
Article

Proteomic Analysis in Valvular Cardiomyopathy: Aortic Regurgitation vs. Aortic Stenosis

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1. Supplementary Materials

1.1. Supplementary Figure

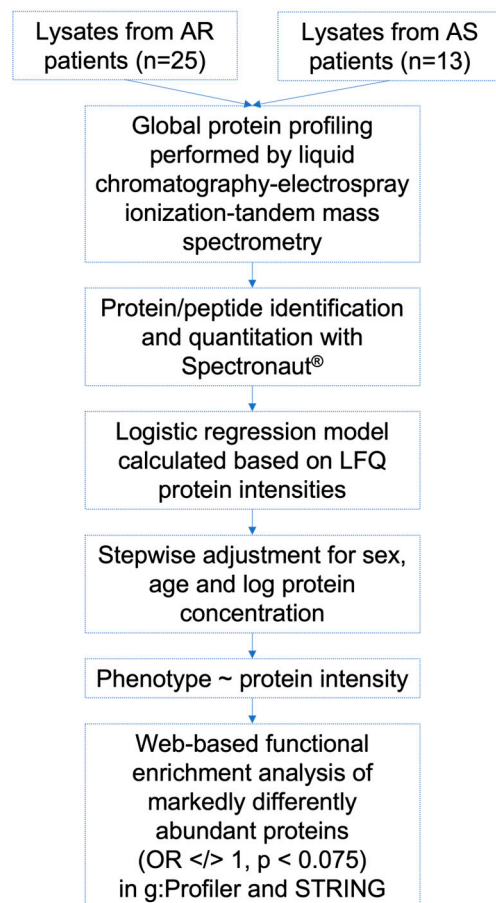


Figure S1. Flow chart for proteomic assessment of final sample set. AR: aortic regurgitation; AS: aortic stenosis; LFQ: label-free quantitation; OR: Odds ratio.

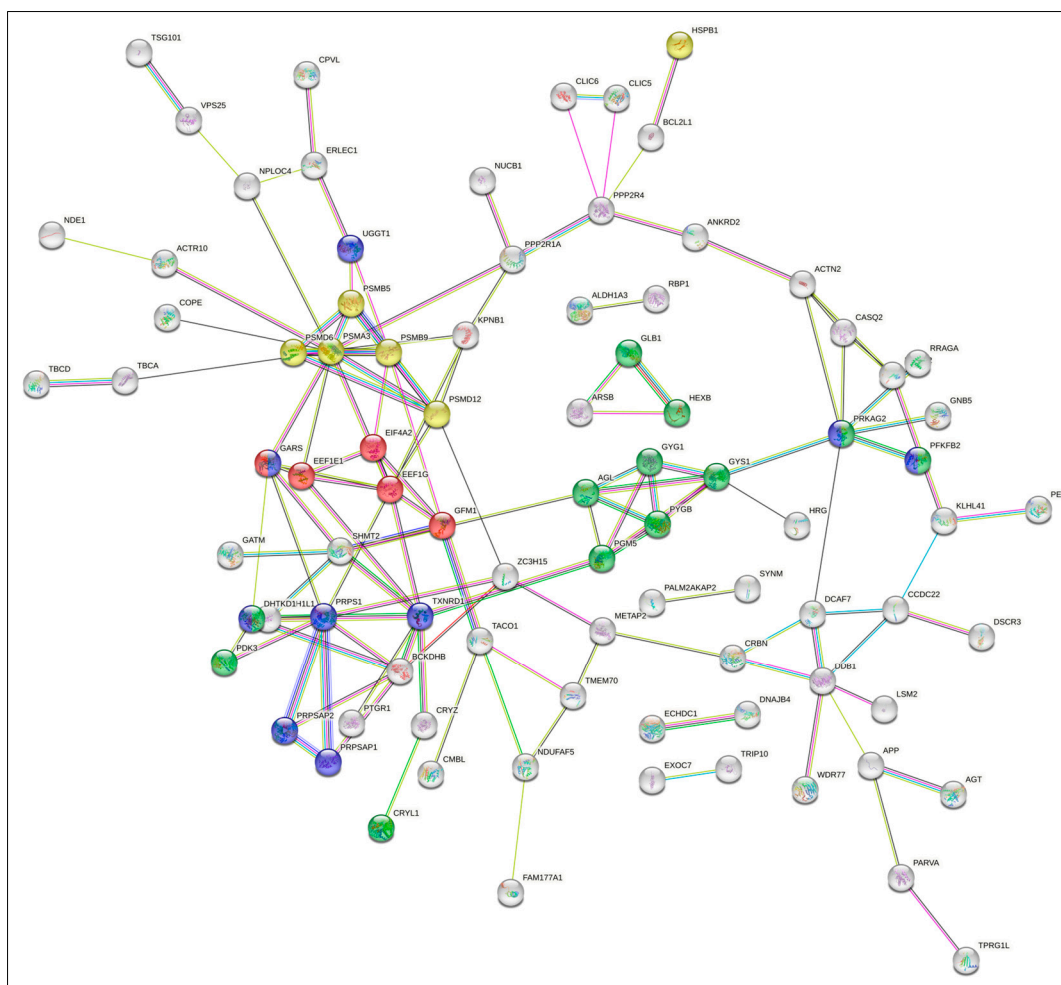


Figure S2. Network visualization of protein-protein interconnections of proteins being characteristic of the aortic regurgitation group using STRING: proteins assigned to carbohydrate metabolism (green), nucleotide metabolism (blue), protein biosynthesis (red), and the proteasome (yellow). Disconnected nodes in the network hidden.

