**Supplementary Table S1:** List of 336 nsSNPs of *SLC30A8* gene in dbSNP database.

|  |  |
| --- | --- |
| Reference SNP ID | Amino acid change |
| rs13266634 | R325W |
| rs144023942 | S24R |
| rs757010863 | M261V |
| rs16889462 | R276P, R276L, R276Q |
| rs73317647 | R165C |
| rs79461372 | Q160K |
| rs115816825 | V165I |
| rs139423387 | I60T |
| rs139489847 | G296R |
| rs139999213 | P310L |
| rs140404252 | L74R |
| rs141202988 | A138G |
| rs141215536 | V208L |
| rs141730422 | H54R, H54P, H54L |
| rs141876609 | C37R |
| rs142407509 | W152R |
| rs143136328 | I237T |
| rs143592691 | E44D |
| rs145638764 | A19V |
| rs145677283 | R165H, R165L |
| rs148780595 | S76L |
| rs149524118 | A216T |
| rs150548337 | G14E |
| rs185043998 | I141S  I190S, I190T |
| rs189800066 | L74F |
| rs199875716 | A139S |
| rs200020886 | G277S |
| rs200572112 | A19T |
| rs200895741 | E17K |
| rs201490156 | Y18D, Y18H |
| rs201697165 | S182F |
| rs202179744 | C151W |
| rs267601738 | E44K |
| rs367555174 | S353F |
| rs367769429 | K241N |
| rs369783320 | D248N |
| rs370648372 | E2G |
| rs374485094 | V330F |
| rs374670777 | T294M |
| rs374796913 | Q227E |
| rs534016412 | Y18C |
| rs547844916 | Y20S, Y20C |
| rs548323063 | G237S |
| rs552729572 | A161T |
| rs558096588 | K16E |
| rs558954279 | S124L |
| rs568845797 | E240K |
| rs746248449 | T348A |
| rs746249658 | R138L, R138Q |
| rs746522664 | I83T |
| rs746834620 | D269H |
| rs748773265 | H154R |
| rs750274669 | N262K |
| rs751109462 | S307Y |
| rs751350884 | S182P |
| rs751920415 | F240S |
| rs752486387 | P367T |
| rs754766609 | R131W |
| rs755057304 | T7K, T7M |
| rs756658613 | E40Q |
| rs757937209 | C26R |
| rs758315809 | F134I |
| rs758804870 | A235T |
| rs758845299 | Y8C |
| rs759290516 | S292N |
| rs759603427 | L27F |
| rs759642504 | S230N |
| rs759922365 | S113N |
| rs760413117 | R283G |
| rs761422383 | W122C |
| rs761514247 | L22R |
| rs761849730 | C184Y |
| rs762384069 | H301N |
| rs762742030 | I179T |
| rs763719329 | H304P, H304L |
| rs763915644 | C319W |
| rs764048971 | G107S |
| rs764354742 | Y244H |
| rs765078317 | D311V |
| rs765852756 | I141N |
| rs765921976 | A286D |
| rs767229202 | N262H |
| rs767594541 | A185T |
| rs767831564 | K34E |
| rs768316866 | K157Q, K157E |
| rs769900402 | P10S |
| rs770224130 | I349F |
| rs770651092 | Q357H |
| rs771191320 | W136G |
| rs771330275 | V219E |
| rs772070239 | L107F |
| rs772589300 | R282Q |
| rs773052424 | A87T |
| rs773218920 | T272I |
| rs774398640 | D246N |
| rs774641324 | P249S |
| rs775528421 | T11K |
| rs775751286 | Q28R |
| rs776072348 | Q279K |
| rs776903420 | S230R |
| rs777483638 | G48R |
| rs778259024 | N11H |
| rs778566684 | A30T |
| rs778716564 | R82Q, R82L |
| rs779578683 | R150S |
| rs779833277 | K126N |
| rs780088312 | C114F |
| rs780519971 | P42S, P42T |
| rs780551100 | I349N |
| rs780969938 | I37F |
| rs781293119 | S222F |
| rs866568009 | E284K |
| rs866700996 | L303R |
| rs867450817 | F204L |
| rs867484704 | L241R |
| rs908272494 | Q308K |
| rs917503960 | C53Y |
| rs927398701 | R41I, R41T |
| rs952851284 | M351T |
| rs974907412 | Q227H |
| rs980364063 | E243V |
| rs984636211 | P38S |
| rs997107604 | S278G |
| rs999243796 | S275G |
| rs1006318663 | T144P |
| rs1035358113 | R215G |
| rs1038379467 | E158Q |
| rs1185587051 | C26Y |
| rs1204184432 | H88Y |
| rs1204842338 | I291T |
| rs1210960262 | S267P |
| rs1216878055 | F33L |
| rs1218396380 | I173T |
| rs1223417676 | P127T |
| rs1226967241 | C26W |
| rs1242547998 | Q263R |
| rs1245619076 | H106N, H106Y |
| rs1246625721 | K9Q |
| rs1251663641 | A212S |
| rs1255515664 | W306C |
| rs1261230269 | A15T |
| rs1264823419 | H137P |
| rs1271726667 | L111R |
| rs1275963745 | Q123E |
| rs1287896728 | C37Y |
| rs1300407122 | V101F |
| rs1300885615 | E140A |
| rs1304394544 | D170H |
| rs1305371427 | S234R |
| rs1309409022 | Y51N |
| rs1325443204 | L308Q |
| rs1328527565 | S8F |
| rs1332425828 | K219E |
| rs1353977131 | H44Y |
| rs1356637421 | C319Y |
| rs1362611064 | I305F |
| rs1364161992 | G49S |
| rs1368760427 | D311N |
| rs1376637053 | E12K |
| rs1381363440 | I305F |
| rs1391496326 | K13T |
| rs1397027939 | F218S |
| rs1419208998 | D307H |
| rs1430961885 | E12G |
| rs1437115060 | A268T |
| rs1441138511 | M261R, M261T |
| rs1448460756 | R39G |
| rs1459420311 | M295T |
| rs1461042881 | N189D |
| rs1466795972 | R331W |
| rs1473828063 | S222T |
| rs1474212116 | K30T |
| rs1481111745 | I254T |
| rs1482242249 | E115K |
| rs1485362192 | V159E |
| rs1489643976 | Y239D |
| rs1563642482 | V294F |
| rs1586585878 | S6R |
| rs1586594545 | I94T |
| rs1586608274 | Q227R |
| rs1586608352 | S234G |
| rs1586625024 | H296N |
| rs1586625087 | Q350H |
| rs1821312676 | N11K |
| rs1821928719 | Q36R |
| rs1821930036 | E44V, E44G |
| rs1821930500 | E46K |
| rs1821932547 | K9R |
| rs1821935420 | K22Q |
| rs1821935671 | W23R |
| rs1822264391 | G47R |
| rs1822266670 | D103V |
| rs1822267911 | L108S |
| rs1822268120 | L108F |
| rs1822269542 | F114S |
| rs1822272743 | K77R |
| rs1822273628 | K81T |
| rs1822277200 | A90T |
| rs1822572727 | P120S |
| rs1822801102 | R150G |
| rs1822805541 | L222R |
| rs1822806700 | S228R |
| rs1822809043 | L187H |
| rs1822809774 | I238F |
| rs1822899509 | M275T |
| rs1823361115 | Q263E |
| rs1823437679 | R283K |
| rs1823438098 | I285T |
| rs1823438953 | F342I |
| rs1823446709 | C319R |
| rs2129784757 | A288S |
| rs2129784848 | S290I |
| rs2130932989 | D12G |
| rs2130933122 | E23G |
| rs2130965851 | P42R |
| rs2130966408 | A77S |
| rs2130985404 | F70V |
| rs2130999340 | L146R |

