576	IQGAP2	IQ motif containing GTPase activating protein 2	3
Q13595	TRA2A	transformer 2 alpha homolog (Drosophila)	1
Q13596	SNX1	sorting nexin 1	1
Q13616	CUL1	cullin 1	3
Q13619	CUL4A	cullin 4A	1
Q13620	CUL4B	cullin 4B	3
Q13630	TSTA3	tissue specific transplantation antigen P35B	2
Q13740	ALCAM	activated leukocyte cell adhesion molecule	4
Q13822	ENPP2	ectonucleotide pyrophosphatase/phosphodiesterase 2	5
Q13867	BLMH	bleomycin hydrolase	11
Q13885	TUBB2A	tubulin, beta 2A class IIa	4
Q13907	IDI1	isopentenyl-diphosphate delta isomerase 1	4
Q14008	CKAP5	cytoskeleton associated protein 5	4
Q14011	CIRBP	cold inducible RNA binding protein	3

Q14103	HNRNPD	heterogeneous nuclear ribonucleoprotein D (AU-rich element RNA binding protein 1, 37kDa)	13
Q14118	DAG1	dystroglycan 1 (dystrophin-associated glycoprotein 1)	7
Q14151	SAFB2	scaffold attachment factor B2	1
Q14152	EIF3A	eukaryotic translation initiation factor 3, subunit A	26
Q14157	UBAP2L	ubiquitin associated protein 2-like	3
Q14166	TTLL12	tubulin tyrosine ligase-like family member 12	4
Q14203	DCTN1	dynactin 1	8
Q14204	DYNC1H1	dynein, cytoplasmic 1, heavy chain 1	33
Q14232	EIF2B1	eukaryotic translation initiation factor 2B, subunit 1 alpha, 26kDa	3
Q14240	EIF4A2*	eukaryotic translation initiation factor 4A2	1
Q14247	CTTN	cortactin	13
Q14320	FAM50A	family with sequence similarity 50, member A	1
Q14353	GAMT	guanidinoacetate N-methyltransferase	5
Q14444	CAPRIN1	cell cycle associated protein 1	4
Q14498	RBM39	RNA binding motif protein 39	1
Q14520	HABP2	hyaluronan binding protein 2	4
Q14566	MCM6	minichromosome maintenance complex component 6	16
Q14651	PLS1	plastin 1	2
Q14683	SMC1A	structural maintenance of chromosomes 1A	7
Q14691	GINS1	GINS complex subunit 1 (Psf1 homolog)	1
Q14694	USP10	ubiquitin specific peptidase 10	1
Q14697	GANAB	glucosidase, alpha; neutral AB	17
Q14847	LASP1	LIM and SH3 protein 1	12
Q14914	PTGR1	prostaglandin reductase 1	10
Q14974	KPNB1	karyopherin (importin) beta 1	19
Q14978	NOLC1	nucleolar and coiled-body phosphoprotein 1	7
Q14980	NUMA1	nuclear mitotic apparatus protein 1	7
Q15008	PSMD6	proteasome (prosome, macropain) 26S subunit, non-ATPase, 6	4
Q15018	FAM175B	family with sequence similarity 175, member B	1
Q15019	DIFF6	septin 2	7
Q15024	EXOSC7	exosome component 7	2
Q15027	ACAP1	ArfGAP with coiled-coil, ankyrin repeat and PH domains 1	2
Q15029	EFTUD2	elongation factor Tu GTP binding domain containing 2	2
Q15046	KARS	lysyl-tRNA synthetase	23
Q15056	EIF4H	eukaryotic translation initiation factor 4H	12
Q15075	EEA1	early endosome antigen 1	7
Q15084	PDIA6	protein disulfide isomerase family A, member 6	2

Q15102	PAFAH1B3	platelet-activating factor acetylhydrolase 1b, catalytic subunit 3 (29kDa)	8
Q15113	PCOLCE	procollagen C-endopeptidase enhancer	2
Q15149	PLEC	plectin	16
Q15181	PPA1	pyrophosphatase (inorganic) 1	7
Q15185	PTGES3	prostaglandin E synthase 3 (cytosolic)	6
Q15233	NONO	non-POU domain containing, octamer-binding	16
Q15257	PPP2R4	protein phosphatase 2A activator, regulatory subunit 4	5
Q15274	QPRT	quinolinate phosphoribosyltransferase	8
Q15286	RAB35	RAB35, member RAS oncogene family	1
Q15365	PCBP1	poly(rC) binding protein 1	7
Q15366	PCBP2	poly(rC) binding protein 2	3
Q15369	TCEB1	transcription elongation factor B (SIII), polypeptide 1 (15kDa, elongin C)	4
Q15393	SF3B3	splicing factor 3b, subunit 3, 130kDa	14
Q15404	RSU1	Ras suppressor protein 1	3
Q15417	CNN3	calponin 3, acidic	7
Q15424	SAFB	scaffold attachment factor B	4
Q15427	SF3B4	splicing factor 3b, subunit 4, 49kDa	2
Q15428	SF3A2	splicing factor 3a, subunit 2, 66kDa	4
Q15435	PPP1R7	protein phosphatase 1, regulatory subunit 7	6
Q15436	SEC23A	Sec23 homolog A (S. cerevisiae)	8
Q15459	SF3A1	splicing factor 3a, subunit 1, 120kDa	3
Q15631	TSN	translin	8
Q15637	SF1	splicing factor 1	2
Q15651	HMGN3	high mobility group nucleosomal binding domain 3	3
Q15691	MAPRE1	microtubule-associated protein, RP/EB family, member 1	7
Q15717	ELAVL1	ELAV like RNA binding protein 1	7
Q15819	UBE2V2	ubiquitin-conjugating enzyme E2 variant 2	3
Q15904	ATP6AP1	ATPase, H+ transporting, lysosomal accessory protein 1	2
Q15907	RAB11B	RAB11B, member RAS oncogene family	10
Q15942	ZYX	zyxin	3
Q16181	CDC10	septin 7	9
Q16186	ADRM1	adhesion regulating molecule 1	6
Q16222	UAP1	UDP-N-acetylglucosamine pyrophosphorylase 1	2
Q16270	IGFBP7	insulin-like growth factor binding protein 7	1
Q16401	PSMD5	proteasome (prosome, macropain) 26S subunit, non-ATPase, 5	5
Q16531	DDB1	damage-specific DNA binding protein 1, 127kDa	27
Q16543	CDC37	cell division cycle 37	8
Q16555	DPYSL2	dihydropyrimidinase-like 2	9

Q16576	RBBP7	retinoblastoma binding protein 7	1
Q16630	CPSF6	cleavage and polyadenylation specific factor 6, 68kDa	6
Q16658	FSCN1	fascin actin-bundling protein 1	7
Q16706	MAN2A1	mannosidase, alpha, class 2A, member 1	3
Q16819	MEP1A	meprin A, alpha (PABA peptide hydrolase)	12
Q16881	TXNRD1	thioredoxin reductase 1	10
Q2TAY7	SMU1	smu-1 suppressor of mec-8 and unc-52 homolog (C. elegans)	3
Q32MZ4	LRRFIP1	leucine rich repeat (in FLII) interacting protein 1	2
Q3KQS4	NOP2	NOP2 nucleolar protein	4
Q3LXA3	DAK	dihydroxyacetone kinase 2 homolog (S. cerevisiae)	8
Q3MHD2	LSM12	LSM12 homolog (S. cerevisiae)	2
Q3ZCW2	LGALSL	lectin, galactoside-binding-like	1
Q4G0F5	VPS26B	vacuolar protein sorting 26 homolog B (S. pombe)	1
Q4LE60	TNPO2	transportin 2	2
Q4V370	SECISBP2	SECIS binding protein 2	1
Q4VC31	CCDC58	coiled-coil domain containing 58	4
Q53GS9	USP39	ubiquitin specific peptidase 39	3
Q53H82	LACTB2	lactamase, beta 2	7
Q562R1	ACTBL2	actin, beta-like 2	1
Q58FF6	HSP90AB4P	heat shock protein 90kDa alpha (cytosolic), class B member 4, pseudogene	1
Q59F66	DDX17	DEAD (Asp-Glu-Ala-Asp) box helicase 17	10
Q5JTJ3	COA6	cytochrome c oxidase assembly factor 6	2
Q5LJA9	UCHL5	ubiquitin carboxyl-terminal hydrolase L5	3
Q5QJE6	DNTTIP2	deoxynucleotidyltransferase, terminal, interacting protein 2	1
Q5QPH3	ACSS2	acyl-CoA synthetase short-chain family member 2	3
Q5R3I4	TTC38	tetratricopeptide repeat domain 38	4
Q5SSJ5	HP1BP3	heterochromatin protein 1, binding protein 3	2
Q5T5C7	SARS	seryl-tRNA synthetase	5
Q5T6V5	C9orf64	chromosome 9 open reading frame 64	8
Q5T9B7	AK1	adenylate kinase 1	6
Q5TDH0	DDI2	DNA-damage inducible 1 homolog 2 (S. cerevisiae)	4
Q5TFE4	NT5DC1	5'-nucleotidase domain containing 1	3
Q5TZA2	CROCC	ciliary rootlet coiled-coil, rootletin	1
Q5VWC4	PSMD4	proteasome (prosome, macropain) 26S subunit, non-ATPase, 4	3
Q5VWZ2	LYPLAL1	lysophospholipase-like 1	1
Q5VYK3	KIAA0368	KIAA0368	4
Q5VZU9	TPP2	tripeptidyl peptidase II	9
Q6EMK4	VASN	vasorin	2

Q6IBS0	TWF2	twinfilin actin-binding protein 2	1
Q6IN85	SMEK1	SMEK homolog 1, suppressor of mek1 (Dictyostelium)	2
Q6P2Q9	PRPF8	pre-mRNA processing factor 8	6
Q6PJT7	ZC3H14	zinc finger CCCH-type containing 14	2
Q6PKG0	LARP1	La ribonucleoprotein domain family, member 1	2
Q6UN15	FIP1L1	factor interacting with PAPOLA and CPSF1	1
Q6UXH0	C19orf80	chromosome 19 open reading frame 80	2
Q6ZMR3	LDHAL6A	lactate dehydrogenase A-like 6A	1
Q6ZN17	LIN28B	lin-28 homolog B (C. elegans)	4
Q6ZRP7	QSOX2	quiescin Q6 sulfhydryl oxidase 2	2
Q71U36	TUBA1A	tubulin, alpha 1a	1
Q7KZF4	SND1	staphylococcal nuclease and tudor domain containing 1	31
Q7L1Q6	BZW1	basic leucine zipper and W2 domains 1	5
Q7L2H7	EIF3M	eukaryotic translation initiation factor 3, subunit M	5
Q7RTV0	PHF5A	PHD finger protein 5A	4
Q7Z4W1	DCXR	dicarbonyl/L-xylulose reductase	2
Q7Z6Z7	HUWE1	HECT, UBA and WWE domain containing 1, E3 ubiquitin protein ligase	11
Q7Z739	YTHDF3	YTH N(6)-methyladenosine RNA binding protein 3	4
Q86SQ4	GPR126	G protein-coupled receptor 126	2
Q86TE4	LUZP2	leucine zipper protein 2	2
Q86TG7	PEG10	paternally expressed 10	2
Q86TI2	DPP9	dipeptidyl-peptidase 9	5
Q86U42	PABPN1	poly(A) binding protein, nuclear 1	4
Q86UD1	OAF	OAF homolog (Drosophila)	2
Q86UK7	ZNF598	zinc finger protein 598	1
Q86UP2	KTN1	kinectin 1 (kinesin receptor)	13
Q86VP6	CAND1	cullin-associated and neddylation-dissociated 1	27
Q86WB0	ZC3HC1	zinc finger, C3HC-type containing 1	1
Q86X29	LSR	lipolysis stimulated lipoprotein receptor	2
Q86YS6	RAB43	RAB43, member RAS oncogene family	1
Q8IVM0	CCDC50	coiled-coil domain containing 50	1
Q8N163	CCAR2	cell cycle and apoptosis regulator 2	5
Q8N1G4	LRRC47	leucine rich repeat containing 47	11
Q8N3J3	C17orf53	chromosome 17 open reading frame 53	2
Q8N4C6	NIN	ninein (GSK3B interacting protein)	2
Q8N684	CPSF7	cleavage and polyadenylation specific factor 7, 59kDa	3
Q8N8S7	ENAH	enabled homolog (Drosophila)	3
Q8NB90	SPATA5	spermatogenesis associated 5	1

Q8NBJ4	GOLM1	golgi membrane protein 1	5
Q8NBP7	PCSK9	proprotein convertase subtilisin/kexin type 9	28
Q8NBS9	TXNDC5	thioredoxin domain containing 5 (endoplasmic reticulum)	6
Q8NC51	SERBP1	SERPINE1 mRNA binding protein 1	14
Q8NCW5	APOA1BP	apolipoprotein A-I binding protein	3
Q8NDC0	MAPK1IP1L	mitogen-activated protein kinase 1 interacting protein 1-like	1
Q8NEY8	PPHLN1	periphilin 1	2
Q8NFC6	BOD1L1	biorientation of chromosomes in cell division 1-like 1	2
Q8TAF3	WDR48	WD repeat domain 48	3
Q8TAT6	NPLOC4	nuclear protein localization 4 homolog (S. cerevisiae)	2
Q8TBC4	UBA3	ubiquitin-like modifier activating enzyme 3	2
Q8TD08	MAPK15	mitogen-activated protein kinase 15	1
Q8TEA8	DTD1	D-tyrosyl-tRNA deacylase 1	1
Q8TF05	PPP4R1	protein phosphatase 4, regulatory subunit 1	2
Q8WUD4	CCDC12	coiled-coil domain containing 12	1
Q8WUW1	BRK1	BRICK1, SCAR/WAVE actin-nucleating complex subunit	3
Q8WVJ2	NUDCD2	NudC domain containing 2	3
Q8WVM8	SCFD1	sec1 family domain containing 1	12
Q8WW12	PCNP	PEST proteolytic signal containing nuclear protein	10
Q8WXF1	PSPC1	paraspeckle component 1	9
Q8WZ42	TTN	titin	9
Q8WZA9	IRGQ	immunity-related GTPase family, Q	3
Q92499	DDX1	DEAD (Asp-Glu-Ala-Asp) box helicase 1	7
Q92520	FAM3C	family with sequence similarity 3, member C	6
Q92526	CCT6B	chaperonin containing TCP1, subunit 6B (zeta 2)	1
Q92597	NDRG1	N-myc downstream regulated 1	2
Q92598	HSPH1	heat shock 105kDa/110kDa protein 1	20
Q92688	ANP32B	acidic (leucine-rich) nuclear phosphoprotein 32 family, member B	8
Q92692	PVRL2	poliovirus receptor-related 2 (herpesvirus entry mediator B)	2
Q92734	TFG	TRK-fused gene	2
Q92747	ARPC1A	actin related protein 2/3 complex, subunit 1A, 41kDa	1
Q92769	HDAC2	histone deacetylase 2	3
Q92804	TAF15	TAF15 RNA polymerase II, TATA box binding protein (TBP)-associated factor, 68kDa	3
Q92820	GGH	gamma-glutamyl hydrolase (conjugase, folylpolygammaglutamyl hydrolase)	5
Q92824	PCSK5	proprotein convertase subtilisin/kexin type 5	2
Q92900	UPF1	UPF1 regulator of nonsense transcripts homolog (yeast)	5

Q92905	COPS5	COP9 signalosome subunit 5	2
Q92922	SMARCC1	SWI/SNF related, matrix associated, actin dependent regulator of chromatin, subfamily c, member 1	3
Q92930	RAB8B	RAB8B, member RAS oncogene family	1
Q92945	KHSRP	KH-type splicing regulatory protein	16
Q92973	TNPO1	transportin 1	1
Q92979	EMG1	EMG1 N1-specific pseudouridine methyltransferase	2
Q93009	USP7	ubiquitin specific peptidase 7 (herpes virus-associated)	5
Q93077	HIST1H2AC	histone cluster 1, H2ac	1
Q93099	HGD	homogentisate 1,2-dioxygenase	11
Q969E8	TSR2	TSR2, 20S rRNA accumulation, homolog (S. cerevisiae)	1
Q969H8	MYDGF	myeloid-derived growth factor	3
Q96A72	MAGOHB	mago-nashi homolog B (Drosophila)	1
Q96AG4	LRRC59	leucine rich repeat containing 59	3
Q96AT1	KIAA1143	KIAA1143	1
Q96AT9	RPE	ribulose-5-phosphate-3-epimerase	2
Q96B54	ZNF428	zinc finger protein 428	1
Q96BP3	PPWD1	peptidylprolyl isomerase domain and WD repeat containing 1	1
Q96C86	DCPS	decapping enzyme, scavenger	2
Q96CN7	ISOC1	isochorismatase domain containing 1	5
Q96CS3	FAF2	Fas associated factor family member 2	2
Q96CT7	CCDC124	coiled-coil domain containing 124	9
Q96EP5	DAZAP1	DAZ associated protein 1	1
Q96FJ2	DYNLL2	dynein, light chain, LC8-type 2	2
Q96FN9	DTD2	D-tyrosyl-tRNA deacylase 2 (putative)	3
Q96FQ6	S100A16	S100 calcium binding protein A16	1
Q96G03	PGM2	phosphoglucomutase 2	3
Q96GD0	PDXP	pyridoxal (pyridoxine, vitamin B6) phosphatase	2
Q96GG9	DCUN1D1	DCN1, defective in cullin neddylation 1, domain containing 1	4
Q96GX9	APIP	APAF1 interacting protein	1
Q96H79	ZC3HAV1L	zinc finger CCCH-type, antiviral 1-like	5
Q96HR9	REEP6	receptor accessory protein 6	2
Q96I15	SCLY	selenocysteine lyase	1
Q96IU4	ABHD14B*	abhydrolase domain containing 14B	1
Q96IY4	CPB2	carboxypeptidase B2 (plasma)	2
Q96K76	USP47	ubiquitin specific peptidase 47	1
Q96KP4	CNDP2	CNDP dipeptidase 2 (metallopeptidase M20 family)	8
Q96MW1	CCDC43	coiled-coil domain containing 43	1

Q96P16	RPRD1A	regulation of nuclear pre-mRNA domain containing 1A	2
Q96QK1	VPS35	vacuolar protein sorting 35 homolog (S. cerevisiae)	5
Q96QR8	PURB	purine-rich element binding protein B	2
Q96RS6	NUDCD1	NudC domain containing 1	1
Q96S82	UBL7	ubiquitin-like 7	1
Q96SI9	STRBP	spermatid perinuclear RNA binding protein	1
Q99426	TBCB	tubulin folding cofactor B	5
Q99436	PSMB7	proteasome (prosome, macropain) subunit, beta type, 7	4
Q99460	PSMD1	proteasome (prosome, macropain) 26S subunit, non-ATPase, 1	4
Q99471	PFDN5	prefoldin subunit 5	3
Q99497	PARK7	parkinson protein 7	12
Q99519	NEU1	sialidase 1 (lysosomal sialidase)	3
Q99536	VAT1	vesicle amine transport 1	4
Q99574	SERPINI1	serpin peptidase inhibitor, clade I (neuroserpin), member 1	4
Q99584	S100A13	S100 calcium binding protein A13	3
Q99598	TSNAX	translin-associated factor X	6
Q99615	DNAJC7	DnaJ (Hsp40) homolog, subfamily C, member 7	4
Q99729	HNRNPAB	heterogeneous nuclear ribonucleoprotein A/B	18
Q99832	CCT7	chaperonin containing TCP1, subunit 7 (eta)	17
Q99856	ARID3A	AT rich interactive domain 3A (BRIGHT-like)	7
Q99961	SH3GL1	SH3-domain GRB2-like 1	4
Q99969	RARRES2	retinoic acid receptor responder (tazarotene induced) 2	5
Q99988	GDF15	growth differentiation factor 15	2
Q9BQ52	ELAC2	elaC ribonuclease Z 2	3
Q9BQ61	C19orf43	chromosome 19 open reading frame 43	3
Q9BQT9	CLSTN3	calsyntenin 3	8
Q9BR76	CORO1B	coronin, actin binding protein, 1B	18
Q9BRA2	TXNDC17	thioredoxin domain containing 17	7
Q9BRF8	CPPED1	calcineurin-like phosphoesterase domain containing 1	1
Q9BRJ6	C7orf50	chromosome 7 open reading frame 50	1
Q9BRK5	SDF4	stromal cell derived factor 4	4
Q9BRP8	WIBG	within bgcn homolog (Drosophila)	2
Q9BS26	ERP44	endoplasmic reticulum protein 44	4
Q9BSE5	AGMAT	agmatine ureohydrolase (agmatinase)	2
Q9BT78	COPS4	COP9 signalosome subunit 4	5
Q9BTT0	ANP32E	acidic (leucine-rich) nuclear phosphoprotein 32 family, member E	3
Q9BTY2	FUCA2	fucosidase, alpha-L- 2, plasma	8
Q9BUQ8	DDX23	DEAD (Asp-Glu-Ala-Asp) box polypeptide 23	4