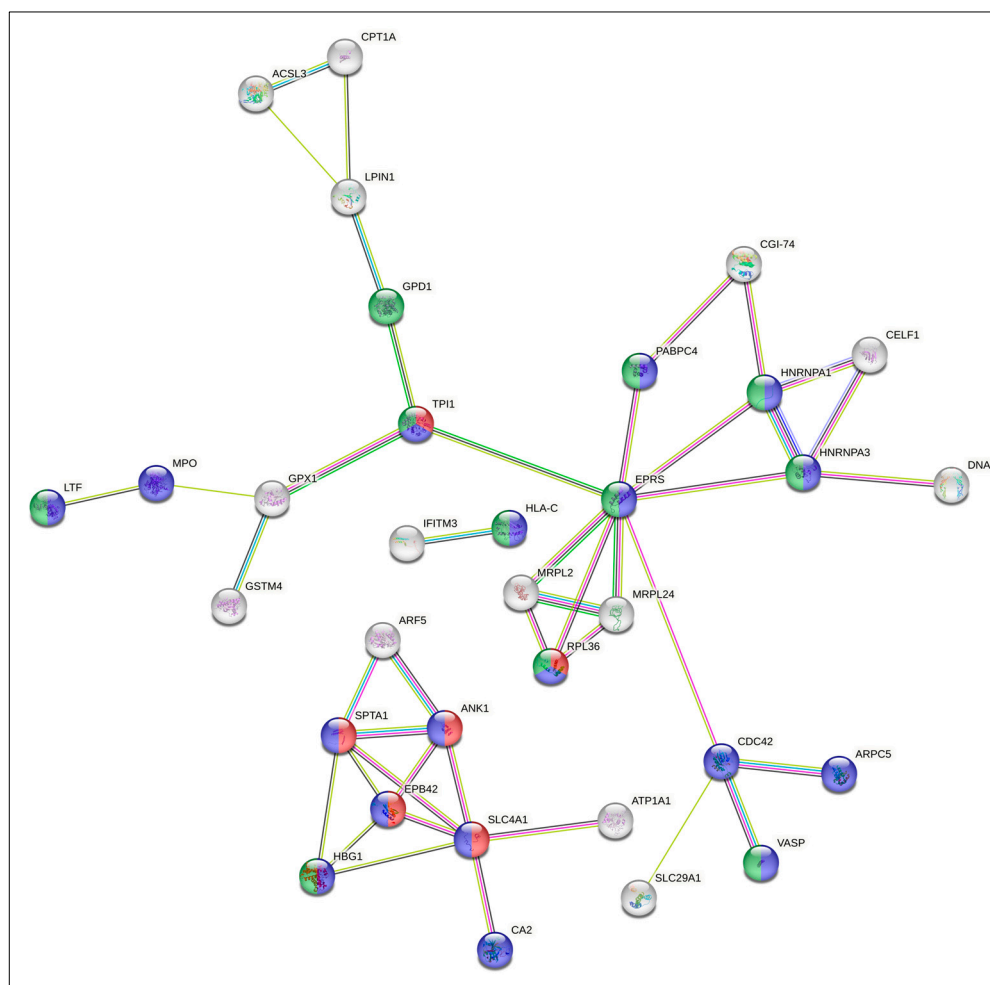


Figure S3. Network visualization of protein-protein interconnections of proteins being characteristic of the aortic stenosis using STRING: proteins assigned to the immune system (green) and hematopoietic system/angiogenesis (blue) and anemia (red) in specific. Disconnected nodes in the network hidden.

1.2. Supplementary Tables

Table S1_A. Liquid chromatography-electrospray ionization-tandem mass spectrometry parameter (data independent mode; quantitative data).

| Data independent analyses (DIA) | |
|--|---|
| reversed phase liquid chromatography | Ultimate 3000 RSLC (Thermo Scientific) |
| Trap column | 75 µm inner diameter, packed with 3 µm C18 particles (Acclaim PepMap100, Thermo Scientific) |
| Analytical column | 75 µm inner diameter, packed with 2.6 µm C18 particles (Accucore, 25 cm, Thermo Scientific) |
| Flow rate | 300 nl/min |
| Column oven temperature | 40°C |
| Buffer system | Binary buffer system consisting of 0.1% acetic acid in HPLC-grade water (buffer A) and 100% ACN in 0.1% acetic acid (buffer B) |
| Gradient | Gradient of buffer B: 2min 2% to 5 %, 8min 5% to 7%, 60min 7% to 25%, 5min 25 to 40%, 2 min 40% to 90%, 6 min 90%, 2 min 90% to 2%, 10 min 2% |
| Mass spectrometer | Exploris 480 |
| Operation mode | Data-independent |
| Electrospray | Nanospray Flex Ion Source |
| Full MS | |
| MS scan resolution | 120,000 |
| Normalized AGC target | 300 % |
| Maximum ion injection time for the MS scan | 60 ms |
| Scan range | 350 to 1200 <i>m/z</i> |
| Spectra data type | Profile |
| DIA scan | |
| Resolution | 30,000 |
| Normalized AGC target | 3000 |
| Maximum ion injection time for the MS/MS scans | Auto |
| Spectra data type | Profile |
| Selection for MS/MS | 1 |
| Isolation window | 65 windows <i>m/z</i> 13 (overlap 2) |
| Fixed first mass | 200 |
| Dissociation mode | Higher energy collisional dissociation (HCD) |
| Normalized collision energy | Stepped, 30 |
| Dissociation mode | HCD |

Table S1_B. Spectronaut parameters for peptide/protein identification and intensity extraction.