**Supplementary Table S2:** List of 25 nsSNPs in SLC30A8 protein via ConSurf analysis.

|  |  |  |
| --- | --- | --- |
| Rs ID | Mutation | Conservation scale |
| rs73317647 | R165C | 9 |
| rs139489847 | G296R | 8 |
| rs140404252 | L74R | 9 |
| rs141730422 | H54R | 9 |
| rs142407509 | W152R | 9 |
| rs145677283 | R165H  R165L | 9 |
| rs149524118 | A216T | 9 |
| rs746249658 | R138Q | 9 |
| rs762384069 | H301N | 9 |
| rs765852756 | I141N | 8 |
| rs771191320 | W136G | 8 |
| rs771330275 | V219E | 8 |
| rs776903420 | S230R | 8 |
| rs780551100 | I349N | 8 |
| rs866700996 | L303R | 9 |
| rs1035358113 | R215G | 8 |
| rs1255515664 | W306C | 9 |
| rs1271726667 | L111R | **5** |
| rs1300885615 | E140A | 9 |
| rs1305371427 | S234R | 9 |
| rs1325443204 | L308Q | 9 |
| rs1586608274 | Q227R | 9 |
| rs1822266670 | D103V | 9 |
| rs1822805541 | L222R | **3** |
| rs2130999340 | L146R | **3** |