Q9BUT1	BDH2	3-hydroxybutyrate dehydrogenase, type 2	2
Q9BV20	MRI1	methylthioribose-1-phosphate isomerase 1	3
Q9BV44	THUMPD3	THUMP domain containing 3	1
Q9BVC6	TMEM109	transmembrane protein 109	4
Q9BWD1	ACAT2	acetyl-CoA acetyltransferase 2	9
Q9BWJ5	SF3B5	splicing factor 3b, subunit 5, 10kDa	3
Q9BX68	HINT2	histidine triad nucleotide binding protein 2	2
Q9BXJ9	NAA15	N(alpha)-acetyltransferase 15, NatA auxiliary subunit	10
Q9BXP5	SRRT	serrate, RNA effector molecule	2
Q9BXS5	AP1M1	adaptor-related protein complex 1, mu 1 subunit	3
Q9BXW9	FANCD2	Fanconi anemia, complementation group D2	2
Q9BY32	ITPA	inosine triphosphatase (nucleoside triphosphate pyrophosphatase)	3
Q9BY44	EIF2A	eukaryotic translation initiation factor 2A, 65kDa	6
Q9BYT8	NLN	neurolysin (metallopeptidase M3 family)	3
Q9BZL1	UBL5	ubiquitin-like 5	3
Q9BZZ5	API5	apoptosis inhibitor 5	8
Q9C005	DPY30	dpy-30 homolog (C. elegans)	2
Q9GZT8	NIF3L1	NIF3 NGG1 interacting factor 3-like 1 (S. cerevisiae)	3
Q9GZZ1	NAA50	N(alpha)-acetyltransferase 50, NatE catalytic subunit	4
Q9H074	PAIP1	poly(A) binding protein interacting protein 1	4
Q9H098	FAM107B	family with sequence similarity 107, member B	2
Q9H0D6	XRN2	5'-3' exoribonuclease 2	2
Q9H0U4	RAB1B	RAB1B, member RAS oncogene family	1
Q9H0W9	C11orf54	chromosome 11 open reading frame 54	4
Q9H1E3	NUCKS1	nuclear casein kinase and cyclin-dependent kinase substrate 1	8
Q9H299	SH3BGRL3	SH3 domain binding glutamate-rich protein like 3	4
Q9H2C0	GAN	gigaxonin	1
Q9H2G2	SLK	STE20-like kinase	4
Q9H2U2	PPA2	pyrophosphatase (inorganic) 2	1
Q9H361	PABPC3	poly(A) binding protein, cytoplasmic 3	1
Q9H3G5	CPVL	carboxypeptidase, vitellogenic-like	9
Q9H3K6	BOLA2/BOLA2B	bolA family member 2	5
Q9H444	CHMP4B	charged multivesicular body protein 4B	3
Q9H446	RWDD1	RWD domain containing 1	2
Q9H479	FN3K	fructosamine 3 kinase	1
Q9H4A4	RNPEP	arginyl aminopeptidase (aminopeptidase B)	5
Q9H4A6	GOLPH3	golgi phosphoprotein 3 (coat-protein)	2
Q9H6F5	CCDC86	coiled-coil domain containing 86	1

Q9H6Z4	RANBP3	RAN binding protein 3	4
Q9H773	DCTPP1	dCTP pyrophosphatase 1	4
Q9H993	C6orf211	chromosome 6 open reading frame 211	3
Q9HA64	FN3KRP	fructosamine 3 kinase related protein	2
Q9HB07	C12orf10	chromosome 12 open reading frame 10	5
Q9HB71	CACYBP	calcyclin binding protein	14
Q9HBR0	SLC38A10	solute carrier family 38, member 10	1
Q9HD42	CHMP1A	charged multivesicular body protein 1A	3
Q9NP61	ARFGAP3	ADP-ribosylation factor GTPase activating protein 3	2
Q9NP77	SSU72	SSU72 RNA polymerase II CTD phosphatase homolog (S. cerevisiae)	2
Q9NP79	VTA1	vesicle (multivesicular body) trafficking 1	3
Q9NQ29	LUC7L	LUC7-like (S. cerevisiae)	1
Q9NQ94	A1CF	APOBEC1 complementation factor	2
Q9NQG5	RPRD1B	regulation of nuclear pre-mRNA domain containing 1B	3
Q9NQI0	DDX4	DEAD (Asp-Glu-Ala-Asp) box polypeptide 4	1
Q9NQR4	NIT2	nitrilase family, member 2	3
Q9NR28	DIABLO	diablo, IAP-binding mitochondrial protein	1
Q9NR30	DDX21	DEAD (Asp-Glu-Ala-Asp) box helicase 21	2
Q9NR45	NANS	N-acetylneuraminic acid synthase	8
Q9NR50	EIF2B3	eukaryotic translation initiation factor 2B, subunit 3 gamma, 58kDa	3
Q9NRF8	CTPS2	CTP synthase 2	1
Q9NRN5	OLFML3	olfactomedin-like 3	3
Q9NRN7	AASDHPPT	aminoadipate-semialdehyde dehydrogenase-phosphopantetheinyl transferase	2
Q9NRV9	HEBP1	heme binding protein 1	3
Q9NRX4	PHPT1	phosphohistidine phosphatase 1	5
Q9NRY2	INIP	INTS3 and NABP interacting protein	1
Q9NS86	LANCL2	LanC lantibiotic synthetase component C-like 2 (bacterial)	1
Q9NSD9	FARSB	phenylalanyl-tRNA synthetase, beta subunit	9
Q9NT62	ATG3	autophagy related 3	2
Q9NTJ3	SMC4	structural maintenance of chromosomes 4	3
Q9NTK5	OLA1	Obg-like ATPase 1	10
Q9NUQ9	FAM49B	family with sequence similarity 49, member B	4
Q9NV58	RNF19A	ring finger protein 19A, RBR E3 ubiquitin protein ligase	2
Q9NVM6	DNAJC17	DnaJ (Hsp40) homolog, subfamily C, member 17	2
Q9NW68	BSDC1	BSD domain containing 1	2
Q9NWV4	C1orf123	chromosome 1 open reading frame 123	1
Q9NX58	LYAR	Ly1 antibody reactive	3

Q9NXG2	THUMPD1	THUMP domain containing 1	3
Q9NXH9	TRMT1	tRNA methyltransferase 1 homolog (S. cerevisiae)	2
Q9NY27	PPP4R2	protein phosphatase 4, regulatory subunit 2	2
Q9NYJ1	COA4	cytochrome c oxidase assembly factor 4 homolog (S. cerevisiae)	2
Q9NZ08	ERAP1	endoplasmic reticulum aminopeptidase 1	3
Q9NZE6	EIF4A2*	eukaryotic translation initiation factor 4A2	1
Q9NZI8	IGF2BP1	insulin-like growth factor 2 mRNA binding protein 1	10
Q9NZL4	HSPBP1	HSPA (heat shock 70kDa) binding protein, cytoplasmic cochaperone 1	5
Q9NZL9	MAT2B	methionine adenosyltransferase II, beta	3
Q9P016	THYN1	thymocyte nuclear protein 1	4
Q9P258	RCC2	regulator of chromosome condensation 2	15
Q9P2E9	RRBP1	ribosome binding protein 1	51
Q9P2J5	LARS	leucyl-tRNA synthetase	5
Q9P2P6	STARD9	StAR-related lipid transfer (START) domain containing 9	4
Q9P2R3	ANKFY1	ankyrin repeat and FYVE domain containing 1	2
Q9UBB4	ATXN10	ataxin 10	1
Q9UBC2	EPS15L1	epidermal growth factor receptor pathway substrate 15-like 1	2
Q9UBE0	SAE1	SUMO1 activating enzyme subunit 1	8
Q9UBQ5	EIF3K	eukaryotic translation initiation factor 3, subunit K	3
Q9UBQ7	GRHPR	glyoxylate reductase/hydroxypyruvate reductase	7
Q9UBR2	CTSZ	cathepsin Z	4
Q9UBT2	UBA2	ubiquitin-like modifier activating enzyme 2	14
Q9UEE9	CFDP1	craniofacial development protein 1	3
Q9UEY8	ADD3	adducin 3 (gamma)	2
Q9UGI8	TES	testis derived transcript (3 LIM domains)	10
Q9UH65	SWAP70	SWAP switching B-cell complex 70kDa subunit	2
Q9UHD1	CHORDC1	cysteine and histidine-rich domain (CHORD) containing 1	5
Q9UHD8	KIAA0991	septin 9	10
Q9UHD9	UBQLN2	ubiquilin 2	2
Q9UHV9	PFDN2	prefoldin subunit 2	5
Q9UHY7	ENOPH1	enolase-phosphatase 1	1
Q9UI30	TRMT112	tRNA methyltransferase 11-2 homolog (S. cerevisiae)	1
Q9UII2	ATPIF1	ATPase inhibitory factor 1	3
Q9UJU6	DBNL*	drebrin-like	1
Q9UJU6	DBNL*	drebrin-like	1
Q9UK22	FBXO2	F-box protein 2	2
Q9UK45	LSM7	LSM7 homolog, U6 small nuclear RNA associated (S. cerevisiae)	4
Q9UK76	HN1	hematological and neurological expressed 1	3

Q9UKD2	MRTO4	mRNA turnover 4 homolog (S. cerevisiae)	1
Q9UKM9	RALY	RALY heterogeneous nuclear ribonucleoprotein	3
Q9UKY7	CDV3	CDV3 homolog (mouse)	6
Q9UKZ9	PCOLCE2	procollagen C-endopeptidase enhancer 2	2
Q9UL25	RAB21	RAB21, member RAS oncogene family	1
Q9UL46	PSME2	proteasome (prosome, macropain) activator subunit 2 (PA28 beta)	5
Q9ULT8	HECTD1	HECT domain containing E3 ubiquitin protein ligase 1	2
Q9UMS4	PRPF19	pre-mRNA processing factor 19	6
Q9UMY4	SNX12	sorting nexin 12	3
Q9UN86	G3BP2	GTPase activating protein (SH3 domain) binding protein 2	4
Q9UNF0	PACSIN2	protein kinase C and casein kinase substrate in neurons 2	3
Q9UNF1	MAGED2	melanoma antigen family D2	3
Q9UNM6	PSMD13	proteasome (prosome, macropain) 26S subunit, non-ATPase, 13	7
Q9UNN5	FAF1	Fas (TNFRSF6) associated factor 1	2
Q9UNS2	COPS3	COP9 signalosome subunit 3	3
Q9UNX3	RPL26L1	ribosomal protein L26-like 1	1
Q9UNZ2	NSFL1C	NSFL1 (p97) cofactor (p47)	15
Q9UPT8	ZC3H4	zinc finger CCCH-type containing 4	1
Q9UQ35	SRRM2	serine/arginine repetitive matrix 2	4
Q9UQ80	PA2G4*	proliferation-associated 2G4, 38kDa	27
Q9UQE7	SMC3	structural maintenance of chromosomes 3	2
Q9Y224	C14orf166	chromosome 14 open reading frame 166	4
Q9Y230	RUVBL2	RuvB-like AAA ATPase 2	14
Q9Y237	PIN4	protein (peptidylprolyl cis/trans isomerase) NIMA-interacting, 4 (parvulin)	4
Q9Y265	RUVBL1	RuvB-like AAA ATPase 1	9
Q9Y266	NUDC	nudC nuclear distribution protein	11
Q9Y281	CFL2	cofilin 2 (muscle)	5
Q9Y295	DRG1	developmentally regulated GTP binding protein 1	2
Q9Y2K7	KDM2A	lysine (K)-specific demethylase 2A	1
Q9Y2S6	TMA7	translation machinery associated 7 homolog (S. cerevisiae)	6
Q9Y2V2	CARHSP1	calcium regulated heat stable protein 1, 24kDa	6
Q9Y2W2	WBP11	WW domain binding protein 11	5
Q9Y2X3	NOP58	NOP58 ribonucleoprotein	1
Q9Y2Z0	SUGT1	SGT1, suppressor of G2 allele of SKP1 (S. cerevisiae)	5
Q9Y333	LSM2	LSM2 homolog, U6 small nuclear RNA associated (S. cerevisiae)	4
Q9Y383	LUC7L2	LUC7-like 2 (S. cerevisiae)	2
Q9Y3A5	SBDS	Shwachman-Bodian-Diamond syndrome	12
Q9Y3B4	SF3B6	splicing factor 3b, subunit 6, 14kDa	3

Q9Y3B9	RRP15	ribosomal RNA processing 15 homolog (S. cerevisiae)	1
Q9Y3C6	PPIL1	peptidylprolyl isomerase (cyclophilin)-like 1	4
Q9Y3C8	UFC1	ubiquitin-fold modifier conjugating enzyme 1	4
Q9Y3I0	RTCB	RNA 2',3'-cyclic phosphate and 5'-OH ligase	8
Q9Y3P9	RABGAP1	RAB GTPase activating protein 1	2
Q9Y3U8	RPL36	ribosomal protein L36	1
Q9Y490	TLN1	talin 1	51
Q9Y4B5	MTCL1	microtubule crosslinking factor 1	2
Q9Y4E8	USP15	ubiquitin specific peptidase 15	2
Q9Y4X5	ARIH1	ariadne RBR E3 ubiquitin protein ligase 1	1
Q9Y4Z0	LSM4	LSM4 homolog, U6 small nuclear RNA associated (S. cerevisiae)	2
Q9Y5B9	SUPT16H	suppressor of Ty 16 homolog (S. cerevisiae)	12
Q9Y5C1	ANGPTL3	angiopoietin-like 3	8
Q9Y5K6	CD2AP	CD2-associated protein	3
Q9Y5S9	RBM8A	RNA binding motif protein 8A	4
Q9Y5X1	SNX9	sorting nexin 9	1
Q9Y5X3	SNX5	sorting nexin 5	7
Q9Y617	PSAT1	phosphoserine aminotransferase 1	22
Q9Y678	COPG1	coatomer protein complex, subunit gamma 1	13
Q9Y696	CLIC4	chloride intracellular channel 4	3
Q9Y6E2	BZW2	basic leucine zipper and W2 domains 2	6
Q9Y6G9	DYNC1LI1	dynein, cytoplasmic 1, light intermediate chain 1	2
Q9Y6W5	WASF2	WAS protein family, member 2	2
U6CYF3	E9PBA7	Methionine adenosyltransferase II	1

Appendix 3. List of proteins quantified by MS/MS SILAC 'spike in' in the analyses of secretomes from HUH7 cells overexpressing PCSK9 and its variants

Gene Name	Protein Name	ANOVA Significant	Cluster	Unique Peptides
EIF4H	eukaryotic translation initiation factor 4H	+	A	12
ERP29	endoplasmic reticulum protein 29	+	A	6
GART	phosphoribosylglycinamide formyltransferase	+	A	7
HSPA4	heat shock 70kDa protein 4	+	A	31
LARS	leucyl-tRNA synthetase	+	A	5
PGP	phosphoglycolate phosphatase	+	A	7
RAD23A	RAD23 homolog A	+	A	1
XRCC5	70 kDa subunit of Ku antigen	+	A	15
ARPC4	actin related protein 2/3 complex, subunit 4, 20kDa	+	B	7
BOLA2/BOLA2B	bolA family member 2	+	B	5
CCT3	chaperonin containing TCP1, subunit 3 (gamma)	+	B	29
CCT5	chaperonin containing TCP1, subunit 5 (epsilon)	+	B	2
DHX9	DEAH (Asp-Glu-Ala-His) box helicase 9	+	B	19
HNRNPM	heterogeneous nuclear ribonucleoprotein M	+	B	23
ILF3	interleukin enhancer binding factor 3, 90kDa	+	B	16
KARS	lysyl-tRNA synthetase	+	B	23
LAP3	leucine aminopeptidase 3	+	B	16
PLIN3	perilipin 3	+	B	6
PSMB2	proteasome subunit, beta type, 2	+	B	9
RPL19	ribosomal protein L19	+	B	8
RPL5	ribosomal protein L5	+	B	23
SUPT16H	suppressor of Ty 16 homolog	+	B	12
TCP1	t-complex 1	+	B	20
VCP	valosin containing protein	+	B	49
VPS26A	vacuolar protein sorting 26 homolog A	+	B	1
VSNL1	visinin-like 1	+	B	2
ACTN3	actinin, alpha 3	+	C	1
HNRNPH1	heterogeneous nuclear ribonucleoprotein H1 (H)	+	C	3
HSPB1	heat shock 27kDa protein 1	+	C	6
PCSK9	proprotein convertase subtilisin/kexin type 9	+	C	28
PSMD3	proteasome 26S subunit, non-ATPase, 3	+	C	8
ISOC1	isochorismatase domain containing 1	+	D	5

LUC7L2	LUC7-like 2	+	D	2
ADM	adrenomedullin	+	E	1
AFP	alpha-fetoprotein	+	E	0
AGT	Angiotensin I	+	E	10
AMBP	alpha-1-microglobulin/bikunin precursor	+	E	21
ANG	angiogenin, ribonuclease, RNase A family, 5	+	E	4
ANGPTL3	angiopoietin-like 3	+	E	8
ANXA2	annexin A2	+	E	10
ANXA5	annexin A5	+	E	5
APOA1	apolipoprotein A-I	+	E	6
APOA2	apolipoprotein A-II	+	E	7
APOB	apolipoprotein B	+	E	0
APOC2	apolipoprotein C-II	+	E	5
APOC3	apolipoprotein C-III	+	E	3
APOE	apolipoprotein E	+	E	0
APOH	apolipoprotein H (beta-2-glycoprotein I)	+	E	13
APOM	apolipoprotein M	+	E	6
APP	amyloid beta (A4) precursor protein	+	E	8
B2M	beta-2-microglobulin	+	E	5
B3GNT1	I-beta-1,3-N-acetylglucosaminyltransferase	+	E	15
BNC2	basonuclin 2	+	E	2
C3	complement component 3	+	E	90
C5	complement component 5	+	E	35
CDH2	cadherin 2, type 1, N-cadherin (neuronal)	+	E	6
CFH	complement factor H	+	E	31
CLSTN1	calsyntenin 1	+	E	16
CLSTN3	calsyntenin 3	+	E	8
CLU	clusterin	+	E	31
COCH	cochlin	+	E	18
COL2A1	collagen, type II, alpha 1	+	E	22
COL5A2	collagen, type V, alpha 2	+	E	18
CPVL	carboxypeptidase, vitellogenic-like	+	E	9
CST3	cystatin C	+	E	8
CTSC	cathepsin C	+	E	6
CTSD	cathepsin D	+	E	7
CTSL	cathepsin L	+	E	1
CXCL5	chemokine (C-X-C motif) ligand 5	+	E	3