| Spectronaut |
|--------------------------|
| 16.0.220606.53000 |

| | |
|--|--|
| Computer Name: AGVOE-SPECTRONAUT | |
| User Domain Name: AGVOE-SPECTRONAUT | |
| User Name: spectronaut | |
| Analysis Mode: UI | |
| Analysis Type: directDIA | |
| Analysis Date: 05-August-2022 | |
| Settings Used: | |
| <u>Pulsar Search\Peptides</u> | |
| Toggle N-terminal M: | True |
| Min Peptide Length: | 7 |
| Max Peptide Length: | 52 |
| Missed Cleavages: | 2 |
| Digest Type: | Specific |
| Enzymes / Cleavage Rules: | Trypsin/P |
| <u>Pulsar Search\Labeling</u> | |
| Channels: | |
| Channel 1: | False |
| Channel 2: | False |
| Channel 3: | False |
| <u>DIA Analysis\Data Extraction</u> | |
| MS1 Mass Tolerance Strategy: | Dynamic |
| Correction Factor: | 1 |
| MS2 Mass Tolerance Strategy: | Dynamic |
| Correction Factor: | 1 |
| Intensity Extraction MS1: | Maximum Intensity |
| Intensity Extraction MS2: | Maximum Intensity |
| <u>DIA Analysis\XIC Extraction</u> | |
| XIC IM Extraction Window: | Dynamic |
| Correction Factor: | 1 |
| XIC RT Extraction Window: | Dynamic |
| Correction Factor: | 1 |
| <u>Pulsar Search\Modifications</u> | |
| Max Variable Modifications: | 5 |
| <u>Database</u> | |
| Original File: | uniprot_Human_2022_01.fasta |
| <u>Select Modifications:</u> | |
| Fixed Modifications:: | Carbamidomethyl (C) |
| Variable Modifications: : | Acetyl (Protein N-term), Oxidation (M) |
| <u>DIA Analysis\Calibration</u> | |
| MS1 Mass Tolerance Strategy: | System Default |

| | |
|---|---|
| MS2 Mass Tolerance Strategy: | System Default |
| Precision iRT: | True |
| iRT <-> RT Regression Type: | Local (Non-Linear) Regression |
| Exclude Deamidated Peptides: | True |
| MZ Extraction Strategy: | Maximum Intensity |
| Allow source specific iRT Calibration: | True |
| <u>DIA Analysis\Identification</u> | |
| Generate Decoys: | True |
| Decoy Limit Strategy: | Dynamic |
| Library Size Fraction: | 0.1 |
| Decoy Method: | Mutated |
| Preferred Fragment Source: | NN Predicted Fragments |
| Machine Learning: | Per Run |
| Exclude Duplicate Assays: | True |
| Precursor PEP Cutoff: | 0.2 |
| Protein Qvalue Cutoff (Experiment): | 0.01 |
| Protein Qvalue Cutoff (Run): | 0.05 |
| Exclude Single Hit Proteins: | False |
| Pvalue Estimator: | Kernel Density Estimator |
| Precursor Qvalue Cutoff: | 0.001 |
| Single Hit Definition: | By Stripped Sequence |
| <u>DIA Analysis\Quantification</u> | |
| Interference Correction: | True |
| MS1 Min: | 2 |
| MS2 Min: | 3 |
| Exclude All Multi-Channel Interferences: | True |
| Only Identified Peptides: | True |
| Protein LFQ Method: | Automatic |
| Major (Protein) Grouping: | by Protein Group Id |
| Minor (Peptide) Grouping: | by Stripped Sequence |
| Minor Group Top N: | False |
| Minor Group Quantity: | Sum precursor quantity |
| Major Group Top N: | True |
| Min: | 2 |
| Max: | 3 |
| Major Group Quantity: | Mean peptide quantity |
| Quantity MS-Level: | MS2 |
| Quantity Type: | Area |
| Proteotypicity Filter: | None (set to proteotypic by in-house analysis R pipeline) |
| Data Filtering: | Identified in % of Runs (Percentile) |
| Fraction: | 0.2 |
| Imputing Strategy: | No Imputing |
| Cross Run Normalization: | True (local) |

DIA Analysis\PTM Work-flow

| | |
|---------------------|-------|
| PTM Localization: | True |
| Probability Cutoff: | 0.75 |
| PTM Analysis: | True |
| Multiplicity: | True |
| Run Clustering: | False |
| PTM Consolidation: | Sum |
| Flanking Region: | 7 |

DIA Analysis\Workflow

| | |
|-----------------------------------|---------------------------------------|
| MS2 DeMultiplexing: | Automatic |
| Run Limit for directDIA Library: | -1 |
| Method Evaluation: | False |
| Profiling Strategy: | iRT Profiling |
| Profiling Row Selection: | Minimum Qvalue Row Selection |
| Qvalue Threshold: | 0.001 |
| Profiling Target Selection: | Profile only non-identified Precursor |
| Identification Criterion: | Qvalue |
| Threshold: | 0.001 |
| Carry-over exact Peak Boundaries: | False |
| Unify Peptide Peaks Strategy: | Select corresponding Peak |

DIA Analysis\Protein Inference

| | |
|-----------------------------|-----------|
| Protein Inference Workflow: | Automatic |
| Inference Algorithm: | IDPicker |

DIA Analysis\Post Analysis

| | |
|--------------------------------------|---|
| Calculate Sample Correlation Matrix: | True |
| Calculate Explained TIC: | Quick |
| Differential Abundance Grouping: | Major Group (Quantification Settings) |
| Smallest Quantitative Unit: | Precursor Ion (Quantification Settings) |
| Use All MS-Level Quantities: | False |
| Differential Abundance Testing: | t-test |
| Group-Wise Testing Correction: | False |
| Run Clustering: | True |
| Distance Metric: | Manhattan Distance |
| Linkage Strategy: | Ward's Method |
| Z-score transformation: | False |
| Order Runs by Clustering: | True |

DIA Analysis\Pipeline Mode

| | |
|------------------------|------|
| Post Analysis Reports: | |
| Scoring Histograms: | True |