**Supplementary Table S3:** Prediction of Phosphorylation sites in wild type RASSF5 using NetPhos 3.1.

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| Netphos 3.1 | Wild Type | | | Netphos 3.1 | Mutant Residues | | |
| Serine (S) | **Position** | **Score** | **Kinase** | **Serine (S)** | **Position** | **Score** | **Kinase** |
| 47 S | 0.514 | cdc2 | 47 S | 0.514 | cdc2 |
| 55 S | 0.797 | unsp | 55 S | 0.797 | unsp |
| 55 S | 0.705 | PKC | 55 S | 0.705 | PKC |
| 97S | 0.804 | PKC | 97S | 0.804 | PKC |
| 97S | 0.648 | PKA | 97S | 0.648 | PKA |
| 97S | 0.535 | DNAPK | 97S | 0.535 | DNAPK |
| 117S | 0.679 | PKA | 117S | 0.679 | PKA |
| 120S | 0.541 | CKI | 120S | 0.541 | CKI |
| 124S | 0.803 | PKC | 124S | 0.803 | PKC |
| 124S | 0.693 | unsp | 124S | 0.693 | unsp |
| 124S | 0.509 | cdc2 | 124S | 0.509 | cdc2 |
| 125S | 0.702 | PKC | 125S | 0.702 | PKC |
| 125S | 0.555 | cdc2 | 125S | 0.555 | cdc2 |
| 129S | 0.994 | unsp | 129S | 0.994 | unsp |
| 129S | 0.628 | PKC | 129S | 0.628 | PKC |
| 183S | 0.509 | cdc2 | 183S | 0.509 | cdc2 |
| 213S | 0.865 | unsp | 213S | 0.865 | unsp |
| 213S | 0.850 | PKC | 213S | 0.850 | PKC |
| 228S | 0.512 | PKA | 228S | 0.512 | PKA |
| 230S | 0.525 | cdc2 | 230S | 0.525 | cdc2 |
| **307S** | **0.629** | **PKA** | **299S** | **0.619** | **PKC** |
| 307S | 0.529 | DNAPK | 307S | 0.636 | DNAPK |
| 307S | 0.524 | cdc2 | 307S | 0.504 | cdc2 |
| 316S | 0.613 | PKC | 316S | 0.613 | PKC |
| 316S | 0.526 | cdc2 | 316S | 0.503 | cdc2 |
| 327S | 0.976 | unsp | 327S | 0.976 | unsp |
| 327S | 0.543 | DNAPK | 327S | 0.543 | DNAPK |
| 327S | 0.529 | ATM | 327S | 0.529 | ATM |
| 339S | 0.967 | unsp | 339S | 0.967 | unsp |
| 339S | 0.605 | cdc2 | 339S | 0.605 | cdc2 |
| 341S | 0.714 | unsp | 341S | 0.714 | unsp |
| 346S | 0.593 | PKA | 346S | 0.647 | PKA |
| 353S | 0.997 | unsp | 353S | 0.998 | unsp |
| Threonine (T) | 60T | 0.753 | unsp | **Threonine (T)** | 60T | 0.753 | unsp |
| 60T | 0.591 | PKC | 60T | 0.591 | PKC |
| 133T | 0.671 | PKC | 133T | 0.756 | PKC |
| 193T | 0.733 | PKC | 193T | 0.733 | PKC |
| Tyrosine (Y) | 69 Y | 0.728 | unsp | **Tyrosine (Y)** | 69 Y | 0.728 | unsp |
| 171Y | 0.562 | EGFR | 171Y | 0.562 | EGFR |
| 244Y | 0.976 | unsp | 244Y | 0.976 | unsp |
| 244Y | 0.595 | EGFR | 244Y | 0.595 | EGFR |