EFEMP1	EGF containing fibulin-like extracellular matrix protein 1	+	E	11
F10	coagulation factor X	+	E	6
F12	coagulation factor XII (Hageman factor)	+	E	7
F2	coagulation factor II (thrombin)	+	E	12
FAM3C	family with sequence similarity 3, member C	+	E	6
FBLN1*	fibulin 1	+	E	4
FBLN1*	fibulin 1	+	E	4
FN1	fibronectin 1	+	E	62
FUCA2	fucosidase, alpha-L- 2, plasma	+	E	8
GALNT2	polypeptide N-acetylgalactosaminyltransferase 2	+	E	3
GANAB	glucosidase, alpha; neutral AB	+	E	17
GC	group-specific component	+	E	25
GLA	galactosidase, alpha	+	E	5
GOLM1	golgi membrane protein 1	+	E	5
HP	haptoglobin	+	E	8
HSPA5	78 kDa glucose-regulated protein	+	E	29
HSPE1	heat shock 10kDa protein 1	+	E	14
IGF2	insulin-like growth factor 2	+	E	10
IGFBP7	insulin-like growth factor binding protein 7	+	E	1
ITIH2	inter-alpha-trypsin inhibitor heavy chain 2	+	E	31
KIF1C	kinesin family member 1C	+	E	1
KNG1	kininogen 1	+	E	24
LAMB1	laminin, beta 1	+	E	19
LAMC1	laminin, gamma 1 (formerly LAMB2)	+	E	10
LYZ	lysozyme	+	E	8
MAN1A1	mannosidase, alpha, class 1A, member 1	+	E	20
MATN3	matrilin 3	+	E	3
MEP1A	meprin A, alpha (PABA peptide hydrolase)	+	E	12
MYH9	myosin, heavy chain 9, non-muscle	+	E	7
NEU1	sialidase 1 (lysosomal sialidase)	+	E	3
NID1	nidogen 1	+	E	37
NPC2	Niemann-Pick disease, type C2	+	E	3
NUCB1	nucleobindin 1	+	E	21
OLFML3	olfactomedin-like 3	+	E	3
ORM1	orosomucoid 1	+	E	4
PDIA3*	protein disulfide isomerase family A, member 3	+	E	10
PLA2G7	phospholipase A2, group VII	+	E	2

PLG	plasminogen	+	E	10
PLOD3	procollagen-lysine, 2-oxoglutarate 5-dioxygenase 3	+	E	10
PROS1	protein S (alpha)	+	E	12
PTPRF	protein tyrosine phosphatase, receptor type, F	+	E	10
RAB1A	RAB1A, member RAS oncogene family	+	E	1
RBP4	retinol binding protein 4, plasma	+	E	14
RNASE4	ribonuclease, RNase A family, 4	+	E	9
RRBP1	ribosome binding protein 1	+	E	51
SDC4	syndecan 4	+	E	2
SDF4	stromal cell derived factor 4	+	E	4
SERPINA1	serpin peptidase inhibitor, clade A, member 1	+	E	29
SERPINA4	serpin peptidase inhibitor, clade A, member 4	+	E	4
SERPINA5	serpin peptidase inhibitor, clade A, member 5	+	E	15
SERPINA6	serpin peptidase inhibitor, clade A, member 6	+	E	5
SERPIND1	serpin peptidase inhibitor, clade D, member 1	+	E	12
SERPINF1	serpin peptidase inhibitor, clade F, member 1	+	E	15
SERPINF2	serpin peptidase inhibitor, clade F, member 2	+	E	12
SERPINI1	serpin peptidase inhibitor, clade I, member 1	+	E	4
SPINK1	serine peptidase inhibitor, Kazal type 1	+	E	7
STC2	stanniocalcin 2	+	E	5
TF	transferrin	+	E	0
THBS1	thrombospondin 1	+	E	2
VTN	vitronectin	+	E	16
AHSG	Alpha-2-HS-glycoprotein	+	E	12
BF	cDNA FLJ55673, highly similar to Complement factor B	+	E	8
A2M	Alpha-2-macroglobulin	+	E	48
BIGH3	Kerato-epithelin	+	E	14
42249	septin 2	NS	N/A	7
42254	septin 7	NS	N/A	9
42256	septin 9	NS	N/A	10
42257	septin 10	NS	N/A	2
42258	septin 11	NS	N/A	8
A1BG	alpha-1-B glycoprotein	NS	N/A	9
AAMDC	adipogenesis associated, Mth938 domain containing	NS	N/A	4
AARS	alanyl-tRNA synthetase	NS	N/A	38
ABCE1	ATP-binding cassette, sub-family E (OABP), member 1	NS	N/A	2

ABHD14B	abhydrolase domain containing 14B	NS	N/A	1
ACAT2	acetyl-CoA acetyltransferase 2	NS	N/A	9
ACLY	ATP citrate lyase	NS	N/A	35
ACO1	aconitase 1, soluble	NS	N/A	5
ACP1	acid phosphatase 1, soluble	NS	N/A	4
ACTBL2	actin, beta-like 2	NS	N/A	1
ACTC1	actin, alpha, cardiac muscle 1	NS	N/A	1
ACTN1	actinin, alpha 1	NS	N/A	16
ACTN4	actinin, alpha 4	NS	N/A	41
ACTR1B	Actin-related protein 1B	NS	N/A	3
ACTR2	ARP2 actin-related protein 2 homolog (yeast)	NS	N/A	8
ACTR3	ARP3 actin-related protein 3 homolog (yeast)	NS	N/A	7
ADH5	alcohol dehydrogenase 5 (class III), chi polypeptide	NS	N/A	17
ADRM1	adhesion regulating molecule 1	NS	N/A	6
ADSL	adenylosuccinate lyase	NS	N/A	8
ADSS	adenylosuccinate synthase	NS	N/A	11
AGMAT	agmatine ureohydrolase (agmatinase)	NS	N/A	2
AGRN	agrin	NS	N/A	9
AHCY	adenosylhomocysteinase	NS	N/A	29
AHNAK	AHNAK nucleoprotein	NS	N/A	9
AHSA1	Activator of 90 kDa heat shock protein ATPase homolog 1	NS	N/A	8
AIFM1	apoptosis-inducing factor, mitochondrion-associated, 1	NS	N/A	6
AIMP1	Endothelial monocyte-activating polypeptide 2	NS	N/A	7
AIMP2	Multisynthase complex auxiliary component p38	NS	N/A	2
AIP	aryl hydrocarbon receptor interacting protein	NS	N/A	11
AK1	adenylate kinase 1	NS	N/A	6
AK2	adenylate kinase 2	NS	N/A	8
AKR1A1	Alcohol dehydrogenase [NADP+]	NS	N/A	9
AKR1B10	Aldo-keto reductase family 1 member B10	NS	N/A	5
AKR1C1/AKR1C2*	aldo-keto reductase family 1, member C2	NS	N/A	5
AKR1C1/AKR1C2*	aldo-keto reductase family 1, member C2	NS	N/A	5
AKR7A2	AFB1 aldehyde reductase 1	NS	N/A	4
ALCAM	activated leukocyte cell adhesion molecule	NS	N/A	4
ALDH1A1	aldehyde dehydrogenase 1 family, member A1	NS	N/A	34
ALDH7A1	aldehyde dehydrogenase 7 family, member A1	NS	N/A	3
ALDH9A1	aldehyde dehydrogenase 9 family, member A1	NS	N/A	2

ALDOA	aldolase A, fructose-bisphosphate	NS	N/A	29
ALDOC	aldolase C, fructose-bisphosphate	NS	N/A	9
ALYREF	Aly/REF export factor	NS	N/A	7
ANP32A	Acidic nuclear phosphoprotein pp32	NS	N/A	7
ANP32B	Acidic protein rich in leucines	NS	N/A	8
ANP32E	LANP-like protein	NS	N/A	3
AP1B1	adaptor-related protein complex 1, beta 1 subunit	NS	N/A	3
AP1G1	adaptor-related protein complex 1, gamma 1 subunit	NS	N/A	6
AP1S1	adaptor-related protein complex 1, sigma 1 subunit	NS	N/A	1
APEH	acylaminoacyl-peptide hydrolase	NS	N/A	9
APEX1	APEX nuclease (multifunctional DNA repair enzyme) 1	NS	N/A	11
API5	apoptosis inhibitor 5	NS	N/A	8
APLP2	amyloid beta (A4) precursor-like protein 2	NS	N/A	4
APOA1BP	apolipoprotein A-I binding protein	NS	N/A	3
APRT	adenine phosphoribosyltransferase	NS	N/A	4
ARCN1	archain 1	NS	N/A	13
ARF3	ADP-ribosylation factor 3	NS	N/A	3
ARF5	ADP-ribosylation factor 5	NS	N/A	1
ARG1	arginase 1	NS	N/A	4
ARHGAP5	Rho GTPase activating protein 5	NS	N/A	8
ARHGDIA	Rho GDP dissociation inhibitor (GDI) alpha	NS	N/A	12
ARID3A	AT rich interactive domain 3A (BRIGHT-like)	NS	N/A	7
ARPC1A	actin related protein 2/3 complex, subunit 1A, 41kDa	NS	N/A	1
ARPC1B	actin related protein 2/3 complex, subunit 1B, 41kDa	NS	N/A	4
ARPC2	actin related protein 2/3 complex, subunit 2, 34kDa	NS	N/A	8
ARPC3	actin related protein 2/3 complex, subunit 3, 21kDa	NS	N/A	4
ARPC5	actin related protein 2/3 complex, subunit 5, 16kDa	NS	N/A	2
ARPP19	cAMP-regulated phosphoprotein, 19kDa	NS	N/A	2
ASL	argininosuccinate lyase	NS	N/A	5
ASNS	asparagine synthetase (glutamine-hydrolyzing)	NS	N/A	11
ATIC	AICAR transformylase	NS	N/A	21
ATOX1	antioxidant 1 copper chaperone	NS	N/A	4
ATP6AP2	ATPase, H+ transporting, lysosomal accessory protein 2	NS	N/A	2
ATP6V1A	ATPase, H+ transporting, lysosomal 70kDa, V1 subunit A	NS	N/A	6
ATP6V1C1	ATPase, H+ transporting, lysosomal 42kDa, V1 subunit C1	NS	N/A	3

ATP6V1E1	ATPase, H+ transporting, lysosomal 31kDa, V1 subunit E1	NS	N/A	2
ATP6V1G1	ATPase, H+ transporting, lysosomal 13kDa, V1 subunit G1	NS	N/A	3
ATPIF1	ATPase inhibitory factor 1	NS	N/A	3
BAX	BCL2-associated X protein	NS	N/A	4
BCAM	basal cell adhesion molecule (Lutheran blood group)	NS	N/A	18
BCAT1	branched chain amino-acid transaminase 1, cytosolic	NS	N/A	3
BDH2	3-hydroxybutyrate dehydrogenase, type 2	NS	N/A	2
BLMH	bleomycin hydrolase	NS	N/A	11
BLVRB	biliverdin reductase B (flavin reductase (NADPH))	NS	N/A	4
BPNT1	3'(2'), 5'-bisphosphate nucleotidase 1	NS	N/A	10
BTD	biotinidase	NS	N/A	2
BUB3	BUB3 mitotic checkpoint protein	NS	N/A	5
BZW1	basic leucine zipper and W2 domains 1	NS	N/A	5
BZW2	basic leucine zipper and W2 domains 2	NS	N/A	6
C11orf54	chromosome 11 open reading frame 54	NS	N/A	4
C12orf10	chromosome 12 open reading frame 10	NS	N/A	5
C14orf166	chromosome 14 open reading frame 166	NS	N/A	4
C19orf10	chromosome 19 open reading frame 10	NS	N/A	3
C19orf43	chromosome 19 open reading frame 43	NS	N/A	3
C1S	complement component 1, s subcomponent	NS	N/A	3
C9orf64	chromosome 9 open reading frame 64	NS	N/A	8
CA2	carbonic anhydrase II	NS	N/A	6
CACYBP	calcyclin binding protein	NS	N/A	14
CAD	Aspartate carbamoyltransferase	NS	N/A	11
CALD1	caldesmon 1	NS	N/A	11
CALM1	calmodulin 1 (phosphorylase kinase, delta)	NS	N/A	#N/A
CALU	calumenin	NS	N/A	5
CAND1	cullin-associated and neddylation-dissociated 1	NS	N/A	27
CAP1	CAP, adenylate cyclase-associated protein 1 (yeast)	NS	N/A	22
CAP2	CAP, adenylate cyclase-associated protein, 2 (yeast)	NS	N/A	2
CAPG	capping protein (actin filament), gelsolin-like	NS	N/A	7
CAPN1	calpain 1, (mu/I) large subunit	NS	N/A	2
CAPRIN1	cell cycle associated protein 1	NS	N/A	4
CAPZA1	capping protein (actin filament) muscle Z-line, alpha 1	NS	N/A	5
CARHSP1	calcium regulated heat stable protein 1, 24kDa	NS	N/A	6

CARS	cysteinyl-tRNA synthetase	NS	N/A	4
CAST	calpastatin	NS	N/A	9
CBR1	carbonyl reductase 1	NS	N/A	8
CBS/LOC102724560	cystathionine-beta-synthase	NS	N/A	#N/A
CBX1	chromobox homolog 1	NS	N/A	3
CBX3	chromobox homolog 3	NS	N/A	8
CBX5	chromobox homolog 5	NS	N/A	9
CCAR2	cell cycle and apoptosis regulator 2	NS	N/A	5
CCDC124	coiled-coil domain containing 124	NS	N/A	9
CCDC50	coiled-coil domain containing 50	NS	N/A	1
CCDC58	coiled-coil domain containing 58	NS	N/A	4
CCS	copper chaperone for superoxide dismutase	NS	N/A	7
CCT2	chaperonin containing TCP1, subunit 2 (beta)	NS	N/A	22
CCT4	chaperonin containing TCP1, subunit 4 (delta)	NS	N/A	21
CCT6A	chaperonin containing TCP1, subunit 6A (zeta 1)	NS	N/A	16
CCT7	chaperonin containing TCP1, subunit 7 (eta)	NS	N/A	17
CCT8	chaperonin containing TCP1, subunit 8 (theta)	NS	N/A	27
CD99	CD99 molecule	NS	N/A	1
CDC37	cell division cycle 37	NS	N/A	8
CDC42	cell division cycle 42	NS	N/A	3
CDK1	cyclin-dependent kinase 1	NS	N/A	7
CDV3	CDV3 homolog (mouse)	NS	N/A	6
CFDP1	craniofacial development protein 1	NS	N/A	3
CFI	complement factor I	NS	N/A	16
CFL1	cofilin 1 (non-muscle)	NS	N/A	14
CFL2	cofilin 2 (muscle)	NS	N/A	5
CHKA	choline kinase alpha	NS	N/A	2
CHMP1A	charged multivesicular body protein 1A	NS	N/A	3
CHMP4B	charged multivesicular body protein 4B	NS	N/A	3
CHORDC1	CHORD domain-containing protein 1	NS	N/A	5
CKAP5	cytoskeleton associated protein 5	NS	N/A	4
CKB	creatine kinase, brain	NS	N/A	13
CLIC1	chloride intracellular channel 1	NS	N/A	13
CLIC4	chloride intracellular channel 4	NS	N/A	3
CLIP1	CAP-GLY domain containing linker protein 1	NS	N/A	2
CLTA	clathrin, light chain A	NS	N/A	7
CLTC	clathrin, heavy chain (Hc)	NS	N/A	39

CMPK1	Cytidine monophosphate kinase	NS	N/A	9
CNBP	CCHC-type zinc finger, nucleic acid binding protein	NS	N/A	11
CNDP2	CNDP dipeptidase 2 (metallopeptidase M20 family)	NS	N/A	8
CNN3	calponin 3, acidic	NS	N/A	7
COA4	cytochrome c oxidase assembly factor 4 homolog	NS	N/A	4
COL1A1	collagen, type I, alpha 1	NS	N/A	3
COPA	coatomer protein complex, subunit alpha	NS	N/A	26
COPB1	coatomer protein complex, subunit beta 1	NS	N/A	6
COPB2	coatomer protein complex, subunit beta 2 (beta prime)	NS	N/A	9
COPE	coatomer protein complex, subunit epsilon	NS	N/A	8
COPG1	coatomer protein complex, subunit gamma 1	NS	N/A	13
COPS4	COP9 signalosome subunit 4	NS	N/A	5
COPS5	COP9 signalosome subunit 5	NS	N/A	2
CORO1B	coronin, actin binding protein, 1B	NS	N/A	18
CORO1C	coronin, actin binding protein, 1C	NS	N/A	17
CP	ceruloplasmin (ferroxidase)	NS	N/A	5
CPB2	carboxypeptidase B2 (plasma)	NS	N/A	2
CPE	carboxypeptidase E	NS	N/A	3
CPN1	carboxypeptidase N, polypeptide 1	NS	N/A	3
CPOX	coproporphyrinogen oxidase	NS	N/A	2
CPPED1	calcineurin-like phosphoesterase domain containing 1	NS	N/A	1
CRYZ	crystallin, zeta (quinone reductase)	NS	N/A	12
CS	citrate synthase	NS	N/A	3
CSE1L	CSE1 chromosome segregation 1-like (yeast)	NS	N/A	17
CTBP1	C-terminal binding protein 1	NS	N/A	3
CTGF	connective tissue growth factor	NS	N/A	6
CTNNB1	catenin (cadherin-associated protein), beta 1, 88kDa	NS	N/A	2
CTPS1	CTP synthase 1	NS	N/A	6
CTSB	cathepsin B	NS	N/A	3
CTSZ	cathepsin Z	NS	N/A	4
CTTN	cortactin	NS	N/A	13
CUL2	cullin 2	NS	N/A	5
CUL4B	cullin 4B	NS	N/A	3
CUTA	cutA divalent cation tolerance homolog (E. coli)	NS	N/A	2
CYCS	cytochrome c, somatic	NS	N/A	7
DAG1	dystroglycan 1 (dystrophin-associated glycoprotein 1)	NS	N/A	7
DAK	dihydroxyacetone kinase 2 homolog	NS	N/A	8

DAP	death-associated protein	NS	N/A	5
DARS	aspartyl-tRNA synthetase	NS	N/A	13
DBI	Acyl-CoA-binding protein	NS	N/A	11
DBN1	drebrin 1	NS	N/A	9
DBNL	drebrin-like	NS	N/A	1
DCTN1	dynactin 1	NS	N/A	8
DCTN2	dynactin 2 (p50)	NS	N/A	7
DCTPP1	dCTP pyrophosphatase 1	NS	N/A	4
DCUN1D1	DCN1-like protein 1	NS	N/A	4
DDAH1	dimethylarginine dimethylaminohydrolase 1	NS	N/A	6
DDAH2	dimethylarginine dimethylaminohydrolase 2	NS	N/A	7
DDB1	damage-specific DNA binding protein 1, 127kDa	NS	N/A	27
DDT	D-dopachrome tautomerase	NS	N/A	4
DDX1	DEAD (Asp-Glu-Ala-Asp) box helicase 1	NS	N/A	7
DDX17	DEAD (Asp-Glu-Ala-Asp) box helicase 17	NS	N/A	10
DDX21	DEAD (Asp-Glu-Ala-Asp) box helicase 21	NS	N/A	2
DDX3X	DEAD (Asp-Glu-Ala-Asp) box helicase 3, X-linked	NS	N/A	5
DDX5	DEAD (Asp-Glu-Ala-Asp) box helicase 5	NS	N/A	8
DDX6	DEAD (Asp-Glu-Ala-Asp) box helicase 6	NS	N/A	3
DEK	DEK proto-oncogene	NS	N/A	4
DENR	density-regulated protein	NS	N/A	4
DHX15	DEAH (Asp-Glu-Ala-His) box helicase 15	NS	N/A	13
DIAPH1	diaphanous-related formin 1	NS	N/A	7
DLD	dihydrolipoamide dehydrogenase	NS	N/A	4
DNAJB1	DnaJ (Hsp40) homolog, subfamily B, member 1	NS	N/A	2
DNAJC8	DnaJ (Hsp40) homolog, subfamily C, member 8	NS	N/A	7
DNM2	dynamin 2	NS	N/A	6
DPP3	dipeptidyl-peptidase 3	NS	N/A	27
DPYSL2	dihydropyrimidinase-like 2	NS	N/A	9
DSTN	destrin (actin depolymerizing factor)	NS	N/A	5
DTD1	D-tyrosyl-tRNA deacylase 1	NS	N/A	1
DUSP3	dual specificity phosphatase 3	NS	N/A	4
DUT	deoxyuridine triphosphatase	NS	N/A	5
DYNC1H1	dynein, cytoplasmic 1, heavy chain 1	NS	N/A	5
DYNLL1	dynein, light chain, LC8-type 1	NS	N/A	2
DYNLL2	dynein, light chain, LC8-type 2	NS	N/A	2
DYNLRB1	dynein, light chain, roadblock-type 1	NS	N/A	4
ECH1	enoyl CoA hydratase 1, peroxisomal	NS	N/A	5