| | |
|--|----------------------------|
| Data Completeness Bar Chart: | True |
| Run Identifications Bar Chart: | True |
| CV Density Line Chart: | True |
| CVs Below X Bar Chart: | True |
| Generate SNE File: | True |
| Store Iontraces in SNE: | False |
| Report Schema: | C_FunGene_complex (Normal) |
| Reporting Unit: | Across Experiment |
| <u>Pulsar Search\Identification</u> | |
| Peptide FDR: | 0.01 |
| Protein Group FDR: | 0.01 |
| PSM FDR: | 0.01 |
| <u>Pulsar Search\Tolerances</u> | |
| <u>Tolerance Parameters:</u> | |
| Thermo Orbitrap: | |
| Calibration Search: | Dynamic |
| MS1 Correction Factor: | 1 |
| MS2 Correction Factor: | 1 |
| Main Search: | Dynamic |
| MS1 Correction Factor: | 1 |
| MS2 Correction Factor: | 1 |
| <u>TOF:</u> | |
| Calibration Search: | Dynamic |
| MS1 Correction Factor: | 1 |
| MS2 Correction Factor: | 1 |
| Main Search: | Dynamic |
| MS1 Correction Factor: | 1 |
| MS2 Correction Factor: | 1 |
| <u>Thermo IonTrap:</u> | |
| Calibration Search: | Dynamic |
| MS1 Correction Factor: | 1 |
| MS2 Correction Factor: | 1 |
| Main Search: | Dynamic |
| MS1 Correction Factor: | 1 |
| MS2 Correction Factor: | 1 |
| <u>Pulsar Search\Workflow</u> | |
| Use DNN Predicted Ion Mobility: | Auto |
| Fragment Ion Selection Strategy: | Intensity Based |
| In-Silico Generate Missing Channels: | False |
| <u>Pulsar Search\Result Filters</u> | |
| <u>Precursors:</u> | |
| Best N Fragments per Peptide: | True |
| Min: | 6 |
| Max: | 10 |

| | |
|------------------------------------|-------|
| Channel Count: | False |
| Modifications: | None |
| Amino Acids: | False |
| Best N Peptides per Protein Group: | False |
| FASTA Matched: | False |
| Missed Cleavage: | False |
| Peptide Charge: | False |
| Proteotypicity: | False |
| <u>Fragment Ions:</u> | |
| m/z : | True |
| Min: | 300 |
| Max: | 1800 |
| Ion Charge: | False |
| Ion Loss Type: | False |
| Ion Type: | False |
| Ion AA Length: | True |
| N: | 3 |
| Relative Intensity: | True |
| Min: | 5 |

Table S2. Aortic regurgitation-associated proteins: OR > 1, $p < 0.075$. OR: Odds ratio, i.e. chance to be assigned to the aortic regurgitation group when protein intensity is high.

| Protein ID | Gene name | Protein name* | OR | p-value | 95% CI lower limit | 95% CI upper limit |
|------------|---------------|--|--------|---------|-----------------------|-----------------------|
| Q9UBV8 | PEF1 | Peflin | 160.23 | 0.028 | 1.75 | 14708.73 |
| Q9NVD7 | PARVA | Alpha-parvin | 63.56 | 0.052 | 0.97 | 4180.91 |
| P46976 | GYG1 | Glycogenin-1 | 27.43 | 0.020 | 1.67 | 449.35 |
| O60826 | CCDC22 | Coiled-coil domain-containing protein 22 | 26.31 | 0.032 | 1.32 | 525.42 |
| O43324 | EEF1E1 | Eukaryotic translation elongation factor 1 epsilon-1 | 25.35 | 0.019 | 1.71 | 375.80 |
| P30153 | PPP2R1A | Serine/threonine-protein phosphatase 2A 65 kDa regulatory subunit A alpha iso-form | 19.04 | 0.051 | 0.99 | 365.84 |
| P25686 | DNAJB2 | DnaJ homolog subfamily B member 2 | 17.56 | 0.043 | 1.10 | 281.40 |
| Q14232 | EIF2B1 | Translation initiation factor eIF-2B subunit alpha | 16.18 | 0.048 | 1.03 | 255.29 |
| Q14558 | PRPSAP1 | Phosphoribosyl pyrophosphate synthase-associated protein 1, | 15.50 | 0.017 | 1.64 | 146.19 |
| O15061 | SYNM | Synemin | 14.64 | 0.021 | 1.51 | 142.18 |
| Q15124 | PGM5 | Phosphoglucomutase-like protein 5 | 13.00 | 0.021 | 1.46 | 115.44 |
| Q9NZ32 | ACTR10 | Actin-related protein 10 | 12.82 | 0.012 | 1.77 | 93.02 |
| Q96IZ0 | PAWR | PRKC apoptosis WT1 regulator protein | 9.07 | 0.025 | 1.32 | 62.46 |
| O60825 | PFKFB2 | 6-phosphofructo-2-kinase/fructose-2,6-bisphosphatase 2 | 8.62 | 0.018 | 1.45 | 51.42 |
| P35573 | AGL | Glycogen debranching enzyme | 8.45 | 0.026 | 1.28 | 55.62 |
| P04196 | HRG | Histidine-rich glycoprotein | 8.26 | 0.031 | 1.22 | 56.02 |
| Q5T0D9 | TPRG1L | Tumor protein p63-regulated gene 1-like protein | 8.20 | 0.050 | 1.00 | 67.16 |
| Q16881 | TXNRD1 | Thioredoxin reductase 1, cytoplasmic | 7.54 | 0.054 | 0.96 | 59.04 |
| O75891 | ALDH1L1 | Cytosolic 10-formyltetrahydrofolate dehydrogenase | 7.53 | 0.024 | 1.30 | 43.66 |
| Q99766 | ATP5S/DMA C2L | ATP synthase subunit s, mitochondrial | 7.47 | 0.049 | 1.01 | 55.14 |