**Supplementary Table S4:** Structural changes, mutations in conserved domains, and amino acid properties of the wild-type and mutant type amino acids from Project Hope.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Amino acid change | Structure | Mutation located in conserved domain | Conservation  (located near a highly conserved position) | Amino acid properties |
| R138Q |  | Yes | Yes | * There is a difference in charge between the wild-type and mutant amino acid. * The charge of the buried wild-type residue is lost by this mutation. * The wild-type and mutant amino acids differ in size. * The mutant residue is smaller than the wild-type residue. * The mutation will cause an empty space in the core of the protein. |
| I141N |  | Yes | Yes | * The wild-type and mutant amino acids differ in size. * The mutant residue is bigger than the wild-type residue. * The wild-type residue was buried in the core of the protein. The mutant residue is bigger and probably will not fit. * The hydrophobicity of the wild-type and mutant residue differs. * The mutation will cause loss of hydrophobic interactions in the core of the protein. |
| W136G |  | Yes | Yes | * The wild-type and mutant amino acids differ in size. * The mutant residue is smaller than the wild-type residue. * This will cause a possible loss of external interactions. * The hydrophobicity of the wild-type and mutant residue differs. * The mutation might cause loss of hydrophobic interactions with other molecules on the surface of the protein. |
| I349N |  | Yes | Yes | * The wild-type and mutant amino acids differ in size. * The mutant residue is bigger than the wild-type residue. * The wild-type residue was buried in the core of the protein. The mutant residue is bigger and probably will not fit. * The hydrophobicity of the wild-type and mutant residue differs. * The mutation will cause loss of hydrophobic interactions in the core of the protein. |
| L303R |  | Yes | Yes | * There is a difference in charge between the wild-type and mutant amino acid. * The mutant residue introduces a charge in a buried residue which can lead to protein folding problems. * The wild-type and mutant amino acids differ in size. * The mutant residue is bigger than the wild-type residue. * The wild-type residue was buried in the core of the protein. The mutant residue is bigger and probably will not fit. * The hydrophobicity of the wild-type and mutant residue differs. * The mutation will cause loss of hydrophobic interactions in the core of the protein. |
| E140A |  | Yes | Yes | * There is a difference in charge between the wild-type and mutant amino acid. * The charge of the buried wild-type residue is lost by this mutation. * The wild-type and mutant amino acids differ in size. * The mutant residue is smaller than the wild-type residue. * The mutation will cause an empty space in the core of the protein. * The hydrophobicity of the wild-type and mutant residue differs. * The mutation will cause loss of hydrogen bonds in the core of the protein and as a result disturb correct folding. |
| W306C |  | Yes | Yes | * The wild-type and mutant amino acids differ in size. * The mutant residue is smaller than the wild-type residue. * The mutation will cause an empty space in the core of the protein. |
| L308Q |  | Yes | Yes | * The wild-type and mutant amino acids differ in size. * The mutant residue is bigger than the wild-type residue. * The wild-type residue was buried in the core of the protein. The mutant residue is bigger and probably will not fit. * The hydrophobicity of the wild-type and mutant residue differs. * The mutation will cause loss of hydrophobic interactions in the core of the protein. |

**Supplementary Table S5:** Effects on AA changes on *SLC30A8* gene from Project Hope.

|  |  |  |
| --- | --- | --- |
| AA change | Structure | Domain |
| R138Q | The mutation could affect the local structure and as a consequence affect this binding site.  The wild-type residue forms a salt bridge with glutamic acid at position 276. The difference in charge will disturb the ionic interaction made by the original, wild-type residue. | The mutated residue is located in a domain that is important for the activity of the protein and in contact with another domain that is also important for the activity. The interaction between these domains could be disturbed by the mutation, which might affect the function of the protein. |
| I141N | The residue is located in a region annotated in the UniProt database as a transmembrane domain.  The mutant residue is bigger than the wild-type residue.  This size difference can affect the contacts with the lipid-membrane.  The wild-type residue is more hydrophobic than the mutant residue.  This differences in hydrophobicity can affect the hydrophobic interactions with the membrane lipids. | The mutated residue is located in a domain that is important for the activity of the protein and in contact with residues in another domain. It is possible that this interaction is important for the correct function of the protein. The mutation can affect this interaction and as such affect protein function. |
| W136G | The mutation introduces a glycine at this position. Glycines are very flexible and can disturb the required rigidity of the protein at this position. | The mutated residue is located in a domain that is important for the activity of the protein and in contact with residues in another domain. It is possible that this interaction is important for the correct function of the protein. The mutation can affect this interaction and as such affect protein function. |
| I349N | In the 3D-structure can be seen that the wild-type residue is located in its preferred secondary structure, a β-strand. The mutant residue prefers to be in another secondary structure; therefore, the local conformation will be slightly destabilized. | The mutated residue is located in a domain that is important for the activity of the protein and in contact with another domain that is also important for the activity. The interaction between these domains could be disturbed by the mutation, which might affect the function of the protein. |
| L303R | The wild-type residue is predicted (using the Reprof software) to be located in an α-helix.  The mutation converts the wild-type residue in a residue that does not prefer α-helices as secondary structure. | The mutated residue is located in a domain that is important for the activity of the protein and in contact with residues in another domain. It is possible that this interaction is important for the correct function of the protein. The mutation can affect this interaction and as such affect protein function. |
| E140A | The wild-type residue forms a hydrogen bond with serine at position 124.  The size difference between wild-type and mutant residue makes that the new residue is not in the correct position to make the same hydrogen bond as the original wild-type residue did.  The difference in hydrophobicity will affect hydrogen bond formation. | The mutated residue is located in a domain that is important for the activity of the protein and in contact with another domain that is also important for the activity. The interaction between these domains could be disturbed by the mutation, which might affect the function of the protein. |
| W306C | Each amino acid has its own specific size, charge, and hydrophobicity-value. The original wild-type residue and newly introduced mutant residue often differ in these properties.  The mutant residue is smaller than the wild-type residue. | The mutated residue is located in a domain that is important for the activity of the protein and in contact with another domain that is also important for the activity. The interaction between these domains could be disturbed by the mutation, which might affect the function of the protein. |
| L308Q | Each amino acid has its own specific size, charge, and hydrophobicity-value. The original wild-type residue and newly introduced mutant residue often differ in these properties.  The mutant residue is bigger than the wild-type residue.  The wild-type residue is more hydrophobic than the mutant residue. | The mutated residue is located in a domain that is important for the activity of the protein and in contact with another domain that is also important for the activity. The interaction between these domains could be disturbed by the mutation, which might affect the function of the protein. |