EDF1	endothelial differentiation-related factor 1	NS	N/A	8
EEA1	early endosome antigen 1	NS	N/A	7
EEF1A1	eukaryotic translation elongation factor 1 alpha 1	NS	N/A	25
EEF1B2	eukaryotic translation elongation factor 1 beta 2	NS	N/A	9
EEF1D	eukaryotic translation elongation factor 1 delta	NS	N/A	2
EEF1E1	eukaryotic translation elongation factor 1 epsilon 1	NS	N/A	2
EEF1G	eukaryotic translation elongation factor 1 gamma	NS	N/A	20
EEF2	eukaryotic translation elongation factor 2	NS	N/A	54
EFNA1	ephrin-A1	NS	N/A	4
EIF1AX	eukaryotic translation initiation factor 1A, X-linked	NS	N/A	6
EIF2A	eukaryotic translation initiation factor 2A, 65kDa	NS	N/A	6
EIF2S1	Eukaryotic translation initiation factor 2 subunit 1	NS	N/A	7
EIF2S2	Eukaryotic translation initiation factor 2 subunit 2	NS	N/A	5
EIF2S3	Eukaryotic translation initiation factor 2 subunit 3	NS	N/A	5
EIF3A	eukaryotic translation initiation factor 3, subunit A	NS	N/A	4
EIF3B	eukaryotic translation initiation factor 3, subunit B	NS	N/A	14
EIF3CL	eukaryotic translation initiation factor 3, subunit C-like	NS	N/A	19
EIF3D	eukaryotic translation initiation factor 3, subunit D	NS	N/A	2
EIF3E	eukaryotic translation initiation factor 3, subunit E	NS	N/A	6
EIF3F	eukaryotic translation initiation factor 3, subunit F	NS	N/A	5
EIF3G	eukaryotic translation initiation factor 3, subunit G	NS	N/A	7
EIF3H	eukaryotic translation initiation factor 3, subunit H	NS	N/A	5
EIF3I	eukaryotic translation initiation factor 3, subunit I	NS	N/A	10
EIF3J	eukaryotic translation initiation factor 3, subunit J	NS	N/A	6
EIF3K	eukaryotic translation initiation factor 3, subunit K	NS	N/A	3
EIF3L	eukaryotic translation initiation factor 3, subunit L	NS	N/A	10
EIF3M	eukaryotic translation initiation factor 3, subunit M	NS	N/A	5
EIF4A1	eukaryotic translation initiation factor 4A1	NS	N/A	10
EIF4A2	eukaryotic translation initiation factor 4A2	NS	N/A	1
EIF4A3	eukaryotic translation initiation factor 4A3	NS	N/A	12
EIF4B	eukaryotic translation initiation factor 4B	NS	N/A	19
EIF4EBP1	Eukaryotic translation initiation factor 4E-binding protein 1	NS	N/A	3
EIF4EBP2	Eukaryotic translation initiation factor 4E-binding protein 2	NS	N/A	1
EIF4G1	eukaryotic translation initiation factor 4 gamma, 1	NS	N/A	8
EIF5	eukaryotic translation initiation factor 5	NS	N/A	7
EIF5A	eukaryotic translation initiation factor 5A	NS	N/A	9

EIF6	eukaryotic translation initiation factor 6	NS	N/A	4
ELAVL1	ELAV like RNA binding protein 1	NS	N/A	7
ENAH	enabled homolog (Drosophila)	NS	N/A	3
ENO1	enolase 1, (alpha)	NS	N/A	50
ENOPH1	enolase-phosphatase 1	NS	N/A	1
EPB41L2	erythrocyte membrane protein band 4.1-like 2	NS	N/A	9
EPRS	glutamyl-prolyl-tRNA synthetase	NS	N/A	31
EPS15L1	Epidermal growth factor receptor substrate 15-like 1	NS	N/A	2
ERH	enhancer of rudimentary homolog (Drosophila)	NS	N/A	9
ERP44	endoplasmic reticulum protein 44	NS	N/A	4
ESD	esterase D	NS	N/A	4
EWSR1	EWS RNA-binding protein 1	NS	N/A	3
EXOSC9	exosome component 9	NS	N/A	1
EZR	ezrin	NS	N/A	27
F5	coagulation factor V (proaccelerin, labile factor)	NS	N/A	6
FABP1	fatty acid binding protein 1, liver	NS	N/A	11
FABP5	fatty acid binding protein 5 (psoriasis-associated)	NS	N/A	16
FAF1	Fas (TNFRSF6) associated factor 1	NS	N/A	2
FAM107B	family with sequence similarity 107, member B	NS	N/A	2
FAM136A	family with sequence similarity 136, member A	NS	N/A	4
FAM175B	family with sequence similarity 175, member B	NS	N/A	1
FARSB	phenylalanyl-tRNA synthetase, beta subunit	NS	N/A	9
FASN	fatty acid synthase	NS	N/A	0
FDPS	farnesyl diphosphate synthase	NS	N/A	9
FEN1	flap structure-specific endonuclease 1	NS	N/A	12
FGA	fibrinogen alpha chain	NS	N/A	38
FGB	fibrinogen beta chain	NS	N/A	20
FGG	fibrinogen gamma chain	NS	N/A	16
FGL1	fibrinogen-like 1	NS	N/A	8
FH	fumarate hydratase	NS	N/A	4
FKBP1A	FK506 binding protein 1A, 12kDa	NS	N/A	6
FKBP3	FK506 binding protein 3, 25kDa	NS	N/A	16
FKBP4	FK506 binding protein 4, 59kDa	NS	N/A	18
FLNA	filamin A, alpha	NS	N/A	25
FLNB	filamin B, beta	NS	N/A	33
FNTA	farnesyltransferase, CAAX box, alpha	NS	N/A	3
FSCN1	fascin actin-bundling protein 1	NS	N/A	7
FST	follistatin	NS	N/A	3

FUBP1	far upstream element (FUSE) binding protein 1	NS	N/A	13
G3BP1	ATP-dependent DNA helicase VIII	NS	N/A	8
G6PD	glucose-6-phosphate dehydrogenase	NS	N/A	20
GAMT	guanidinoacetate N-methyltransferase	NS	N/A	5
GAPDH	glyceraldehyde-3-phosphate dehydrogenase	NS	N/A	25
GARS	glycyl-tRNA synthetase	NS	N/A	14
GATM	glycine amidinotransferase	NS	N/A	4
GBE1	glucan (1,4-alpha-), branching enzyme 1	NS	N/A	2
GCSH	glycine cleavage system protein H	NS	N/A	1
GDF15	growth differentiation factor 15	NS	N/A	2
GDI1	GDP dissociation inhibitor 1	NS	N/A	3
GFPT1	glutamine--fructose-6-phosphate transaminase 1	NS	N/A	7
GGCT	gamma-glutamylcyclotransferase	NS	N/A	6
GGH	gamma-glutamyl hydrolase	NS	N/A	5
GLO1	glyoxalase I	NS	N/A	10
GLRX	glutaredoxin	NS	N/A	3
GLRX3	glutaredoxin 3	NS	N/A	2
GLUL	glutamate-ammonia ligase	NS	N/A	7
GMPS	guanine monphosphate synthase	NS	N/A	3
GNB1	guanine nucleotide binding protein , beta polypeptide 1	NS	N/A	3
GNB2L1	Cell proliferation-inducing gene 21 protein	NS	N/A	15
GNG5	guanine nucleotide binding protein, gamma 5	NS	N/A	2
GNPDA1	glucosamine-6-phosphate deaminase 1	NS	N/A	5
GNS	glucosamine (N-acetyl)-6-sulfatase	NS	N/A	3
GOLPH3	golgi phosphoprotein 3	NS	N/A	2
GOPC	golgi-associated PDZ and coiled-coil motif containing	NS	N/A	2
GORASP2	golgi reassembly stacking protein 2, 55kDa	NS	N/A	4
GOT1	glutamic-oxaloacetic transaminase 1, soluble	NS	N/A	16
GPC3	glypican 3	NS	N/A	4
GPI	glucose-6-phosphate isomerase	NS	N/A	27
GPR126	G protein-coupled receptor 126	NS	N/A	2
GPS1	G protein pathway suppressor 1	NS	N/A	6
GRB2	growth factor receptor-bound protein 2	NS	N/A	8
GRHPR	glyoxylate reductase/hydroxypyruvate reductase	NS	N/A	7
GRN	granulin	NS	N/A	10
GSPT1	G1 to S phase transition 1	NS	N/A	7
GSR	glutathione reductase	NS	N/A	12

GSS	glutathione synthetase	NS	N/A	17
GSTO1	glutathione S-transferase omega 1	NS	N/A	10
GSTP1	glutathione S-transferase pi 1	NS	N/A	9
HARS	histidyl-tRNA synthetase	NS	N/A	12
HAT1	histone acetyltransferase 1	NS	N/A	5
HDAC2	histone deacetylase 2	NS	N/A	3
HDGF	hepatoma-derived growth factor	NS	N/A	14
HDGFRP2	hepatoma-derived growth factor-related protein 2	NS	N/A	5
HDLBP	high density lipoprotein binding protein	NS	N/A	14
HECTD1	HECT domain containing E3 ubiquitin protein ligase 1	NS	N/A	2
HEXA	hexosaminidase A (alpha polypeptide)	NS	N/A	3
HGD	homogentisate 1,2-dioxygenase	NS	N/A	11
HINT1	histidine triad nucleotide binding protein 1	NS	N/A	7
HIST1H1B	histone cluster 1, H1b	NS	N/A	7
HIST1H1E	histone cluster 1, H1e	NS	N/A	5
HIST1H2AC	histone cluster 1, H2ac	NS	N/A	1
HIST2H2BE	histone cluster 2, H2be	NS	N/A	0
HMGA1	high mobility group AT-hook 1	NS	N/A	3
HMGB1	high mobility group box 1	NS	N/A	18
HMGB2	high mobility group box 2	NS	N/A	14
HMGB3	high mobility group box 3	NS	N/A	9
HMGCS1	3-hydroxy-3-methylglutaryl-CoA synthase 1	NS	N/A	19
HMGN1	high mobility group nucleosome binding domain 1	NS	N/A	5
HMGN2	high mobility group nucleosomal binding domain 2	NS	N/A	5
HN1	hematological and neurological expressed 1	NS	N/A	3
HN1L	hematological and neurological expressed 1-like	NS	N/A	7
HNMT	histamine N-methyltransferase	NS	N/A	8
HNRNPA0	heterogeneous nuclear ribonucleoprotein A0	NS	N/A	4
HNRNPA1	heterogeneous nuclear ribonucleoprotein A1	NS	N/A	19
HNRNPA2B1	heterogeneous nuclear ribonucleoprotein A2/B1	NS	N/A	4
HNRNPA3	heterogeneous nuclear ribonucleoprotein A3	NS	N/A	2
HNRNPAB	heterogeneous nuclear ribonucleoprotein A/B	NS	N/A	18
HNRNPC	heterogeneous nuclear ribonucleoprotein C (C1/C2)	NS	N/A	20
HNRNPD	heterogeneous nuclear ribonucleoprotein D	NS	N/A	13
HNRNPDL	heterogeneous nuclear ribonucleoprotein D-like	NS	N/A	3
HNRNPF	heterogeneous nuclear ribonucleoprotein F	NS	N/A	3
HNRNPK	heterogeneous nuclear ribonucleoprotein K	NS	N/A	22

HNRNPL*	heterogeneous nuclear ribonucleoprotein L	NS	N/A	13
HNRNPL*	heterogeneous nuclear ribonucleoprotein L	NS	N/A	13
HNRNPR	heterogeneous nuclear ribonucleoprotein R	NS	N/A	8
HNRNPU	heterogeneous nuclear ribonucleoprotein U	NS	N/A	16
HPX	hemopexin	NS	N/A	6
HRSP12	heat-responsive protein 12	NS	N/A	2
HSP90AA1	Heat shock 86 kDa	NS	N/A	0
HSP90AB1	Heat shock 84 kDa	NS	N/A	0
HSP90B1	heat shock protein 90kDa beta (Grp94), member 1	NS	N/A	21
HSPA8	heat shock 70kDa protein 8	NS	N/A	0
HSPA9	heat shock 70kDa protein 9 (mortalin)	NS	N/A	14
HSPBP1	Heat shock protein-binding protein 1	NS	N/A	5
HSPD1	heat shock 60kDa protein 1 (chaperonin)	NS	N/A	19
HSPG2	heparan sulfate proteoglycan 2	NS	N/A	11
HSPH1	heat shock 105kDa/110kDa protein 1	NS	N/A	20
HUWE1	ARF-binding protein 1	NS	N/A	11
IARS	isoleucyl-tRNA synthetase	NS	N/A	9
IDE	insulin-degrading enzyme	NS	N/A	5
IDH1	isocitrate dehydrogenase 1 (NADP+), soluble	NS	N/A	33
IGF2BP1	insulin-like growth factor 2 mRNA binding protein 1	NS	N/A	10
IGF2BP3	insulin-like growth factor 2 mRNA binding protein 3	NS	N/A	2
ILF2	interleukin enhancer binding factor 2	NS	N/A	5
IMPA1	inositol(myo)-1(or 4)-monophosphatase 1	NS	N/A	3
IMPA2	inositol(myo)-1(or 4)-monophosphatase 2	NS	N/A	3
IMPDH2	IMP (inosine 5'-monophosphate) dehydrogenase 2	NS	N/A	10
INS	insulin	NS	N/A	2
IPO5	importin 5	NS	N/A	18
IPO7	importin 7	NS	N/A	4
IRGQ	immunity-related GTPase family, Q	NS	N/A	3
JAG1	jagged 1	NS	N/A	6
KHDRBS1	GAP-associated tyrosine phosphoprotein p62	NS	N/A	12
KHSRP	KH-type splicing regulatory protein	NS	N/A	16
KIAA1143	KIAA1143	NS	N/A	1
KIAA1598	KIAA1598	NS	N/A	2
KIF1B	kinesin family member 1B	NS	N/A	1
KIF2A	kinesin heavy chain member 2A	NS	N/A	3
KIF5B	kinesin family member 5B	NS	N/A	20
KPNA2	karyopherin alpha 2 (RAG cohort 1, importin alpha 1)	NS	N/A	7

KPNA3	karyopherin alpha 3 (importin alpha 4)	NS	N/A	3
KPNA4	karyopherin alpha 4 (importin alpha 3)	NS	N/A	1
KPNB1	karyopherin (importin) beta 1	NS	N/A	19
KRT18	keratin 18	NS	N/A	24
KTN1	kinectin 1 (kinesin receptor)	NS	N/A	13
LACTB2	lactamase, beta 2	NS	N/A	7
LASP1	LIM and SH3 protein 1	NS	N/A	12
LDHA	lactate dehydrogenase A	NS	N/A	23
LGALS1	lectin, galactoside-binding, soluble, 1	NS	N/A	5
LGALS3	lectin, galactoside-binding, soluble, 3	NS	N/A	4
LGALS3BP	lectin, galactoside-binding, soluble, 3 binding protein	NS	N/A	7
LIN28B	lin-28 homolog B (C. elegans)	NS	N/A	4
LMAN2	lectin, mannose-binding 2	NS	N/A	5
LMNA	lamin A/C	NS	N/A	13
LMNB1	lamin B1	NS	N/A	7
LMNB2	lamin B2	NS	N/A	8
LRG1	leucine-rich alpha-2-glycoprotein 1	NS	N/A	5
LRRC47	leucine rich repeat containing 47	NS	N/A	11
LSM3	LSM3 homolog, U6 small nuclear RNA associated	NS	N/A	3
LSM6	LSM6 homolog, U6 small nuclear RNA associated	NS	N/A	5
LSM7	LSM7 homolog, U6 small nuclear RNA associated	NS	N/A	4
LSM8	LSM8 homolog, U6 small nuclear RNA associated	NS	N/A	2
LSR	lipolysis stimulated lipoprotein receptor	NS	N/A	2
LTA4H	leukotriene A4 hydrolase	NS	N/A	8
LUM	lumican	NS	N/A	1
LYPLAL1	lysophospholipase-like 1	NS	N/A	1
MAGOH	Protein mago nashi homolog	NS	N/A	1
MAGOHB	mago-nashi homolog B (Drosophila)	NS	N/A	1
MAN2A1	mannosidase, alpha, class 2A, member 1	NS	N/A	3
MAP4	microtubule-associated protein 4	NS	N/A	26
MAPK1	mitogen-activated protein kinase 1	NS	N/A	9
MAPK1IP1L	MAPK-interacting and spindle-stabilizing protein-like	NS	N/A	1
MAPRE1	APC-binding protein EB1	NS	N/A	7
MARCKS	myristoylated alanine-rich protein kinase C substrate	NS	N/A	4
MARCKSL1	MARCKS-like 1	NS	N/A	3
MARS	methionyl-tRNA synthetase	NS	N/A	8
MASP1	mannan-binding lectin serine peptidase 1	NS	N/A	5

MAT2A	methionine adenosyltransferase II, alpha	NS	N/A	6
MATR3	matrin 3	NS	N/A	6
MCM3	minichromosome maintenance complex component 3	NS	N/A	18
MCM4	minichromosome maintenance complex component 4	NS	N/A	10
MCM5	minichromosome maintenance complex component 5	NS	N/A	11
MCM6	minichromosome maintenance complex component 6	NS	N/A	16
MCM7	minichromosome maintenance complex component 7	NS	N/A	14
MDH1	malate dehydrogenase 1, NAD (soluble)	NS	N/A	20
MDH2	malate dehydrogenase 2, NAD (mitochondrial)	NS	N/A	16
MDK	midkine (neurite growth-promoting factor 2)	NS	N/A	8
ME1	malic enzyme 1, NADP(+)-dependent, cytosolic	NS	N/A	9
METAP1	methionyl aminopeptidase 1	NS	N/A	1
MGMT	O-6-methylguanine-DNA methyltransferase	NS	N/A	1
MIF	macrophage migration inhibitory factor	NS	N/A	4
MOB1B	MOB kinase activator 1B	NS	N/A	7
MPST	mercaptopyruvate sulfurtransferase	NS	N/A	3
MRI1	methylthioribose-1-phosphate isomerase 1	NS	N/A	3
MT1E	metallothionein 1E	NS	N/A	0
MT1X	metallothionein 1X	NS	N/A	1
MT2A	metallothionein 2A	NS	N/A	1
MTAP	methylthioadenosine phosphorylase	NS	N/A	4
MTHFD1	C-1-tetrahydrofolate synthase, cytoplasmic	NS	N/A	5
MTPN	myotrophin	NS	N/A	3
MTTP	microsomal triglyceride transfer protein	NS	N/A	4
MVD	mevalonate (diphospho) decarboxylase	NS	N/A	10
MYL12B	myosin, light chain 12B, regulatory	NS	N/A	2
NAA15	N(alpha)-acetyltransferase 15, NatA auxiliary subunit	NS	N/A	10
NAA50	N(alpha)-acetyltransferase 50, NatE catalytic subunit	NS	N/A	4
NACA	nascent polypeptide-associated complex alpha subunit	NS	N/A	6
NAMPT	nicotinamide phosphoribosyltransferase	NS	N/A	7
NANS	N-acetylneuraminic acid synthase	NS	N/A	8
NAP1L1	nucleosome assembly protein 1-like 1	NS	N/A	1
NAPA	Alpha-soluble NSF attachment protein	NS	N/A	2
NAPRT	nicotinate phosphoribosyltransferase	NS	N/A	4
NARS	asparaginyl-tRNA synthetase	NS	N/A	8