| | | | | | | |
|--------|---------|---|------|-------|------|-------|
| P26641 | EEF1G | Elongation factor 1-gamma | 7.44 | 0.027 | 1.26 | 44.00 |
| Q7Z4V5 | HDGFL2 | Hepatoma-derived growth factor-related protein 2 | 7.33 | 0.015 | 1.47 | 36.56 |
| P60891 | PRPS1 | Ribose-phosphate pyrophosphokinase 1 | 7.08 | 0.021 | 1.35 | 37.11 |
| Q14956 | GPNMB | Transmembrane glycoprotein NMB | 6.86 | 0.011 | 1.56 | 30.25 |
| P04792 | HSPB1 | Heat shock protein beta-1, HspB1 | 6.65 | 0.055 | 0.96 | 45.91 |
| O60256 | PRPSAP2 | Phosphoribosyl pyrophosphate synthase-associated protein 2 | 6.63 | 0.017 | 1.40 | 31.39 |
| P09455 | RBP1 | Retinol-binding protein 1 | 6.58 | 0.030 | 1.20 | 36.21 |
| P55809 | OXCT1 | Succinyl-CoA:3-ketoacid coenzyme A transferase 1, mitochondrial | 6.45 | 0.053 | 0.97 | 42.75 |
| Q07817 | BCL2L1 | Bcl-2-like protein 1 | 6.43 | 0.024 | 1.27 | 32.48 |
| Q02818 | NUCB1 | Nucleobindin-1 | 6.27 | 0.074 | 0.84 | 47.05 |
| P11216 | PYGB | Glycogen phosphorylase, brain form | 6.26 | 0.038 | 1.11 | 35.33 |
| Q96DZ1 | ERLEC1 | Endoplasmic reticulum lectin 1 | 6.15 | 0.074 | 0.84 | 45.01 |
| P01019 | AGT | Angiotensinogen | 5.90 | 0.028 | 1.21 | 28.72 |
| Q15120 | PDK3 | [Pyruvate dehydrogenase (acetyl-transferring)] kinase isozyme 3 | 5.73 | 0.041 | 1.07 | 30.66 |
| P28074 | PSMB5 | Proteasome subunit beta type-5 | 5.47 | 0.044 | 1.05 | 28.54 |
| Q16531 | DDB1 | DNA damage-binding protein 1 | 5.22 | 0.054 | 0.97 | 28.02 |
| P35609 | ACTN2 | Alpha-actinin-2 | 5.22 | 0.048 | 1.01 | 26.89 |
| Q9NYL2 | MAP3K20 | Mitogen-activated protein kinase kinase kinase 20 | 5.21 | 0.060 | 0.94 | 29.03 |
| Q9Y316 | MEMO1 | Protein MEMO1 | 5.11 | 0.019 | 1.30 | 20.05 |
| Q9NVT9 | ARMC1 | Armadillo repeat-containing protein 1 | 5.02 | 0.025 | 1.22 | 20.59 |
| Q6VY07 | PACS1 | Phosphofurin acidic cluster sorting protein 1 | 4.97 | 0.029 | 1.18 | 20.91 |
| Q9NYU2 | UGGT1 | UDP-glucose:glycoprotein glucosyltransferase 1 | 4.88 | 0.069 | 0.89 | 26.93 |
| P61962 | DCAF7 | DDB1- and CUL4-associated factor 7 | 4.86 | 0.044 | 1.04 | 22.74 |
| P28065 | PSMB9 | Proteasome subunit beta type-9 | 4.67 | 0.072 | 0.87 | 25.03 |
| Q96HY7 | DHTKD1 | 2-oxoadipate dehydrogenase complex component E1 | 4.58 | 0.045 | 1.04 | 20.21 |

| | | | | | | |
|--------|----------|---|------|-------|------|-------|
| Q9NXR1 | NDE1 | Nuclear distribution protein nudE homo- log 1 | 4.51 | 0.051 | 0.99 | 20.51 |
| Q8N8S7 | ENAH | Protein enabled homolog | 4.51 | 0.023 | 1.24 | 16.45 |
| P05067 | APP | Amyloid-beta precursor protein | 4.48 | 0.059 | 0.94 | 21.35 |
| P15848 | ARSB | Arylsulfatase B | 4.47 | 0.030 | 1.15 | 17.37 |
| Q9BRG1 | VPS25 | Vacuolar protein-sorting-associated pro- tein 25 | 4.46 | 0.046 | 1.02 | 19.45 |
| Q08257 | CRYZ | Quinone oxidoreductase | 4.45 | 0.023 | 1.23 | 16.04 |
| Q16658 | FSCN1 | Fascin | 4.45 | 0.038 | 1.09 | 18.21 |
| Q9H3G5 | CPVL | Probable serine carboxypeptidase CPVL | 4.44 | 0.033 | 1.12 | 17.54 |
| Q9HAB8 | PPCS | Phosphopantothenate--cysteine ligase, Phosphopantothenoylcysteine synthetase | 4.44 | 0.039 | 1.07 | 18.35 |
| P16278 | GLB1 | Beta-galactosidase | 4.43 | 0.044 | 1.04 | 18.85 |
| Q9BYV6 | TRIM55 | Tripartite motif-containing protein 55 | 4.40 | 0.053 | 0.98 | 19.76 |
| O14958 | CASQ2 | Calsequestrin-2 | 4.37 | 0.062 | 0.93 | 20.66 |
| Q9UPT5 | EXOC7 | Exocyst complex component 7 | 4.31 | 0.063 | 0.92 | 20.09 |
| Q99816 | TSG101 | Tumor susceptibility gene 101 protein | 4.30 | 0.064 | 0.92 | 20.13 |
| O60662 | KLHL41 | Kelch-like protein 41 | 4.28 | 0.040 | 1.07 | 17.21 |
| Q7L523 | RRAGA | Ras-related GTP-binding protein A | 4.28 | 0.030 | 1.15 | 15.89 |
| P78385 | KRT83 | Keratin, type II cuticular Hb3 | 4.28 | 0.051 | 0.99 | 18.39 |
| Q14974 | KPNB1 | Importin subunit beta-1 | 4.21 | 0.067 | 0.90 | 19.64 |
| P08185 | SERPINA6 | Corticosteroid-binding globulin | 4.17 | 0.061 | 0.94 | 18.50 |
| Q15642 | TRIP10 | Cdc42-interacting protein 4 | 4.14 | 0.072 | 0.88 | 19.49 |
| Q5TEU4 | NDUFAF5 | Arginine-hydroxylase NDUFAF5, mito- chondrial | 4.12 | 0.054 | 0.97 | 17.46 |
| Q9Y2D5 | AKAP2 | A-kinase anchor protein 2 | 4.08 | 0.065 | 0.92 | 18.18 |
| Q9NRK6 | ABCB10 | ATP-binding cassette sub-family B mem- ber 10, mitochondrial | 4.06 | 0.061 | 0.94 | 17.61 |
| P50579 | METAP2 | Methionine aminopeptidase 2 | 4.00 | 0.074 | 0.87 | 18.30 |
| P07686 | HEXB | Beta-hexosaminidase subunit beta | 3.94 | 0.052 | 0.99 | 15.72 |
| O00232 | PSMD12 | 26S proteasome non-ATPase regulatory subunit 12 | 3.89 | 0.040 | 1.06 | 14.28 |
| P05452 | CLEC3B | Tetranectin | 3.86 | 0.042 | 1.05 | 14.15 |
| Q9NZA1 | CLIC5 | Chloride intracellular channel protein 5 | 3.85 | 0.068 | 0.90 | 16.43 |