**Supplementary Table S6:** Functions of proteins connected with SLC30A8.

|  |  |  |
| --- | --- | --- |
| Serial no | Protein name | Protein function |
| 01 | Receptor-type tyrosine-protein phosphatase-like N (PTPRN) | * Plays a role in vesicle-mediated secretory processes. * Required for the accumulation of normal levels of insulin- containing vesicles and preventing their degradation. * Plays a role in insulin secretion in response to glucose stimuli. Required for normal accumulation of the neurotransmitter’s norepinephrine, dopamine and serotonin in the brain |
| 02 | Hematopoietically-expressed homeobox protein (HHEX) | * Recognizes the DNA sequence 5'-ATTAA-3'. * Transcriptional repressor. May play a role in hematopoietic differentiation. * Establishes anterior identity at two levels; acts early to enhance canonical WNT-signaling by repressing expression of TLE4 |
| 03 | Threonylcarbamoyladenosine tRNA methylthiotransferase (CDKAL1) | * Catalyzes the methylthiolation of N6- threonylcarbamoyladenosine (t (6)A), * leading to the formation of 2- methylthio-N6-threonylcarbamoyladenosine (ms(2)t(6)A) at position 37 in tRNAs that read codons beginning with adenine |
| 04 | Insulin A chain (INS) | * Insulin decreases blood glucose concentration. * It increases cell permeability to monosaccharides, amino acids and fatty acids. * It accelerates glycolysis, the pentose phosphate cycle, and glycogen synthesis in liver. |
| 05 | Transcription factor 7-like 2 (TCF7L2) | * Participates in the Wnt signaling pathway and modulates MYC expression by binding to its promoter in a sequence-specific manner. * Acts as repressor in the absence of CTNNB1, and as activator in its presence. * Activates transcription from promoters with several copies of the Tcf motif 5'-CCTTTGATC-3' in the presence of CTNNB1 |
| 06 | Glutamate decarboxylase 2 (GAD2) | * Catalyzes the production of GABA |
| 07 | Insulin-like growth factor 2 mRNA-binding protein 2 (IGF2BP2) | * RNA-binding factor that recruits target transcripts to cytoplasmic protein-RNA complexes (mRNPs). This transcript 'caging' into mRNPs allows mRNA transport and transient storage. * It also modulates the rate and location at which target transcripts encounter the translational apparatus and shields them from endonuclease attacks or microRNA-mediated degradation |
| 08 | ATP-sensitive inward rectifier potassium channel 11 (KCNJ11) | * This receptor is controlled by G proteins. * Inward rectifier potassium channels are characterized by a greater tendency to allow potassium to flow into the cell rather than out of it. |
| 09 | Alpha-ketoglutarate-dependent dioxygenase (FTO) | * RNA demethylase that mediates oxidative demethylation of different RNA species, such as mRNAs, tRNAs and snRNAs, and acts as a regulator of fat mass, adipogenesis and energy homeostasis. * N (6)- methyladenosine (m6A) demethylation by FTO affects mRNA expression and stability. |
| 10 | Kinesin-like protein (KIF11) | * Motor protein required for establishing a bipolar spindle during mitosis. Required in non-mitotic cells for transport of secretory proteins from the Golgi complex to the cell surface |