NASP	nuclear autoantigenic sperm protein (histone-binding)	NS	N/A	14
NCL	nucleolin	NS	N/A	53
NDRG1	N-myc downstream regulated 1	NS	N/A	2
NEDD8-MDP1	NEDD8-MDP1 readthrough	NS	N/A	3
NHP2L1	NHP2 non-histone chromosome protein 2-like 1	NS	N/A	4
NIT2	nitrilase family, member 2	NS	N/A	3
NLN	neurolysin (metallopeptidase M3 family)	NS	N/A	3
NME1-NME2	NME1-NME2 readthrough	NS	N/A	0
NMT1	N-myristoyltransferase 1	NS	N/A	9
NOLC1	nucleolar and coiled-body phosphoprotein 1	NS	N/A	7
NONO	non-POU domain containing, octamer-binding	NS	N/A	16
NOP2	NOP2 nucleolar protein	NS	N/A	4
NPEPPS	aminopeptidase puromycin sensitive	NS	N/A	26
NPM1*	nucleophosmin	NS	N/A	5
NPM1*	nucleophosmin	NS	N/A	5
NQO1	NAD(P)H dehydrogenase, quinone 1	NS	N/A	8
NRD1	nardilysin (N-arginine dibasic convertase)	NS	N/A	6
NSF	N-ethylmaleimide-sensitive factor	NS	N/A	4
NSFL1C	NSFL1 (p97) cofactor (p47)	NS	N/A	15
NT5DC1	5'-nucleotidase domain containing 1	NS	N/A	3
NTS	neurotensin	NS	N/A	6
NUCKS1	nuclear casein kinase and cyclin-dependent kinase substrate 1	NS	N/A	8
NUDC	nudC nuclear distribution protein	NS	N/A	11
NUDCD1	NudC domain containing 1	NS	N/A	1
NUDCD2	NudC domain containing 2	NS	N/A	3
NUDT21	nudix (nucleoside diphosphate linked moiety X)-type motif 21	NS	N/A	6
NUDT5	nudix (nucleoside diphosphate linked moiety X)-type motif 5	NS	N/A	7
NUMA1	nuclear mitotic apparatus protein 1	NS	N/A	7
NUTF2	nuclear transport factor 2	NS	N/A	4
OLA1	Obg-like ATPase 1	NS	N/A	10
ORM2	orosomucoid 2	NS	N/A	3
OSBP	oxysterol binding protein	NS	N/A	10
OTUB1	OTU deubiquitinase, ubiquitin aldehyde binding 1	NS	N/A	6
P4HB	prolyl 4-hydroxylase, beta polypeptide	NS	N/A	19
PA2G4	proliferation-associated 2G4, 38kDa	NS	N/A	27

PABPC1	poly(A) binding protein, cytoplasmic 1	NS	N/A	7
PABPC4	poly(A) binding protein, cytoplasmic 4 (inducible form)	NS	N/A	6
PABPN1	poly(A) binding protein, nuclear 1	NS	N/A	4
PACSIN2	protein kinase C and casein kinase substrate in neurons 2	NS	N/A	3
PAFAH1B1	Lissencephaly-1 protein	NS	N/A	6
PAFAH1B2	PAF acetylhydrolase 30 kDa subunit	NS	N/A	2
PAFAH1B3	PAF acetylhydrolase 29 kDa subunit	NS	N/A	8
PAICS	phosphoribosylaminoimidazole carboxylase	NS	N/A	14
PAIP1	poly(A) binding protein interacting protein 1	NS	N/A	4
PAK2	p21 protein (Cdc42/Rac)-activated kinase 2	NS	N/A	10
PARK7	parkinson protein 7	NS	N/A	12
PARP1	poly (ADP-ribose) polymerase 1	NS	N/A	24
PCBP1	poly(rC) binding protein 1	NS	N/A	7
PCBP2	poly(rC) binding protein 2	NS	N/A	3
PCMT1	protein-L-isoaspartate (D-aspartate) O-methyltransferase	NS	N/A	6
PCNA	proliferating cell nuclear antigen	NS	N/A	14
PCNP	PEST proteolytic signal containing nuclear protein	NS	N/A	10
PCOLCE	procollagen C-endopeptidase enhancer	NS	N/A	2
PCOLCE2	procollagen C-endopeptidase enhancer 2	NS	N/A	2
PCYT2	phosphate cytidylyltransferase 2, ethanolamine	NS	N/A	7
PDAP1	PDGFA associated protein 1	NS	N/A	9
PDCD5	programmed cell death 5	NS	N/A	10
PDCD6IP	programmed cell death 6 interacting protein	NS	N/A	23
PDE6H	phosphodiesterase 6H, cGMP-specific, cone, gamma	NS	N/A	7
PDIA3*	protein disulfide isomerase family A, member 3	NS	N/A	10
PDIA4	protein disulfide isomerase family A, member 4	NS	N/A	16
PDLIM1	PDZ and LIM domain 1	NS	N/A	10
PDLIM5	PDZ and LIM domain 5	NS	N/A	3
PDXP	pyridoxal (pyridoxine, vitamin B6) phosphatase	NS	N/A	2
PEBP1	phosphatidylethanolamine binding protein 1	NS	N/A	12
PEPD	peptidase D	NS	N/A	8
PFAS	phosphoribosylformylglycinamidine synthase	NS	N/A	20
PFDN1	prefoldin subunit 1	NS	N/A	4
PFDN2	prefoldin subunit 2	NS	N/A	5
PFDN6	prefoldin subunit 6	NS	N/A	6
PFKL	phosphofructokinase, liver	NS	N/A	3

PFN1	profilin 1	NS	N/A	17
PFN2	profilin 2	NS	N/A	2
PGAM1	phosphoglycerate mutase 1 (brain)	NS	N/A	15
PGD	phosphogluconate dehydrogenase	NS	N/A	17
PGK1	phosphoglycerate kinase 1	NS	N/A	28
PGLS	6-phosphogluconolactonase	NS	N/A	7
PGM1	phosphoglucomutase 1	NS	N/A	13
PGM2	phosphoglucomutase 2	NS	N/A	3
PGM3	phosphoglucomutase 3	NS	N/A	11
PHGDH	phosphoglycerate dehydrogenase	NS	N/A	18
PHPT1	phosphohistidine phosphatase 1	NS	N/A	5
PIN1	peptidylprolyl cis/trans isomerase, NIMA-interacting 1	NS	N/A	5
PIN4	Parvulin-14	NS	N/A	4
PITPNB	phosphatidylinositol transfer protein, beta	NS	N/A	5
PKM	pyruvate kinase, muscle	NS	N/A	29
PLEC	plectin	NS	N/A	9
PLOD1	procollagen-lysine, 2-oxoglutarate 5-dioxygenase 1	NS	N/A	4
PLS1	plastin 1	NS	N/A	2
PLS3	plastin 3	NS	N/A	16
PNP	purine nucleoside phosphorylase	NS	N/A	7
PPA1	pyrophosphatase (inorganic) 1	NS	N/A	7
PPIA	peptidylprolyl isomerase A (cyclophilin A)	NS	N/A	20
PPIB	peptidylprolyl isomerase B (cyclophilin B)	NS	N/A	15
PPIC	peptidylprolyl isomerase C (cyclophilin C)	NS	N/A	2
PPID	peptidylprolyl isomerase D	NS	N/A	6
PPIL1	peptidylprolyl isomerase (cyclophilin)-like 1	NS	N/A	4
PPM1G	protein phosphatase, Mg2+/Mn2+ dependent, 1G	NS	N/A	11
PPP1CA	protein phosphatase 1, catalytic subunit, alpha isozyme	NS	N/A	4
PPP1R7	protein phosphatase 1, regulatory subunit 7	NS	N/A	6
PPP2CA	protein phosphatase 2, catalytic subunit, alpha isozyme	NS	N/A	5
PPP2R1A	protein phosphatase 2, regulatory subunit A, alpha	NS	N/A	13
PPP2R4	protein phosphatase 2A activator, regulatory subunit 4	NS	N/A	5
PPP5C	protein phosphatase 5, catalytic subunit	NS	N/A	11
PRDX1	peroxiredoxin 1	NS	N/A	20
PRDX2	peroxiredoxin 2	NS	N/A	11

PRDX3	peroxiredoxin 3	NS	N/A	6
PRDX4	peroxiredoxin 4	NS	N/A	3
PRDX5	peroxiredoxin 5	NS	N/A	4
PRDX6	peroxiredoxin 6	NS	N/A	12
PREP	prolyl endopeptidase	NS	N/A	8
PRKACA	protein kinase, cAMP-dependent, catalytic, alpha	NS	N/A	5
PRKAR1A	protein kinase, cAMP-dependent, regulatory, type I, alpha	NS	N/A	2
PRKAR2A	protein kinase, cAMP-dependent, regulatory, type II, alpha	NS	N/A	4
PRKCSH	protein kinase C substrate 80K-H	NS	N/A	11
PRKDC	protein kinase, DNA-activated, catalytic polypeptide	NS	N/A	41
PRMT1	protein arginine methyltransferase 1	NS	N/A	10
PRMT5	protein arginine methyltransferase 5	NS	N/A	8
PROC	protein C	NS	N/A	3
PROSC	proline synthetase co-transcribed homolog (bacterial)	NS	N/A	1
PRPF19	pre-mRNA processing factor 19	NS	N/A	6
PRPF8	pre-mRNA processing factor 8	NS	N/A	6
PSAP	prosaposin	NS	N/A	14
PSAT1	phosphoserine aminotransferase 1	NS	N/A	22
PSIP1	PC4 and SFRS1 interacting protein 1	NS	N/A	3
PSMA1	proteasome subunit, alpha type, 1	NS	N/A	14
PSMA2	proteasome subunit, alpha type, 2	NS	N/A	6
PSMA3	proteasome subunit, alpha type, 3	NS	N/A	14
PSMA4	proteasome subunit, alpha type, 4	NS	N/A	10
PSMA5	proteasome subunit, alpha type, 5	NS	N/A	10
PSMA6	proteasome subunit, alpha type, 6	NS	N/A	14
PSMA7	proteasome subunit, alpha type, 7	NS	N/A	12
PSMB1	proteasome subunit, beta type, 1	NS	N/A	9
PSMB3	proteasome subunit, beta type, 3	NS	N/A	7
PSMB4	proteasome subunit, beta type, 4	NS	N/A	5
PSMB5	proteasome subunit, beta type, 5	NS	N/A	11
PSMB6	proteasome subunit, beta type, 6	NS	N/A	5
PSMB7	proteasome subunit, beta type, 7	NS	N/A	4
PSMC1	proteasome 26S subunit, ATPase, 1	NS	N/A	9
PSMC2	proteasome 26S subunit, ATPase, 2	NS	N/A	9
PSMC3	proteasome 26S subunit, ATPase, 3	NS	N/A	6
PSMC4	proteasome 26S subunit, ATPase, 4	NS	N/A	3

PSMC5	proteasome 26S subunit, ATPase, 5	NS	N/A	5
PSMC6	proteasome 26S subunit, ATPase, 6	NS	N/A	4
PSMD1	proteasome 26S subunit, non-ATPase, 1	NS	N/A	4
PSMD11	proteasome 26S subunit, non-ATPase, 11	NS	N/A	8
PSMD13	proteasome 26S subunit, non-ATPase, 13	NS	N/A	7
PSMD2	proteasome 26S subunit, non-ATPase, 2	NS	N/A	7
PSMD4	proteasome 26S subunit, non-ATPase, 4	NS	N/A	3
PSMD5	proteasome 26S subunit, non-ATPase, 5	NS	N/A	5
PSMD7	proteasome 26S subunit, non-ATPase, 7	NS	N/A	4
PSME1	proteasome activator subunit 1 (PA28 alpha)	NS	N/A	5
PSME2	proteasome activator subunit 2 (PA28 beta)	NS	N/A	5
PSME3	proteasome activator subunit 3 (PA28 gamma; Ki)	NS	N/A	3
PSPC1	paraspeckle component 1	NS	N/A	9
PSPH	phosphoserine phosphatase	NS	N/A	11
PTBP1	polypyrimidine tract binding protein 1	NS	N/A	15
PTGES3	prostaglandin E synthase 3 (cytosolic)	NS	N/A	6
PTGR1	prostaglandin reductase 1	NS	N/A	10
PTMA	prothymosin, alpha	NS	N/A	9
PTMS	parathymosin	NS	N/A	8
PTPN11	protein tyrosine phosphatase, non-receptor type 11	NS	N/A	4
PTPRK	protein tyrosine phosphatase, receptor type, K	NS	N/A	5
PVR	poliovirus receptor	NS	N/A	3
PYGB	phosphorylase, glycogen; brain	NS	N/A	4
PYGL	phosphorylase, glycogen, liver	NS	N/A	11
QARS	glutaminyl-tRNA synthetase	NS	N/A	10
QDPR	quinoid dihydropteridine reductase	NS	N/A	10
QPRT	quinolinate phosphoribosyltransferase	NS	N/A	8
QSOX1	quiescin Q6 sulfhydryl oxidase 1	NS	N/A	8
RAB11B	RAB11B, member RAS oncogene family	NS	N/A	10
RAB14	RAB14, member RAS oncogene family	NS	N/A	3
RAB1B	RAB1B, member RAS oncogene family	NS	N/A	1
RAB21	RAB21, member RAS oncogene family	NS	N/A	1
RAB5C	RAB5C, member RAS oncogene family	NS	N/A	4
RAB7A	RAB7A, member RAS oncogene family	NS	N/A	8
RAC1	ras-related C3 botulinum toxin substrate 1	NS	N/A	9
RAD23B	RAD23 homolog B	NS	N/A	2
RALY	RALY heterogeneous nuclear ribonucleoprotein	NS	N/A	3
RAN	RAN, member RAS oncogene family	NS	N/A	14

RANBP1	RAN binding protein 1	NS	N/A	13
RANGAP1	Ran GTPase activating protein 1	NS	N/A	7
RAP1B	RAP1B, member of RAS oncogene family	NS	N/A	2
RARRES2	retinoic acid receptor responder (tazarotene induced) 2	NS	N/A	5
RARS	arginyl-tRNA synthetase	NS	N/A	14
RBBP4	retinoblastoma binding protein 4	NS	N/A	4
RBM3	RNA binding motif (RNP1, RRM) protein 3	NS	N/A	4
RBM8A	RNA binding motif protein 8A	NS	N/A	4
RBMX	RNA binding motif protein, X-linked	NS	N/A	8
RBP2	retinol binding protein 2, cellular	NS	N/A	2
RCC2	regulator of chromosome condensation 2	NS	N/A	15
RDX	radixin	NS	N/A	22
RHOA	ras homolog family member A	NS	N/A	4
RNH1	ribonuclease/angiogenin inhibitor 1	NS	N/A	9
RNMT	RNA (guanine-7-) methyltransferase	NS	N/A	3
RNPEP	arginyl aminopeptidase (aminopeptidase B)	NS	N/A	5
ROCK1	Rho-associated, coiled-coil containing protein kinase 1	NS	N/A	3
RPA3	replication protein A3, 14kDa	NS	N/A	2
RPE	ribulose-5-phosphate-3-epimerase	NS	N/A	2
RPL10A	ribosomal protein L10a	NS	N/A	13
RPL11	ribosomal protein L11	NS	N/A	7
RPL12	ribosomal protein L12	NS	N/A	8
RPL13	ribosomal protein L13	NS	N/A	5
RPL14	ribosomal protein L14	NS	N/A	11
RPL15	ribosomal protein L15	NS	N/A	5
RPL17	ribosomal protein L17	NS	N/A	3
RPL18	ribosomal protein L18	NS	N/A	6
RPL18A	ribosomal protein L18a	NS	N/A	4
RPL21	ribosomal protein L21	NS	N/A	5
RPL22	ribosomal protein L22	NS	N/A	3
RPL23	ribosomal protein L23	NS	N/A	4
RPL23A	ribosomal protein L23a	NS	N/A	4
RPL24	ribosomal protein L24	NS	N/A	8
RPL26	ribosomal protein L26	NS	N/A	1
RPL27	ribosomal protein L27	NS	N/A	5
RPL27A	ribosomal protein L27a	NS	N/A	3

RPL28	ribosomal protein L28	NS	N/A	9
RPL29	ribosomal protein L29	NS	N/A	4
RPL3	ribosomal protein L3	NS	N/A	9
RPL30	ribosomal protein L30	NS	N/A	7
RPL31	ribosomal protein L31	NS	N/A	2
RPL32	ribosomal protein L32	NS	N/A	5
RPL34	ribosomal protein L34	NS	N/A	9
RPL35	ribosomal protein L35	NS	N/A	8
RPL35A	ribosomal protein L35a	NS	N/A	7
RPL36A	ribosomal protein L36a	NS	N/A	3
RPL37	ribosomal protein L37	NS	N/A	4
RPL37A	ribosomal protein L37a	NS	N/A	2
RPL4	ribosomal protein L4	NS	N/A	14
RPL6	ribosomal protein L6	NS	N/A	6
RPL7	ribosomal protein L7	NS	N/A	6
RPL7A	ribosomal protein L7a	NS	N/A	6
RPL8	ribosomal protein L8	NS	N/A	6
RPLP0	ribosomal protein, large, P0	NS	N/A	12
RPLP2	ribosomal protein, large, P2	NS	N/A	3
RPRD1B	regulation of nuclear pre-mRNA domain containing 1B	NS	N/A	3
RPS10	ribosomal protein S10	NS	N/A	7
RPS11	ribosomal protein S11	NS	N/A	17
RPS12	ribosomal protein S12	NS	N/A	10
RPS13	ribosomal protein S13	NS	N/A	6
RPS14	ribosomal protein S14	NS	N/A	7
RPS15	ribosomal protein S15	NS	N/A	2
RPS15A	ribosomal protein S15a	NS	N/A	6
RPS16	ribosomal protein S16	NS	N/A	12
RPS18	ribosomal protein S18	NS	N/A	8
RPS19	ribosomal protein S19	NS	N/A	14
RPS2	ribosomal protein S2	NS	N/A	11
RPS20	ribosomal protein S20	NS	N/A	8
RPS21	ribosomal protein S21	NS	N/A	9
RPS23	ribosomal protein S23	NS	N/A	5
RPS24	ribosomal protein S24	NS	N/A	4
RPS25	ribosomal protein S25	NS	N/A	6
RPS26	ribosomal protein S26	NS	N/A	4