| | | | | | | |
|--------|---------|--|------|-------|------|-------|
| O75347 | TBCA | Tubulin-specific chaperone A | 3.84 | 0.041 | 1.06 | 13.95 |
| Q9Y2S2 | CRYL1 | Lambda-crystallin homolog | 3.78 | 0.046 | 1.03 | 13.94 |
| O14972 | VPS26C | Vacuolar protein sorting-associated protein 26C | 3.78 | 0.068 | 0.91 | 15.76 |
| P25788 | PSMA3 | Proteasome subunit alpha type-3 (Macropain subunit C8) | 3.68 | 0.058 | 0.96 | 14.17 |
| Q9BSH4 | TACO1 | Translational activator of cytochrome c oxidase 1 | 3.66 | 0.071 | 0.90 | 14.95 |
| Q9UGJ0 | PRKAG2 | 5'-AMP-activated protein kinase subunit gamma-2 | 3.60 | 0.057 | 0.96 | 13.46 |
| Q9NTX5 | ECHDC1 | Ethylmalonyl-CoA decarboxylase | 3.60 | 0.053 | 0.99 | 13.12 |
| Q9Y333 | LSM2 | U6 snRNA-associated Sm-like protein LSm2 | 3.58 | 0.053 | 0.99 | 13.03 |
| P35659 | DEK | Protein DEK | 3.50 | 0.069 | 0.91 | 13.50 |
| Q96NY7 | CLIC6 | Chloride intracellular channel protein 6 | 3.49 | 0.049 | 1.01 | 12.11 |
| Q9BTW9 | TBCD | Tubulin-specific chaperone D | 3.48 | 0.029 | 1.14 | 10.65 |
| Q9BUB7 | TMEM70 | Transmembrane protein 70 | 3.47 | 0.067 | 0.91 | 13.19 |
| Q92599 | SEPTIN8 | Septin-8 | 3.47 | 0.036 | 1.08 | 11.10 |
| Q14914 | PTGR1 | Prostaglandin reductase 1 | 3.45 | 0.036 | 1.09 | 10.97 |
| Q3ZCW2 | LGALSL | Galectin-related protein | 3.42 | 0.040 | 1.06 | 11.04 |
| Q96DG6 | CMBL | Carboxymethylenebutenolidase homolog | 3.41 | 0.039 | 1.06 | 10.96 |
| Q8WU90 | ZC3H15 | Zinc finger CCCH domain-containing protein 15 | 3.41 | 0.036 | 1.08 | 10.74 |
| Q9BQA1 | WDR77 | Methylosome protein 50 | 3.31 | 0.048 | 1.01 | 10.85 |
| P41250 | GARS1 | Glycine--tRNA ligase | 3.27 | 0.052 | 0.99 | 10.79 |
| Q14240 | EIF4A2 | Eukaryotic initiation factor 4A-II | 3.22 | 0.051 | 1.00 | 10.44 |
| Q96RP9 | GFM1 | Elongation factor G, mitochondrial | 3.11 | 0.066 | 0.93 | 10.42 |
| Q9GZV1 | ANKRD2 | Ankyrin repeat domain-containing protein 2 | 3.09 | 0.054 | 0.98 | 9.72 |
| Q15008 | PSMD6 | 26S proteasome non-ATPase regulatory subunit 6 | 3.08 | 0.072 | 0.91 | 10.45 |
| Q8WZA9 | IRGQ | Immunity-related GTPase family Q protein | 3.07 | 0.073 | 0.90 | 10.43 |
| Q86TI2 | DPP9 | Dipeptidyl peptidase 9 | 3.04 | 0.054 | 0.98 | 9.45 |

| | | | | | | |
|--------|----------|--|------|-------|------|------|
| Q9NPH2 | ISYNA1 | Inositol-3-phosphate synthase 1 | 3.01 | 0.052 | 0.99 | 9.17 |
| P50440 | GATM | Glycine amidinotransferase, mitochondrial | 2.96 | 0.071 | 0.91 | 9.62 |
| O14775 | GNB5 | Guanine nucleotide-binding protein subunit beta-5 | 2.96 | 0.069 | 0.92 | 9.53 |
| P47895 | ALDH1A3 | Aldehyde dehydrogenase family 1 member A3 | 2.95 | 0.056 | 0.97 | 8.94 |
| Q8N128 | FAM177A1 | Protein FAM177A1 | 2.91 | 0.068 | 0.92 | 9.18 |
| Q9UHI8 | ADAMTS1 | A disintegrin and metalloproteinase with thrombospondin motifs 1 | 2.90 | 0.057 | 0.97 | 8.69 |
| Q63HQ2 | EGFLAM | Pikachurin | 2.89 | 0.063 | 0.95 | 8.83 |
| P21953 | BCKDHB | 2-oxoisovalerate dehydrogenase subunit beta, mitochondrial | 2.88 | 0.054 | 0.98 | 8.47 |
| P46926 | GNPDA1 | Glucosamine-6-phosphate isomerase 1 | 2.88 | 0.067 | 0.93 | 8.97 |
| Q9UDY4 | DNAJB4 | DnaJ homolog subfamily B member 4 | 2.86 | 0.050 | 1.00 | 8.18 |
| Q15257 | PTPA | Serine/threonine-protein phosphatase 2A activator | 2.86 | 0.067 | 0.93 | 8.79 |
| P14324 | FDPS | Farnesyl pyrophosphate synthase | 2.85 | 0.048 | 1.01 | 8.07 |
| P34897 | SHMT2 | Serine hydroxymethyltransferase, mitochondrial | 2.81 | 0.075 | 0.90 | 8.75 |
| Q8TAT6 | NPLOC4 | Nuclear protein localization protein 4 homolog | 2.78 | 0.064 | 0.94 | 8.20 |
| Q96SW2 | CRBN | Protein cereblon | 2.74 | 0.048 | 1.01 | 7.45 |
| Q9P2T1 | GMPR2 | GMP reductase 2 | 2.70 | 0.075 | 0.91 | 8.08 |
| Q6P5Q4 | LMOD2 | Leiomodin-2 | 2.65 | 0.049 | 1.00 | 7.02 |
| P13807 | GYS1 | Glycogen [starch] synthase, muscle | 2.49 | 0.059 | 0.97 | 6.43 |
| O14579 | COPE | Coatomer subunit epsilon | 2.48 | 0.065 | 0.94 | 6.53 |

* Color-coding corresponding to color-coding used in Supplementary Figure S2: proteins assigned to carbohydrate metabolism (green), nucleotide metabolism (blue), protein biosynthesis (red), and the proteasome (yellow). *p*-values and Odds ratios were calculated by the *glm* function, confident intervals (95%) by the *confint.default* function of the *stats* package in R using LFQ protein intensities.

Table S3. Aortic stenosis-associated proteins: OR < 1, *p* < 0.075. OR: Odds ratio, i.e. chance to be assigned to the aortic stenosis group when protein intensity is high.

| Protein ID | Gene name | Protein name* | OR | p-value | 95% CI lower limit | 95% CI upper limit |
|------------|-----------|--|------|---------|-----------------------|-----------------------|
| Q8NFW8 | CMA5 | N-acylneuraminate cytidyltransferase | 0.01 | 0.024 | 0.00 | 0.50 |
| Q687X5 | STEAP4 | Metalloreductase STEAP4 | 0.01 | 0.032 | 0.00 | 0.66 |
| Q709C8 | VPS13C | Intermembrane lipid transfer protein VPS13C | 0.01 | 0.031 | 0.00 | 0.67 |
| Q32MK0 | MYLK3 | Myosin light chain kinase 3 | 0.06 | 0.028 | 0.00 | 0.74 |
| O95573 | ACSL3 | Fatty acid CoA ligase Acsl3 | 0.06 | 0.047 | 0.00 | 0.96 |
| O15511 | ARPC5 | Actin-related protein 2/3 complex subunit 5 | 0.07 | 0.019 | 0.01 | 0.65 |
| Q9Y383 | LUC7L2 | Putative RNA-binding protein Luc7-like 2 | 0.10 | 0.042 | 0.01 | 0.92 |
| P02549 | SPTA1 | Spectrin alpha chain, erythrocytic 1 | 0.10 | 0.021 | 0.01 | 0.71 |
| P60174 | TPI1 | Triosephosphate isomerase | 0.10 | 0.043 | 0.01 | 0.93 |
| Q03013 | GSTM4 | Glutathione S-transferase Mu 4 | 0.11 | 0.054 | 0.01 | 1.04 |
| P10321 | HLA-C | HLA class I histocompatibility antigen, C alpha chain | 0.11 | 0.033 | 0.01 | 0.84 |
| Q4KMQ2 | ANO6 | Anoctamin-6 | 0.11 | 0.031 | 0.02 | 0.82 |
| P00918 | CA2 | Carbonic anhydrase 2 | 0.11 | 0.024 | 0.02 | 0.75 |
| Q92879 | CELF1 | CUGBP Elav-like family member 1 | 0.13 | 0.071 | 0.01 | 1.19 |
| Q5T653 | MRPL2 | 39S ribosomal protein L2, mitochondrial, L2mt, MRP-L2 | 0.15 | 0.059 | 0.02 | 1.07 |
| P07814 | EPRS1 | Bifunctional glutamate/proline--tRNA ligase | 0.16 | 0.068 | 0.02 | 1.14 |
| P46937 | YAP1 | Transcriptional coactivator YAP1, Yes-associated protein 1 | 0.16 | 0.034 | 0.03 | 0.87 |
| Q14693 | LPIN1 | Phosphatidate phosphatase LPIN1 | 0.18 | 0.048 | 0.03 | 0.99 |
| Q8NE62 | CHDH | Choline dehydrogenase, mitochondrial | 0.19 | 0.056 | 0.04 | 1.04 |
| P51991 | HNRNPA3 | Heterogeneous nuclear ribonucleoprotein A3 | 0.20 | 0.042 | 0.04 | 0.94 |
| P09651 | HNRNPA1 | Heterogeneous nuclear ribonucleoprotein A1, hnRNP A1 | 0.20 | 0.053 | 0.04 | 1.02 |
| Q9UBV2 | SEL1L | Protein sel-1 homolog 1 | 0.21 | 0.042 | 0.05 | 0.95 |
| P07203 | GPX1 | Glutathione peroxidase 1 | 0.22 | 0.044 | 0.05 | 0.96 |