RPS27A	ribosomal protein S27a	NS	N/A	3
RPS27L	ribosomal protein S27-like	NS	N/A	3
RPS28	ribosomal protein S28	NS	N/A	10
RPS29	ribosomal protein S29	NS	N/A	3
RPS3	ribosomal protein S3	NS	N/A	16
RPS3A	ribosomal protein S3A	NS	N/A	20
RPS4X	ribosomal protein S4, X-linked	NS	N/A	9
RPS5	ribosomal protein S5	NS	N/A	8
RPS6	ribosomal protein S6	NS	N/A	16
RPS6KA3	ribosomal protein S6 kinase, 90kDa, polypeptide 3	NS	N/A	13
RPS7	ribosomal protein S7	NS	N/A	5
RPS8	ribosomal protein S8	NS	N/A	11
RPS9	ribosomal protein S9	NS	N/A	12
RPSAP58	ribosomal protein SA pseudogene 58	NS	N/A	9
RRP15	ribosomal RNA processing 15 homolog	NS	N/A	1
RSL1D1	ribosomal L1 domain containing 1	NS	N/A	2
RTCB	RNA 2',3'-cyclic phosphate and 5'-OH ligase	NS	N/A	8
RUVBL1	RuvB-like AAA ATPase 1	NS	N/A	9
RUVBL2	RuvB-like AAA ATPase 2	NS	N/A	14
S100A11	S100 calcium binding protein A11	NS	N/A	5
S100A13	S100 calcium binding protein A13	NS	N/A	3
S100P	S100 calcium binding protein P	NS	N/A	6
SAE1	SUMO1 activating enzyme subunit 1	NS	N/A	8
SAFB	scaffold attachment factor B	NS	N/A	4
SARNP	SAP domain containing ribonucleoprotein	NS	N/A	11
SARS	seryl-tRNA synthetase	NS	N/A	5
SBDS	Shwachman-Bodian-Diamond syndrome	NS	N/A	12
SCFD1	sec1 family domain containing 1	NS	N/A	12
SCP2	sterol carrier protein 2	NS	N/A	2
SCRN1	secernin 1	NS	N/A	5
SEC13	SEC13 homolog	NS	N/A	4
SEC22B	SEC22 vesicle trafficking protein homolog B	NS	N/A	2
SEC23A	Sec23 homolog A	NS	N/A	8
SEC31A	SEC31 homolog A	NS	N/A	9
SERBP1	SERPINE1 mRNA binding protein 1	NS	N/A	14
SERF2	small EDRK-rich factor 2	NS	N/A	5
SERPINB1	serpin peptidase inhibitor, clade B, member 1	NS	N/A	11
SERPINB6	serpin peptidase inhibitor, clade B, member 6	NS	N/A	4

SERPINB9	serpin peptidase inhibitor, clade B, member 9	NS	N/A	5
SERPINC1	serpin peptidase inhibitor, clade C, member 1	NS	N/A	10
SERPINE1	serpin peptidase inhibitor, clade E, member 1	NS	N/A	8
SERPING1	serpin peptidase inhibitor, clade G, member 1	NS	N/A	14
SERPINH1	serpin peptidase inhibitor, clade H , member 1	NS	N/A	1
SET	SET nuclear proto-oncogene	NS	N/A	10
SF1	splicing factor 1	NS	N/A	2
SF3A2	splicing factor 3a, subunit 2, 66kDa	NS	N/A	4
SF3A3	splicing factor 3a, subunit 3, 60kDa	NS	N/A	8
SF3B1	splicing factor 3b, subunit 1, 155kDa	NS	N/A	4
SF3B2	splicing factor 3b, subunit 2, 145kDa	NS	N/A	7
SF3B3	splicing factor 3b, subunit 3, 130kDa	NS	N/A	14
SF3B4	splicing factor 3b, subunit 4, 49kDa	NS	N/A	2
SF3B6	splicing factor 3b, subunit 6, 14kDa	NS	N/A	5
SFPQ	splicing factor proline/glutamine-rich	NS	N/A	28
SGTA	Alpha-SGT	NS	N/A	9
SH3BGRL3	SH3 domain binding glutamate-rich protein like 3	NS	N/A	4
SHMT1	serine hydroxymethyltransferase 1 (soluble)	NS	N/A	3
SHMT2	serine hydroxymethyltransferase 2 (mitochondrial)	NS	N/A	7
SKP1	S-phase kinase-associated protein 1	NS	N/A	5
SLC9A3R1	solute carrier family 9, subfamily A, member 3 regulator 1	NS	N/A	14
SMARCC1	SWI/SNF complex subunit SMARCC1	NS	N/A	3
SMC1A	structural maintenance of chromosomes 1A	NS	N/A	7
SMEK1	SMEK homolog 1, suppressor of mek1 (Dictyostelium)	NS	N/A	2
SMS	spermine synthase	NS	N/A	4
SMU1	smu-1 suppressor of mec-8 and unc-52 homolog (C. elegans)	NS	N/A	3
SND1	staphylococcal nuclease and tudor domain containing 1	NS	N/A	31
SNRNP200	small nuclear ribonucleoprotein 200kDa (U5)	NS	N/A	8
SNRNP70	small nuclear ribonucleoprotein 70kDa (U1)	NS	N/A	8
SNRPA	small nuclear ribonucleoprotein polypeptide A	NS	N/A	4
SNRPA1	small nuclear ribonucleoprotein polypeptide A'	NS	N/A	2
SNRPB2	small nuclear ribonucleoprotein polypeptide B	NS	N/A	3
SNRPD1	small nuclear ribonucleoprotein D1 polypeptide 16kDa	NS	N/A	1
SNRPD2	small nuclear ribonucleoprotein D2 polypeptide 16.5kDa	NS	N/A	3

SNRPD3	small nuclear ribonucleoprotein D3 polypeptide 18kDa	NS	N/A	3
SNRPF	small nuclear ribonucleoprotein polypeptide F	NS	N/A	3
SNRPG	small nuclear ribonucleoprotein polypeptide G	NS	N/A	3
SNW1	SNW domain containing 1	NS	N/A	2
SNX12	sorting nexin 12	NS	N/A	3
SNX2	sorting nexin 2	NS	N/A	3
SNX3	sorting nexin 3	NS	N/A	4
SOD1	superoxide dismutase 1, soluble	NS	N/A	5
SORD	sorbitol dehydrogenase	NS	N/A	11
SPTAN1	spectrin, alpha, non-erythrocytic 1	NS	N/A	54
SPTBN1	spectrin, beta, non-erythrocytic 1	NS	N/A	5
SRI	sorcin	NS	N/A	3
SRM	spermidine synthase	NS	N/A	7
SRP14	18 kDa Alu RNA-binding protein	NS	N/A	7
SRP19	signal recognition particle 19kDa	NS	N/A	1
SRP9	signal recognition particle 9kDa	NS	N/A	10
SRRT	serrate, RNA effector molecule	NS	N/A	2
SRSF1	serine/arginine-rich splicing factor 1	NS	N/A	7
SRSF2	serine/arginine-rich splicing factor 2	NS	N/A	9
SRSF3	serine/arginine-rich splicing factor 3	NS	N/A	5
SRSF6	serine/arginine-rich splicing factor 6	NS	N/A	4
SSB	Sjogren syndrome antigen B (autoantigen La)	NS	N/A	19
SSRP1	structure specific recognition protein 1	NS	N/A	12
ST13	suppression of tumorigenicity 13 (colon carcinoma)	NS	N/A	13
ST6GAL1	ST6 beta-galactosamide alpha-2,6-sialyltranferase 1	NS	N/A	9
STIP1	stress-induced phosphoprotein 1	NS	N/A	34
STMN1	stathmin 1	NS	N/A	25
STRAP	serine/threonine kinase receptor associated protein	NS	N/A	10
SUB1	SUB1 homolog	NS	N/A	7
SUGT1	SGT1, suppressor of G2 allele of SKP1	NS	N/A	5
SULT2A1	Bile salt sulfotransferase	NS	N/A	3
SUMO1	small ubiquitin-like modifier 1	NS	N/A	7
SUMO2	small ubiquitin-like modifier 2	NS	N/A	5
SUPT5H	suppressor of Ty 5 homolog	NS	N/A	2
SYNCRIP	Glycine- and tyrosine-rich RNA-binding protein	NS	N/A	0
TAF15	68 kDa TATA-binding protein-associated factor	NS	N/A	3
TAGLN2	transgelin 2	NS	N/A	12

TALDO1	transaldolase 1	NS	N/A	27
TARS	threonyl-tRNA synthetase	NS	N/A	17
TBCA	tubulin folding cofactor A	NS	N/A	16
TBCB	tubulin folding cofactor B	NS	N/A	5
TCEA1	transcription elongation factor A (SII), 1	NS	N/A	11
TCEB1	Elongin 15 kDa subunit	NS	N/A	4
TES	testis derived transcript (3 LIM domains)	NS	N/A	10
TFG	TRK-fused gene	NS	N/A	2
TFPI	tissue factor pathway inhibitor	NS	N/A	3
THBS4	thrombospondin 4	NS	N/A	6
THOP1	thimet oligopeptidase 1	NS	N/A	11
THUMPD1	THUMP domain containing 1	NS	N/A	3
THUMPD3	THUMP domain containing 3	NS	N/A	1
THYN1	thymocyte nuclear protein 1	NS	N/A	4
TJP2	tight junction protein 2	NS	N/A	5
TKT	transketolase	NS	N/A	29
TLN1	talin 1	NS	N/A	51
TMA7	translation machinery associated 7 homolog	NS	N/A	9
TMEM109	transmembrane protein 109	NS	N/A	4
TMPO	thymopoietin	NS	N/A	6
TMSB10/TMSB4X*	thymosin beta 10	NS	N/A	9
TMSB10/TMSB4X*	thymosin beta 10	NS	N/A	9
TNPO1	transportin 1	NS	N/A	1
TNPO2	transportin 2	NS	N/A	2
TNPO3	transportin 3	NS	N/A	4
TOP1	topoisomerase (DNA) I	NS	N/A	4
TPD52	tumor protein D52	NS	N/A	4
TPD52L2	tumor protein D52-like 2	NS	N/A	4
TPI1	triosephosphate isomerase 1	NS	N/A	19
TPM1	tropomyosin 1 (alpha)	NS	N/A	5
TPM3	tropomyosin 3	NS	N/A	17
TPM4	tropomyosin 4	NS	N/A	8
TPP2	tripeptidyl peptidase II	NS	N/A	9
TPT1	tumor protein, translationally-controlled 1	NS	N/A	9
TRA2B	transformer 2 beta homolog (Drosophila)	NS	N/A	2
TRAPPC3	trafficking protein particle complex 3	NS	N/A	2
TRIM28	tripartite motif containing 28	NS	N/A	19

TROVE2	TROVE domain family, member 2	NS	N/A	2
TSN	translin	NS	N/A	8
TSNAX	translin-associated factor X	NS	N/A	6
TSTA3	tissue specific transplantation antigen P35B	NS	N/A	2
TUBB	tubulin, beta class I	NS	N/A	3
TUBB2A	tubulin, beta 2A class IIa	NS	N/A	4
TUBB4B	tubulin, beta 4B class IVb	NS	N/A	8
TWF1	twinfilin actin-binding protein 1	NS	N/A	4
TXLNA	taxilin alpha	NS	N/A	3
TXN	thioredoxin	NS	N/A	9
TXNDC17	thioredoxin domain containing 17	NS	N/A	7
TXNDC5	thioredoxin domain containing 5	NS	N/A	6
TXNRD1	thioredoxin reductase 1	NS	N/A	10
U2AF2	U2 small nuclear RNA auxiliary factor 2	NS	N/A	7
UBA1	ubiquitin-like modifier activating enzyme 1	NS	N/A	26
UBA2	ubiquitin-like modifier activating enzyme 2	NS	N/A	14
UBA6	ubiquitin-like modifier activating enzyme 6	NS	N/A	4
UBAP2L	ubiquitin associated protein 2-like	NS	N/A	3
UBE2D3	ubiquitin-conjugating enzyme E2D 3	NS	N/A	2
UBE2I	ubiquitin-conjugating enzyme E2I	NS	N/A	4
UBE2K	ubiquitin-conjugating enzyme E2K	NS	N/A	5
UBE2L3	ubiquitin-conjugating enzyme E2L 3	NS	N/A	8
UBE2M	ubiquitin-conjugating enzyme E2M	NS	N/A	9
UBE2N	ubiquitin-conjugating enzyme E2N	NS	N/A	8
UBE2V1	ubiquitin-conjugating enzyme E2 variant 1	NS	N/A	5
UBE2V2	ubiquitin-conjugating enzyme E2 variant 2	NS	N/A	3
UBQLN2	ubiquilin 2	NS	N/A	2
UCHL1	Neuron cytoplasmic protein 9.5	NS	N/A	5
UCHL5	ubiquitin carboxyl-terminal hydrolase L5	NS	N/A	3
UFC1	ubiquitin-fold modifier conjugating enzyme 1	NS	N/A	4
UGDH	UDP-glucose 6-dehydrogenase	NS	N/A	16
UGP2	UDP-glucose pyrophosphorylase 2	NS	N/A	13
UPF1	UPF1 regulator of nonsense transcripts homolog (yeast)	NS	N/A	5
UROD	uroporphyrinogen decarboxylase	NS	N/A	4
USO1	USO1 vesicle transport factor	NS	N/A	7
USP14	ubiquitin specific peptidase 14	NS	N/A	11
USP15	ubiquitin specific peptidase 15	NS	N/A	2

USP5	ubiquitin specific peptidase 5	NS	N/A	11
USP7	ubiquitin specific peptidase 7	NS	N/A	5
VASN	vasorin	NS	N/A	2
VASP	vasodilator-stimulated phosphoprotein	NS	N/A	4
VAT1	vesicle amine transport 1	NS	N/A	4
VBP1	von Hippel-Lindau binding protein 1	NS	N/A	6
VCAN	versican	NS	N/A	2
VCL	vinculin	NS	N/A	16
VIL1	villin 1	NS	N/A	20
VIM	vimentin	NS	N/A	15
VPS35	vacuolar protein sorting 35 homolog	NS	N/A	5
VTA1	vesicle (multivesicular body) trafficking 1	NS	N/A	3
WARS	tryptophanyl-tRNA synthetase	NS	N/A	8
WBP11	WW domain binding protein 11	NS	N/A	5
WDR1	WD repeat domain 1	NS	N/A	20
WIBG	within bgcn homolog (Drosophila)	NS	N/A	2
XPO1	exportin 1	NS	N/A	11
XPOT	exportin, tRNA	NS	N/A	3
XRCC6	5-3 exoribonuclease 2	NS	N/A	19
XRN2	5'-3' exoribonuclease 2	NS	N/A	2
XYLB	xylulokinase homolog (H. influenzae)	NS	N/A	6
YARS	tyrosyl-tRNA synthetase	NS	N/A	13
YBX1	Y box binding protein 1	NS	N/A	14
YKT6	YKT6 v-SNARE homolog	NS	N/A	3
YTHDF3	YTH domain family, member 3	NS	N/A	4
YWHAB*	tyrosine 3-monooxygenase, beta	NS	N/A	1
YWHAB*	tyrosine 3-monooxygenase, beta	NS	N/A	1
YWHAE	tyrosine 3-monooxygenase, epsilon	NS	N/A	18
YWHAG	tyrosine 3-monooxygenase, gamma	NS	N/A	13
YWHAH	tyrosine 3-monooxygenase, eta	NS	N/A	9
YWHAQ	tyrosine 3-monooxygenase, theta	NS	N/A	13
YWHAZ	tyrosine 3-monooxygenase, zeta	NS	N/A	17
ZC3HAV1L	zinc finger CCCH-type, antiviral 1-like	NS	N/A	5
EIF3D	Eukaryotic translation initiation factor 3, subunit D	NS	N/A	2
HPRT	Hypoxanthine-guanine phosphoribosyltransferase	NS	N/A	3
DNCI2	Cytoplasmic dynein 1 intermediate chain 2	NS	N/A	4
CDH1	Cadherin-1	NS	N/A	3
PALB	Prealbumin	NS	N/A	2

H3FA	Histone H3.1	NS	N/A	8
KIAA0664	Putative uncharacterized protein KIAA0664	NS	N/A	9
TCOF1	Treacher Collins syndrome protein	NS	N/A	9
GMFB	Glia maturation factor beta	NS	N/A	5
TOLLIP	Toll-interacting protein	NS	N/A	3
DAAP-21F2.2-001	Valyl-tRNA synthetase	NS	N/A	9
H2AFJ	Histone H2A	NS	N/A	1
BAT3	HLA-B-associated transcript 3	NS	N/A	2
hCG_29955	SMARCA4 isoform 2	NS	N/A	2
FAM21A	WASH complex subunit FAM21A	NS	N/A	1
H4/A	Histone H4	NS	N/A	10
H2BFA	Histone H2B type 1-C/E/F/G/I	NS	N/A	0
HSPA1	Heat shock 70 kDa protein 1/2	NS	N/A	14
BM28	DNA replication licensing factor MCM2	NS	N/A	25
RPS17	40S ribosomal protein S17	NS	N/A	5
C17orf25	Glyoxalase domain-containing protein 4	NS	N/A	10
NAP1L4	Nucleosome assembly protein 1-like 4b	NS	N/A	6
GDI2	Guanosine diphosphate dissociation inhibitor 2	NS	N/A	21
CAPZB	Capping protein (Actin filament) muscle Z-line, beta	NS	N/A	11
BAT1	56 kDa U2AF65-associated protein	NS	N/A	19
UCHL3	Ubiquitin carboxyl-terminal hydrolase isozyme L3	NS	N/A	5
TIA1	Nucleolysin TIA-1 isoform p15	NS	N/A	1

Abbreviation: Not significant, NS; Not clustered, N/A.

Appendix 4. Clustering of proteins significantly changed in the analyses of secretomes from HUH7 cells overexpressing PCSK9 and its variants

Gene Name	Protein Name	ANOVA Significant	Cluster	Unique Peptides	Cellular Location
EIF4H	eukaryotic translation initiation factor 4H	+	A	12	C
ERP29	endoplasmic reticulum protein 29	+	A	6	C
GART	phosphoribosylglycinamide formyltransferase	+	A	7	C
HSPA4	heat shock 70kDa protein 4	+	A	31	C
LARS	leucyl-tRNA synthetase	+	A	5	C
PGP	phosphoglycolate phosphatase	+	A	7	C
RAD23A	RAD23 homolog A	+	A	1	N
XRCC5	70 kDa subunit of Ku antigen	+	A	15	N
ARPC4	actin related protein 2/3 complex, subunit 4, 20kDa	+	B	7	C
BOLA2/BOLA2B	bolA family member 2	+	B	5	C
CCT3	chaperonin containing TCP1, subunit 3 (gamma)	+	B	29	C
CCT5	chaperonin containing TCP1, subunit 5 (epsilon)	+	B	2	C
DHX9	DEAH (Asp-Glu-Ala-His) box helicase 9	+	B	19	N
HNRNPM	heterogeneous nuclear ribonucleoprotein M	+	B	23	N
ILF3	interleukin enhancer binding factor 3, 90kDa	+	B	16	N
KARS	lysyl-tRNA synthetase	+	B	23	C
LAP3	leucine aminopeptidase 3	+	B	16	C
PLIN3	perilipin 3	+	B	6	C
PSMB2	proteasome subunit, beta type, 2	+	B	9	C
RPL19	ribosomal protein L19	+	B	8	C
RPL5	ribosomal protein L5	+	B	23	C
SUPT16H	suppressor of Ty 16 homolog	+	B	12	N
TCP1	t-complex 1	+	B	20	C
VCP	valosin containing protein	+	B	49	C
VPS26A	vacuolar protein sorting 26 homolog A	+	B	1	C
VSNL1	visinin-like 1	+	B	2	C
ACTN3	actinin, alpha 3	+	C	1	PM
HNRNPH1	heterogeneous nuclear ribonucleoprotein H1 (H)	+	C	3	N
HSPB1	heat shock 27kDa protein 1	+	C	6	C
PCSK9	proprotein convertase subtilisin/kexin type 9	+	C	28	ES
PSMD3	proteasome 26S subunit, non-ATPase, 3	+	C	8	C
ISOC1	isochorismatase domain containing 1	+	D	5	C
LUC7L2	LUC7-like 2	+	D	2	N
ADM	adrenomedullin	+	E	1	ES
AFP	alpha-fetoprotein	+	E	0	ES

AGT	Angiotensin I	+	E	10	ES
AMBP	alpha-1-microglobulin/bikunin precursor	+	E	21	ES
ANG	angiogenin, ribonuclease, RNase A family, 5	+	E	4	ES
ANGPTL3	angiopoietin-like 3	+	E	8	ES
ANXA2	annexin A2	+	E	10	PM
ANXA5	annexin A5	+	E	5	PM
APOA1	apolipoprotein A-I	+	E	6	ES
APOA2	apolipoprotein A-II	+	E	7	ES
APOB	apolipoprotein B	+	E	0	ES
APOC2	apolipoprotein C-II	+	E	5	ES
APOC3	apolipoprotein C-III	+	E	3	ES
APOE	apolipoprotein E	+	E	0	ES
APOH	apolipoprotein H (beta-2-glycoprotein I)	+	E	13	ES
APOM	apolipoprotein M	+	E	6	PM
APP	amyloid beta (A4) precursor protein	+	E	8	PM
B2M	beta-2-microglobulin	+	E	5	PM
B3GNT1	I-beta-1,3-N-acetylglucosaminyltransferase	+	E	15	C
BNC2	basonuclin 2	+	E	2	N
C3	complement component 3	+	E	90	ES
C5	complement component 5	+	E	35	ES
CDH2	cadherin 2, type 1, N-cadherin (neuronal)	+	E	6	PM
CFH	complement factor H	+	E	31	ES
CLSTN1	calsyntenin 1	+	E	16	PM
CLSTN3	calsyntenin 3	+	E	8	PM
CLU	clusterin	+	E	31	C
COCH	cochlin	+	E	18	ES
COL2A1	collagen, type II, alpha 1	+	E	22	ES
COL5A2	collagen, type V, alpha 2	+	E	18	ES
CPVL	carboxypeptidase, vitellogenic-like	+	E	9	C
CST3	cystatin C	+	E	8	ES
CTSC	cathepsin C	+	E	6	C
CTSD	cathepsin D	+	E	7	C
CTSL	cathepsin L	+	E	1	C
CXCL5	chemokine (C-X-C motif) ligand 5	+	E	3	ES
EFEMP1	EGF containing fibulin-like extracellular matrix protein 1	+	E	11	ES
F10	coagulation factor X	+	E	6	ES
F12	coagulation factor XII (Hageman factor)	+	E	7	ES
F2	coagulation factor II (thrombin)	+	E	12	ES
FAM3C	family with sequence similarity 3, member C	+	E	6	ES
FBLN1*	fibulin 1	+	E	4	ES
FBLN1*	fibulin 1	+	E	4	ES
FN1	fibronectin 1	+	E	62	ES

FUCA2	fucosidase, alpha-L- 2, plasma	+	E	8	ES
GALNT2	polypeptide N-acetylgalactosaminyltransferase 2	+	E	3	C
GANAB	glucosidase, alpha; neutral AB	+	E	17	C
GC	group-specific component	+	E	25	ES
GLA	galactosidase, alpha	+	E	5	C
GOLM1	golgi membrane protein 1	+	E	5	C
HP	haptoglobin	+	E	8	ES
HSPA5	78 kDa glucose-regulated protein	+	E	29	C
HSPE1	heat shock 10kDa protein 1	+	E	14	C
IGF2	insulin-like growth factor 2	+	E	10	ES
IGFBP7	insulin-like growth factor binding protein 7	+	E	1	ES
ITIH2	inter-alpha-trypsin inhibitor heavy chain 2	+	E	31	ES
KIF1C	kinesin family member 1C	+	E	1	C
KNG1	kininogen 1	+	E	24	ES
LAMB1	laminin, beta 1	+	E	19	ES
LAMC1	laminin, gamma 1 (formerly LAMB2)	+	E	10	ES
LYZ	lysozyme	+	E	8	ES
MAN1A1	mannosidase, alpha, class 1A, member 1	+	E	20	C
MATN3	matrilin 3	+	E	3	ES
MEP1A	meprin A, alpha (PABA peptide hydrolase)	+	E	12	PM
MYH9	myosin, heavy chain 9, non-muscle	+	E	7	C
NEU1	sialidase 1 (lysosomal sialidase)	+	E	3	C
NID1	nidogen 1	+	E	37	ES
NPC2	Niemann-Pick disease, type C2	+	E	3	ES
NUCB1	nucleobindin 1	+	E	21	C
OLFML3	olfactomedin-like 3	+	E	3	ES
ORM1	orosomucoid 1	+	E	4	ES
PDIA3*	protein disulfide isomerase family A, member 3	+	E	10	C
PLA2G7	phospholipase A2, group VII	+	E	2	ES
PLG	plasminogen	+	E	10	ES
PLOD3	procollagen-lysine, 2-oxoglutarate 5-dioxygenase 3	+	E	10	C
PROS1	protein S (alpha)	+	E	12	ES
PTPRF	protein tyrosine phosphatase, receptor type, F	+	E	10	PM
RAB1A	RAB1A, member RAS oncogene family	+	E	1	C
RBP4	retinol binding protein 4, plasma	+	E	14	ES
RNASE4	ribonuclease, RNase A family, 4	+	E	9	ES
RRBP1	ribosome binding protein 1	+	E	51	C
SDC4	syndecan 4	+	E	2	PM
SDF4	stromal cell derived factor 4	+	E	4	C
SERPINA1	serpin peptidase inhibitor, clade A, member 1	+	E	29	ES
SERPINA4	serpin peptidase inhibitor, clade A, member 4	+	E	4	ES
SERPINA5	serpin peptidase inhibitor, clade A, member 5	+	E	15	ES

SERPINA6	serpin peptidase inhibitor, clade A, member 6	+	E	5	ES
SERPIND1	serpin peptidase inhibitor, clade D, member 1	+	E	12	ES
SERPINF1	serpin peptidase inhibitor, clade F, member 1	+	E	15	ES
SERPINF2	serpin peptidase inhibitor, clade F, member 2	+	E	12	ES
SERPINI1	serpin peptidase inhibitor, clade I, member 1	+	E	4	ES
SPINK1	serine peptidase inhibitor, Kazal type 1	+	E	7	ES
STC2	stanniocalcin 2	+	E	5	ES
TF	transferrin	+	E	0	ES
THBS1	thrombospondin 1	+	E	2	ES
VTN	vitronectin	+	E	16	ES
AHSG	Alpha-2-HS-glycoprotein	+	E	12	ES
CFB	Complement factor B	+	E	8	ES
A2M	Alpha-2-macroglobulin	+	E	48	ES
BIGH3	Kerato-epithelin	+	E	14	ES

Abbreviations: Cytoplasm, C; Plasma membrane, PM; Extracellular space, ES; Nucleus, N; Not annotated, N/A.

Appendix 5. Clustering of co-immunoprecipited secretome proteins that significantly changed between any two of EV, PCSK9 WT and its variants

Gene Names	Protein Names	Cluster	ANOVA p value
PLOD2	procollagen-lysine, 2-oxoglutarate 5-dioxygenase 2	A	1.7E-04
CCT6A	chaperonin containing TCP1, subunit 6A (zeta 1)	A	2.2E-04
UBE2N	ubiquitin-conjugating enzyme E2N	A	8.1E-04
MGAT5	C-1-tetrahydrofolate synthase	A	8.8E-04
ST6GAL1	ST6 beta-galactosamide alpha-2,6-sialyltranferase 1	A	3.2E-03
B4GALT4	Beta-1,4-galactosyltransferase 4	A	4.1E-03
EIF5A	eukaryotic translation initiation factor 5A	A	4.5E-03
KRT18	keratin 18	A	4.9E-03
MDH1	malate dehydrogenase 1, NAD (soluble)	A	6.8E-03
GYG1	glycogenin 1	A	7.2E-03
EEF1D	Antigen NY-CO-4	A	8.2E-03
TOMM70A	Antigen NY-CO-4	A	8.4E-03
API5	apoptosis inhibitor 5	A	1.2E-02
CKB	creatine kinase, brain	A	1.6E-02
PRDX1	peroxiredoxin 1	A	1.6E-02
NSFL1C	NSFL1 (p97) cofactor (p47)	A	1.8E-02
ALDOA	aldolase A, fructose-bisphosphate	A	2.1E-02
SAE1	SUMO1 activating enzyme subunit 1	A	2.8E-02
PTPN11	protein tyrosine phosphatase, non-receptor type 11	A	3.3E-02
SERBP1	SERPINE1 mRNA binding protein 1	A	3.5E-02
APOE	apolipoprotein E	A	2.5E-06
FN1	fibronectin 1	A	4.7E-05
FGG	fibrinogen gamma chain	A	2.5E-04
CFH	complement factor H	A	3.8E-04
NTS	neurotensin	A	9.0E-04
SERPINF1	serpin peptidase inhibitor, clade F member 1	A	1.4E-03
GPI	glucose-6-phosphate isomerase	A	1.0E-02
SERPINH1	serpin peptidase inhibitor, clade H member 1	A	2.3E-02
RBM4B	RNA binding motif protein 4B	A	9.0E-04
SFPQ	splicing factor proline/glutamine-rich	A	2.9E-03
HNRNPH3	heterogeneous nuclear ribonucleoprotein H3 (2H9)	A	3.5E-03
PTBP1	polypyrimidine tract binding protein 1	A	6.0E-03
DDX23	DEAD (Asp-Glu-Ala-Asp) box polypeptide 23	A	6.5E-03
SF3A3	splicing factor 3a, subunit 3, 60kDa	A	1.1E-02
LBX2	ladybird homeobox 2	A	1.9E-02
FKBP4	FK506 binding protein 4, 59kDa	A	2.0E-02
DNAJB1	DnaJ (Hsp40) homolog, subfamily B, member 1	A	2.3E-02

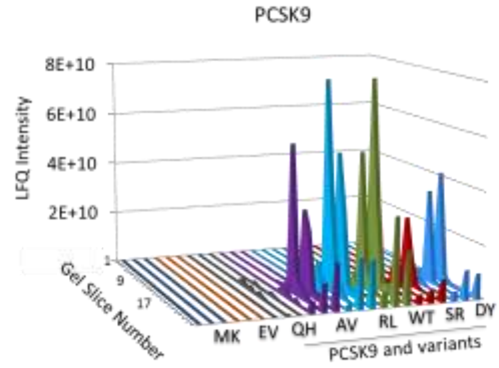
NCL	nucleolin	A	3.3E-02
PCBP2	poly(rC) binding protein 2	A	3.3E-02
PCBP1	poly(rC) binding protein 1	A	3.6E-02
SRSF7	serine/arginine-rich splicing factor 7	A	4.2E-02
HNRNPAB	heterogeneous nuclear ribonucleoprotein A/B	A	4.4E-02
GPS1	G protein pathway suppressor 1	A	1.8E-02
TLN1	talin 1	A	1.7E-02
NAP1L4	Exportin-7	A	7.3E-03
HNRNPD	heterogeneous nuclear ribonucleoprotein D	A	4.4E-02
CTNNA1	catenin (cadherin-associated protein), alpha 1, 102kDa	A	4.4E-02
XRN2	5'-3' exoribonuclease 2	B	3.7E-02
NCAPD2	non-SMC condensin I complex, subunit D2	B	3.7E-02
FASN	fatty acid synthase	C	1.9E-10
PSMD2	proteasome (prosome, macropain) 26S subunit, non-ATPase, 2	C	1.6E-08
IARS	isoleucyl-tRNA synthetase	C	7.4E-08
PSME4	proteasome (prosome, macropain) activator subunit 4	C	8.5E-07
COPA	coatomer protein complex, subunit alpha	C	9.1E-07
COPB1	coatomer protein complex, subunit beta 1	C	9.5E-07
AHCY	adenosylhomocysteinase	C	9.7E-07
ACACA	acetyl-CoA carboxylase alpha	C	9.7E-07
NADK2	NAD kinase 2, mitochondrial	C	1.1E-06
GMPPA	GDP-mannose pyrophosphorylase A	C	1.2E-06
GANAB	glucosidase, alpha; neutral AB	C	1.9E-06
ACTR1A	ARP1 actin-related protein 1 homolog A, centractin alpha (yeast)	C	2.2E-06
MARS	methionyl-tRNA synthetase	C	2.2E-06
EIF3B	eukaryotic translation initiation factor 3, subunit B	C	2.8E-06
DYNC1H1	dynein, cytoplasmic 1, heavy chain 1	C	3.9E-06
LARS	leucyl-tRNA synthetase	C	6.2E-06
ACTG1	actin, gamma 1	C	6.2E-06
IKBKAP	Elongator complex protein 1	C	6.9E-06
GCN1L1	GCN1 general control of amino-acid synthesis 1-like 1 (yeast)	C	6.9E-06
GALK1	galactokinase 1	C	1.4E-05
EPRS	glutamyl-prolyl-tRNA synthetase	C	1.5E-05
COPG1	coatomer protein complex, subunit gamma 1	C	3.8E-05
ACO1	aconitase 1, soluble	C	4.1E-05
RTCB	RNA 2',3'-cyclic phosphate and 5'-OH ligase	C	4.7E-05
COPB2	coatomer protein complex, subunit beta 2 (beta prime)	C	5.7E-05
EEF2	eukaryotic translation elongation factor 2	C	9.0E-05
PCYOX1	prenylcysteine oxidase 1	C	1.2E-04
GNB2L1	Elongator complex protein 1	C	1.3E-04
DYNC1LI1	dynein, cytoplasmic 1, light intermediate chain 1	C	1.6E-04
UBA1	ubiquitin-like modifier activating enzyme 1	C	1.7E-04
RPSA	ribosomal protein SA	C	1.7E-04

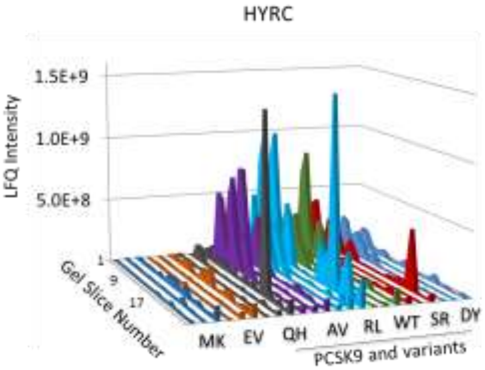
HTT	huntingtin	C	1.8E-04
CLUH	clustered mitochondria (cluA/CLU1) homolog	C	1.9E-04
PFAS	phosphoribosylformylglycinamidine synthase	C	2.2E-04
EIF3I	eukaryotic translation initiation factor 3, subunit I	C	2.5E-04
DARS	aspartyl-tRNA synthetase	C	3.0E-04
RPL5	ribosomal protein L5	C	3.2E-04
PPAT	phosphoribosyl pyrophosphate amidotransferase	C	3.8E-04
CS	citrate synthase	C	4.2E-04
CAD	Aspartate carbamoyltransferase	C	4.2E-04
MTHFD1	C-1-tetrahydrofolate synthase	C	5.2E-04
RPS18	ribosomal protein S18	C	7.4E-04
KIAA0368	KIAA0368	C	7.9E-04
ARPC1B	actin related protein 2/3 complex, subunit 1B, 41kDa	C	7.9E-04
QDPR	quinoid dihydropteridine reductase	C	8.3E-04
PSMD3	proteasome (prosome, macropain) 26S subunit, non-ATPase, 3	C	9.7E-04
SARS	seryl-tRNA synthetase	C	9.7E-04
COASY	CoA synthase	C	1.1E-03
PSMD1	proteasome (prosome, macropain) 26S subunit, non-ATPase, 1	C	1.6E-03
SEPT9	septin 9	C	1.7E-03
PSMD6	proteasome (prosome, macropain) 26S subunit, non-ATPase, 6	C	2.4E-03
NANS	N-acetylneuraminic acid synthase	C	2.4E-03
PYGB	phosphorylase, glycogen; brain	C	2.4E-03
MYH9	myosin, heavy chain 9, non-muscle	C	2.4E-03
EIF3A	eukaryotic translation initiation factor 3, subunit A	C	2.5E-03
KARS	lysyl-tRNA synthetase	C	3.0E-03
NQO1	NAD(P)H dehydrogenase, quinone 1	C	3.6E-03
HMGCS1	3-hydroxy-3-methylglutaryl-CoA synthase 1 (soluble)	C	3.6E-03
PYGL	phosphorylase, glycogen, liver	C	3.9E-03
RPS16	ribosomal protein S16	C	4.1E-03
TNPO3	transportin 3	C	6.5E-03
EIF2S1	eukaryotic translation initiation factor 2, subunit 1 alpha, 35kDa	C	6.8E-03
RPS3	ribosomal protein S3	C	6.8E-03
HECTD3	HECT domain containing E3 ubiquitin protein ligase 3	C	7.1E-03
AP2A1	adaptor-related protein complex 2, alpha 1 subunit	C	7.2E-03
RPS11	ribosomal protein S11	C	7.2E-03
TBCD	tubulin folding cofactor D	C	8.4E-03
IGF2BP1	insulin-like growth factor 2 mRNA binding protein 1	C	9.4E-03
DCTN1	dynactin 1	C	9.5E-03
GLDC	glycine dehydrogenase (decarboxylating)	C	1.0E-02
MON2	MON2 homolog (S. cerevisiae)	C	1.2E-02
UBE4B	ubiquitination factor E4B	C	1.3E-02
OAT	ornithine aminotransferase	C	1.3E-02
EIF4A1	eukaryotic translation initiation factor 4A1	C	1.3E-02

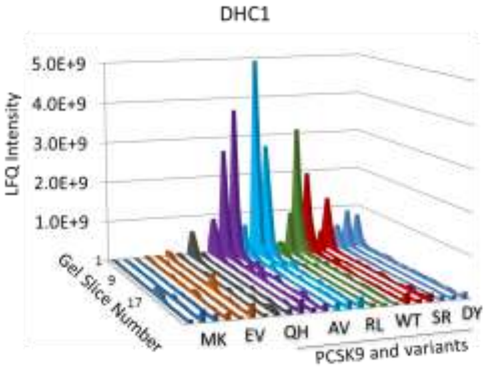
HYOU1	hypoxia up-regulated 1	C	1.4E-02
TUBB6	tubulin, beta 6 class V	C	1.5E-02
PC	pyruvate carboxylase	C	1.8E-02
MPST	mercaptopyruvate sulfurtransferase	C	1.9E-02
MAPK3	mitogen-activated protein kinase 3	C	2.0E-02
FARSA	phenylalanyl-tRNA synthetase, alpha subunit	C	2.9E-02
MTTP	microsomal triglyceride transfer protein	C	2.9E-02
NAGK	N-acetylglucosamine kinase	C	4.4E-02
LIN28B	lin-28 homolog B (C. elegans)	C	4.7E-02
PCSK9	proprotein convertase subtilisin/kexin type 9	C	8.3E-07
PCOLCE2	procollagen C-endopeptidase enhancer 2	C	5.0E-04
VTN	vitronectin	C	8.5E-03
NAA15	N(alpha)-acetyltransferase 15, NatA auxiliary subunit	C	7.2E-07
SNRNP200	small nuclear ribonucleoprotein 200kDa (U5)	C	1.1E-06
SND1	staphylococcal nuclease and tudor domain containing 1	C	3.9E-06
PSMC2	proteasome (prosome, macropain) 26S subunit, ATPase, 2	C	4.2E-06
EIF4A3	eukaryotic translation initiation factor 4A3	C	4.3E-06
CPSF1	cleavage and polyadenylation specific factor 1, 160kDa	C	6.2E-06
SKIV2L	superkiller viralicidic activity 2-like (S. cerevisiae)	C	6.9E-06
DHX9	DEAH (Asp-Glu-Ala-His) box helicase 9	C	1.4E-05
SRRT	serrate, RNA effector molecule	C	2.1E-05
HDAC2	histone deacetylase 2	C	4.1E-05
BUB3	BUB3 mitotic checkpoint protein	C	5.4E-05
MCM4	minichromosome maintenance complex component 4	C	8.7E-05
DDX19A	DEAD (Asp-Glu-Ala-Asp) box polypeptide 19A	C	1.0E-04
RUVBL1	RuvB-like AAA ATPase 1	C	1.5E-04
RAN	RAN, member RAS oncogene family	C	1.5E-04
XPO1	exportin 1	C	1.6E-04
PSMC1	proteasome (prosome, macropain) 26S subunit, ATPase, 1	C	1.6E-04
RRM1	ribonucleotide reductase M1	C	1.7E-04
MSH6	mutS homolog 6	C	1.7E-04
FEN1	flap structure-specific endonuclease 1	C	2.5E-04
PRKDC	protein kinase, DNA-activated, catalytic polypeptide	C	3.2E-04
HDAC6	histone deacetylase 6	C	6.1E-04
XPOT	exportin, tRNA	C	9.7E-04
HNRNPM	heterogeneous nuclear ribonucleoprotein M	C	9.7E-04
CTPS1	CTP synthase 1	C	9.7E-04
DHX15	DEAH (Asp-Glu-Ala-His) box helicase 15	C	1.0E-03
SMC4	structural maintenance of chromosomes 4	C	1.1E-03
MATR3	matrin 3	C	1.3E-03
MCM6	minichromosome maintenance complex component 6	C	1.5E-03
NSUN2	NOP2/Sun RNA methyltransferase family, member 2	C	1.6E-03
UBR4	ubiquitin protein ligase E3 component n-recognin 4	C	1.8E-03

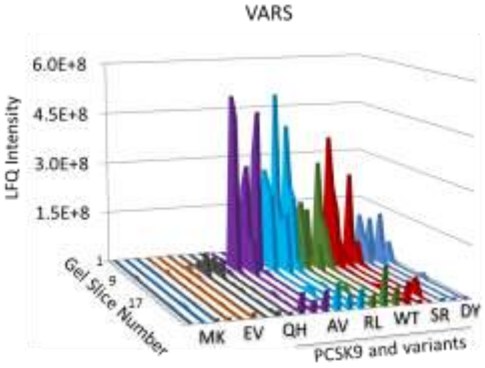
SMCHD1	structural maintenance of chromosomes flexible hinge domain containing 1	C	2.1E-03
PPP6C	protein phosphatase 6, catalytic subunit	C	2.4E-03
SMC2	structural maintenance of chromosomes 2	C	2.5E-03
XPO5	exportin 5	C	4.8E-03
MYBBP1A	MYB binding protein (P160) 1a	C	5.2E-03
TARDBP	TAR DNA binding protein	C	5.3E-03
PSMC6	proteasome (prosome, macropain) 26S subunit, ATPase, 6	C	6.5E-03
EFTUD2	elongation factor Tu GTP binding domain containing 2	C	6.5E-03
NONO	non-POU domain containing, octamer-binding	C	7.3E-03
CDK1	cyclin-dependent kinase 1	C	7.3E-03
DIS3	DIS3 exosome endoribonuclease and 3'-5' exoribonuclease	C	7.3E-03
KNTC1	kinetochore associated 1	C	1.0E-02
IPO8	importin 8	C	1.3E-02
PRPF8	pre-mRNA processing factor 8	C	1.6E-02
STAT1	signal transducer and activator of transcription 1, 91kDa	C	1.7E-02
SRP68	signal recognition particle 68kDa	C	1.8E-02
UPF1	UPF1 regulator of nonsense transcripts homolog (yeast)	C	1.9E-02
MSH2	mutS homolog 2	C	1.9E-02
DNAJA2	DnaJ (Hsp40) homolog, subfamily A, member 2	C	2.0E-02
PSMC5	proteasome (prosome, macropain) 26S subunit, ATPase, 5	C	2.1E-02
POLR2B	polymerase (RNA) II (DNA directed) polypeptide B, 140kDa	C	2.4E-02
AQR	aquarius intron-binding spliceosomal factor	C	3.0E-02
POLR2A	polymerase (RNA) II (DNA directed) polypeptide A, 220kDa	C	3.9E-02
FAM115A	family with sequence similarity 115, member A	C	1.6E-04
ZC3HAV1L	zinc finger CCCH-type, antiviral 1-like	C	3.0E-03
C12orf10	chromosome 12 open reading frame 10	C	6.1E-03
ATP6V1A	ATPase, H+ transporting, lysosomal 70kDa, V1 subunit A	C	6.9E-05
CLTC	clathrin, heavy chain (Hc)	C	5.0E-04
STRAP	serine/threonine kinase receptor associated protein	C	6.4E-04
GNB1	guanine nucleotide binding protein (G protein), beta polypeptide 1	C	7.2E-04
IGF2R	insulin-like growth factor 2 receptor	C	8.9E-04
GNAS	GNAS complex locus	C	5.1E-03
USP9X	ubiquitin specific peptidase 9, X-linked	C	6.5E-03
VARS	Valyl-tRNA synthetase	C	9.7E-07
ACLY	Valyl-tRNA synthetase	C	4.8E-05
BM28	ATP-citrate (pro-S-)-lyase	C	7.9E-04
KIAA0251	DNA replication licensing factor MCM2	C	2.4E-03
AOF2	Pyridoxal-dependent decarboxylase domain-containing protein 1	C	3.4E-03
KIAA0745	BRAF35-HDAC complex protein BHC110	C	2.3E-02
HSPA2	Nucleosome assembly protein 1-like 4b	C	4.6E-02
SERPINA5	serpin peptidase inhibitor, clade A member 5	C	2.4E-02
TRIM24	tripartite motif containing 24	C	4.7E-02

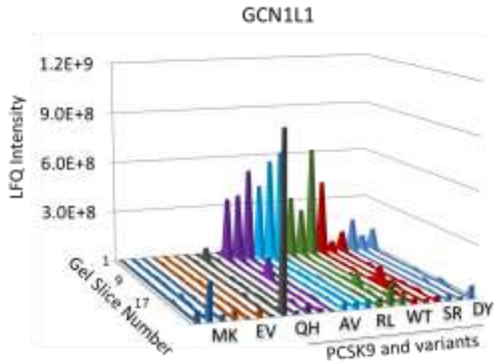
Appendix 6. LFQ intensity histograms of the top 20 potential PCSK9 interactors with greatest numbers of unique peptides and largest fold changes quantified by individual gel slices

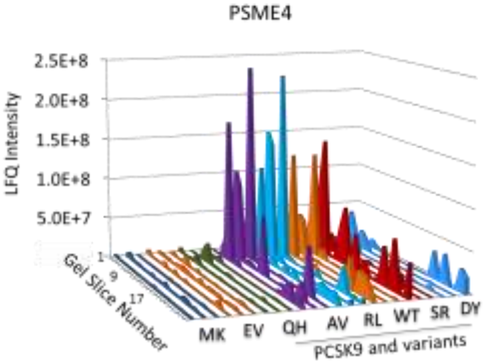


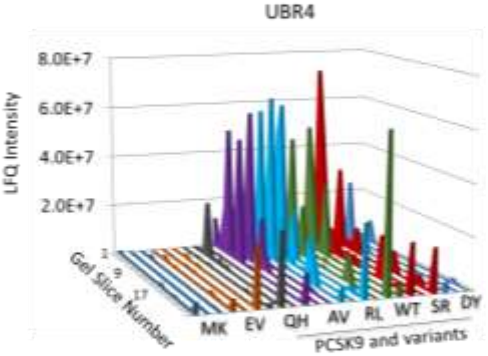


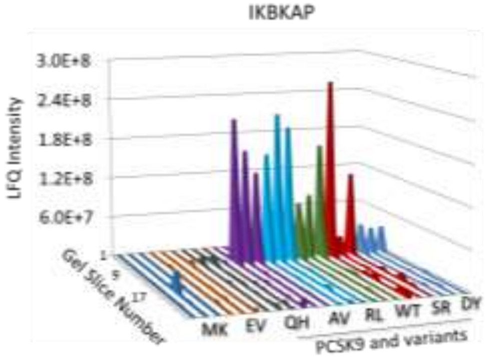


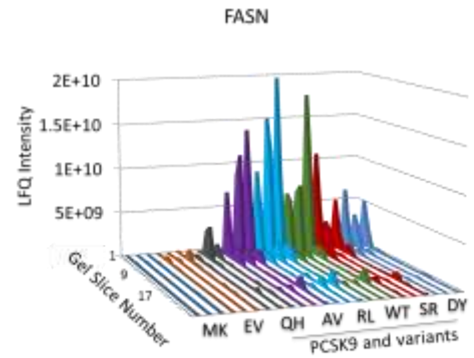







UBR4
LFQ Intensity
8.0E+7
6.0E+7
4.0E+7
2.0E+7
1
9
17
Gel Slice Number
MK EV QH AV RL WT SR DY
PCSK9 and variants


IKBKAP
LFQ Intensity
3.0E+8
2.4E+8
1.8E+8
1.2E+8
6.0E+7
1
9
17
Gel Slice Number
MK EV QH AV RL WT SR DY
PCSK9 and variants


FASN
LFQ Intensity
2E+10
1.5E+10
1E+10
5E+09
1
9
17
Gel Slice Number
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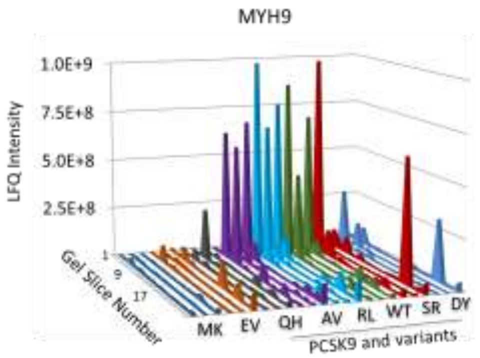
PCSK9 and variants


MYH9
LFQ Intensity
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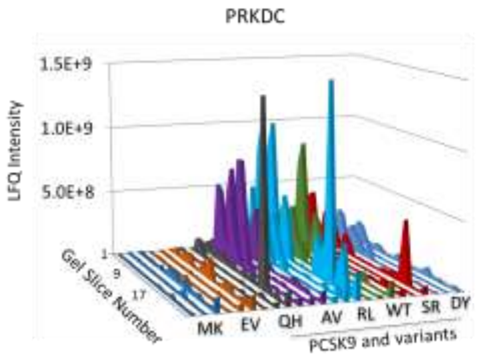
7.5E+8
5.0E+8
2.5E+8
1
9
17
Gel Slice Number
MK EV QH AV RL WT SR DY
PCSK9 and variants


PRKDC
LFQ Intensity
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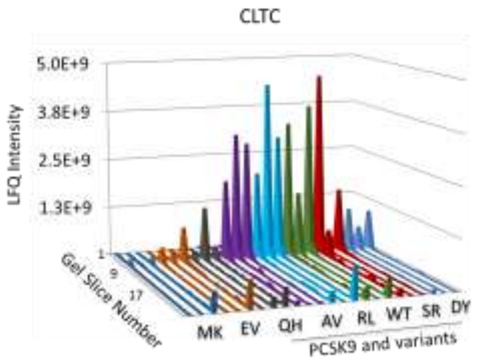
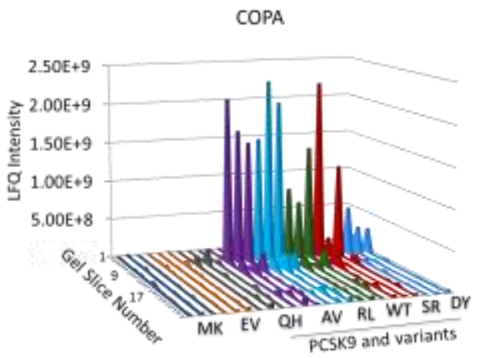
1.0E+9
5.0E+8
1
9
17
Gel Slice Number
MK EV QH AV RL WT SR DY
PCSK9 and variants


CLTC
LFQ Intensity
5.0E+9
3.8E+9
2.5E+9
1.3E+9
1
9
17
Gel Slice Number
MK EV QH AV RL WT SR DY
PCSK9 and variants


COPA
LFQ Intensity
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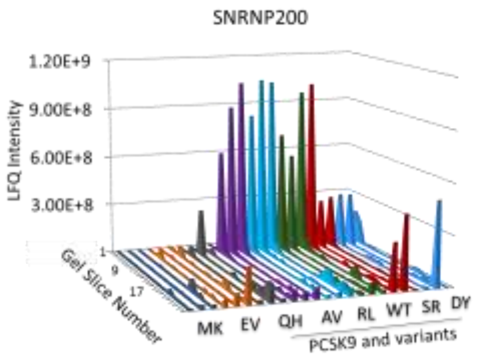
2.00E+9
1.50E+9
1.00E+9
5.00E+8
1
9
17
Gel Slice Number
MK EV QH AV RL WT SR DY
PCSK9 and variants


SNRNP200
LFQ Intensity
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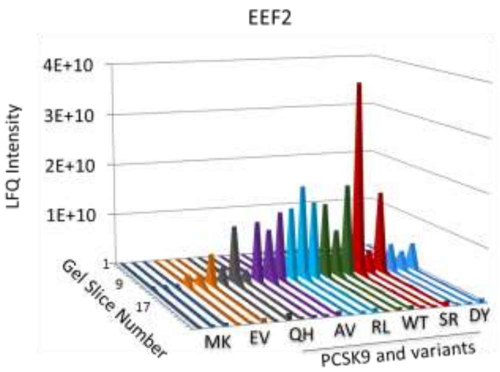
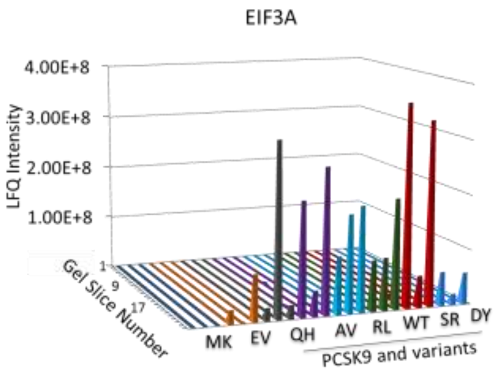
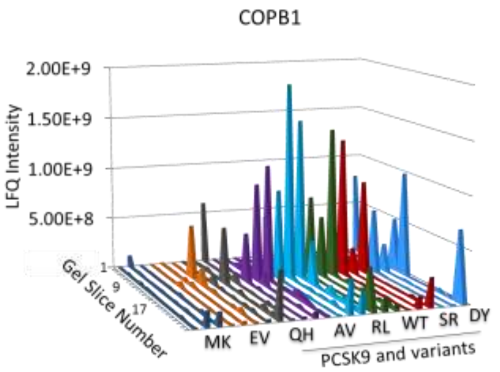
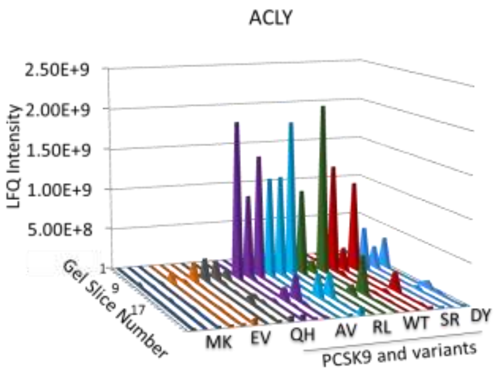
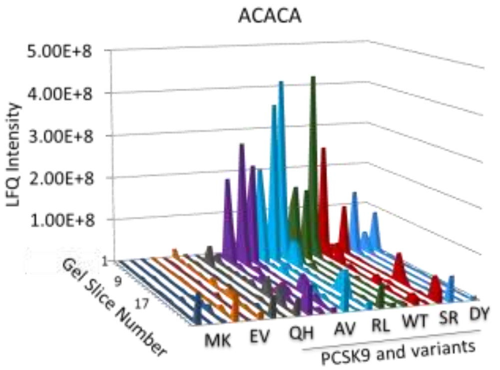
9.00E+8
6.00E+8
3.00E+8
1
9
17
Gel Slice Number
MK EV QH AV RL WT SR DY
PCSK9 and variants

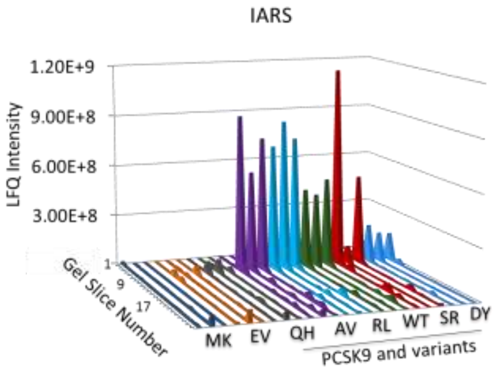

EEF2


EIF3A


COPB1


ACLY


ACACA


IARS

Abbreviations:

PCSK9	proprotein convertase subtilisin/kexin type 9
HYRC	DNA-dependent protein kinase catalytic subunit
DHC1	Cytoplasmic dynein 1 heavy chain 1
VARS	Valine--tRNA ligase
GCN1L1	GCN1 general control of amino-acid synthesis 1-like 1
PSME4	proteasome (prosome, macropain) activator subunit 4
UBR4	ubiquitin protein ligase E3 component n-recognin 4
IKBKAP	Elongator complex protein 1
FASN	fatty acid synthase
MYH9	myosin, heavy chain 9, non-muscle

PRKDC	protein kinase, DNA-activated, catalytic polypeptide
CLTC	clathrin, heavy chain
COPA	coatomer protein complex, subunit alpha
SNRNP200	small nuclear ribonucleoprotein 200kDa
EEF2	eukaryotic translation elongation factor 2
EIF3A	eukaryotic translation initiation factor 3, subunit A
COPB1	coatomer protein complex, subunit beta 1
ACLY	Valyl-tRNA synthetase
ACACA	acetyl-CoA carboxylase alpha
IARS	isoleucyl-tRNA synthetase

Contributions of Collaborators

In this study, the work in its entirety was performed by myself, Ge Chu, with the exception of the following: the site directed mutagenesis of V5-tagged PCSK9 variants were generated and sequence confirmed by Dr. Janice Mayne; in the study on protein subcellular distribution analysis section, the algorithms for normalization of MS/MS SILAC 'spike in' data and for the generation of the $\log_2$ L/H ratio curves were written and produced by Dr. Zhibin Ning; Cells stably transfected with wild type PCSK9-V5, variants D374Y-V5 and Q152H-V5 and empty vector were generated by Dr. Alexandre Noah; In the proteomic analyses and identification of the secreted interactome of PCSK9 and its variants section, the computing script for ANOVA and *post hoc* tests was written and operated by Dr. Zhibin Ning.

Curriculum vitae

Ge Chu

QUALIFICATIONS

- Proactive interpersonal communication skills gained from work and volunteer experience
- Excellent multi-task handling, problem-defining and critical evaluating skills proved by research experience

EDUCATION

M.Sc. in Biochemistry, University of Ottawa 2013-present

B.Sc. in Biotechnology, East China Normal University, China 2007-2011

RESEARCH EXPERIENCE

Proteomics-based study on effects of protein PCSK9 and its variants

M.Sc. student in Daniel Figeys' lab, University of Ottawa 2013-present

- Identified and quantified protein subcellular distribution and secretome changes affected by PCSK9 variants overexpression in cell culture utilizing proteomics and bioinformatics tools
- Identified secreted PCSK9 interactor candidates by co-immunoprecipitation and mass spectrometry analysis

KEY SKILLS

MS related skills:

- Protein sample preparation for LC-MS analysis
- LC-MS measurement and quantification
- Statistical analysis and bioinformatics processes

Biochemistry related skills:

- Cell culture and transfection
- DNA amplification and quantification
- Subcellular fractionation by density gradient ultracentrifugation
- Immunoblotting and ELISA

PRESENTATIONS

- Seminar

Department Research Symposium	University of Ottawa	2014

- Posters

Ottawa Institute of Systems Biology Symposium	University of Ottawa	May 2015
Department Graduate Students Poster Day	University of Ottawa	2014
Ottawa Institute of Systems Biology Symposium	University of Ottawa	June 2013

WORK AND VOLUNTEER EXPERIENCE

Let's Talk Science Volunteer

University of Ottawa May 2013-Present

- Demonstrating experimental procedures for national youth lab tours
- Demonstrating Chemistry Magic Show for sponsor appreciation event

Teaching Assistant / Marker

University of Ottawa Sept.-Dec. 2013

- Corrected tests, midterms and final exams and proctored exams

Market Service Assistant

T&T Supermarket, Mississauga April 2012-Nov. 2013

SCHOLARSHIPS AND AWARDS

Admission Scholarship, University of Ottawa	2013-2015
Excellence Scholarship, East China Normal University	2008-2010
Honor Prize, Daxia Cup Essay Competition, East China Normal University	2010