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|---------------|---------|--|------|-------|------|------|
| Q96A35 | MRPL24 | 39S ribosomal protein L24, mitochondrial | 0.22 | 0.035 | 0.05 | 0.90 |
| P16452 | EPB42 | Protein 4.2 | 0.22 | 0.032 | 0.06 | 0.88 |
| Q9Y6C2 | EMILIN1 | EMILIN-1 | 0.22 | 0.035 | 0.06 | 0.90 |
| P00414 | MT-CO3 | Cytochrome c oxidase subunit 3 | 0.23 | 0.042 | 0.05 | 0.95 |
| Q8N163 | CCAR2 | Cell cycle and apoptosis regulator protein 2 | 0.23 | 0.065 | 0.05 | 1.09 |
| P60953 | CDC42 | Cell division control protein 42 homolog | 0.24 | 0.042 | 0.06 | 0.95 |
| P05164 | MPO | Myeloperoxidase | 0.24 | 0.027 | 0.07 | 0.85 |
| Q01628 | IFITM3 | Interferon-induced transmembrane protein 3 | 0.24 | 0.054 | 0.06 | 1.02 |
| Q99808 | SLC29A1 | Equilibrative nucleoside transporter 1 | 0.24 | 0.061 | 0.06 | 1.06 |
| P54578 | USP14 | Ubiquitin carboxyl-terminal hydrolase 14 | 0.25 | 0.039 | 0.06 | 0.93 |
| Q9BQJ4 | TMEM47 | Transmembrane protein 47 | 0.25 | 0.028 | 0.07 | 0.86 |
| Q13310 | PABPC4 | Polyadenylate-binding protein 4 | 0.25 | 0.031 | 0.07 | 0.88 |
| P05023 | ATP1A1 | Sodium/potassium-transporting ATPase subunit alpha-1 | 0.26 | 0.047 | 0.07 | 0.98 |
| P02788 | LTF | Lactotransferrin | 0.27 | 0.023 | 0.09 | 0.83 |
| P21695 | GPD1 | Glycerol-3-phosphate dehydrogenase | 0.28 | 0.070 | 0.07 | 1.11 |
| Q9ULP0 | NDRG4 | Protein NDRG4 | 0.28 | 0.075 | 0.07 | 1.13 |
| Q9Y3U8 | RPL36 | 60S ribosomal protein L36 | 0.29 | 0.049 | 0.08 | 1.00 |
| O60884 | DNAJA2 | DnaJ homolog subfamily A member 2 | 0.29 | 0.053 | 0.08 | 1.02 |
| P42229_P51692 | STAT5A | Signal transducer and activator of transcription 5A | 0.30 | 0.047 | 0.09 | 0.98 |
| Q14766 | LTBP1 | Latent-transforming growth factor beta-binding protein 1 | 0.30 | 0.049 | 0.09 | 1.00 |
| P50552 | VASP | Vasodilator-stimulated phosphoprotein | 0.30 | 0.050 | 0.09 | 1.00 |
| Q92629 | SGCD | Delta-sarcoglycan | 0.31 | 0.066 | 0.09 | 1.08 |
| P50416 | CPT1A | Carnitine O-palmitoyltransferase 1, liver isoform | 0.31 | 0.041 | 0.10 | 0.95 |
| P84085 | ARF5 | ADP-ribosylation factor 5 | 0.31 | 0.056 | 0.09 | 1.03 |
| Q9NZ45 | CISD1 | CDGSH iron-sulfur domain-containing protein 1 | 0.32 | 0.053 | 0.10 | 1.02 |
| Q8NBX0 | SCCPDH | Saccharopine dehydrogenase-like oxidoreductase | 0.33 | 0.059 | 0.11 | 1.04 |

| | | | | | | |
|--------|--------|--------------------------------|------|-------|------|------|
| P16157 | ANK1 | Ankyrin-1 | 0.34 | 0.046 | 0.12 | 0.98 |
| P09382 | LGALS1 | Galectin-1 | 0.35 | 0.057 | 0.12 | 1.03 |
| P20336 | RAB3A | Ras-related protein Rab-3A | 0.38 | 0.055 | 0.14 | 1.02 |
| P02730 | SLC4A1 | Band 3 anion transport protein | 0.39 | 0.075 | 0.14 | 1.10 |
| P69891 | HBG1 | Hemoglobin subunit gamma-1 | 0.39 | 0.066 | 0.15 | 1.06 |

* Color-coding corresponding to color-coding used in Supplementary Figure S3: proteins assigned to the immune system (green) and hematopoietic system/angiogenesis (blue) and anemia (red) in specific. *p*-values and Odds ratios were calculated by the *glm* function, confident intervals (95%) by the *confint.default* function of the *stats* package in R using LFQ protein intensities.

Table S4. Results of g:Profiler enrichment analysis of proteins with $p < 0.075$ in the logistic regression analysis. Categories with adjusted p -value < 0.05 (calculated by the g:SCS approach in g:Profiler) in either one or both analyses of proteins with Odds ratio > 1 or Odds ratio < 1 shown.

| | | term_id | term_size (no of mole- cules in category) | - proteins | | | | - proteins | | | |
|-----------|--|------------|---|--------------------|----------------------------|----------------------|--------------------------|--------------------|----------------------------|----------------------|--------------------------|
| term_name | | | | adj pValue (AR) | log10adj pValue (AR) | list size (AR) | with OR > 1 (AR) | adj pValue (AS) | log10adj pValue (AS) | list size (AS) | with OR < 1 (AS) |
| GO:BP | organonitrogen compound metabolic process | GO:1901564 | 6522 | 9.9E-05 | 4.00 | 114 | 63 | | | | |
| GO:BP | small molecule metabolic process | GO:0044281 | 1837 | 1.7E-04 | 3.77 | 114 | 29 | | | | |
| GO:BP | catabolic process | GO:0009056 | 2587 | 2.5E-04 | 3.60 | 114 | 35 | | | | |
| GO:BP | cellular catabolic process | GO:0044248 | 2121 | 1.0E-03 | 2.98 | 114 | 30 | | | | |
| GO:BP | nucleobase-containing small molecule metabolic process | GO:0055086 | 674 | 1.2E-03 | 2.92 | 114 | 16 | | | | |
| GO:BP | carbohydrate metabolic process | GO:0005975 | 594 | 1.3E-03 | 2.89 | 114 | 15 | | | | |
| GO:BP | carbohydrate catabolic process | GO:0016052 | 154 | 3.3E-03 | 2.48 | 114 | 8 | | | | |
| GO:BP | organic substance catabolic process | GO:1901575 | 2128 | 3.6E-03 | 2.44 | 114 | 29 | | | | |
| GO:BP | 5-phosphoribose 1-diphosphate biosynthetic process | GO:0006015 | 6 | 5.2E-03 | 2.28 | 114 | 3 | | | | |
| GO:BP | 5-phosphoribose 1-diphosphate metabolic process | GO:0046391 | 6 | 5.2E-03 | 2.28 | 114 | 3 | | | | |
| GO:BP | generation of precursor metabolites and energy | GO:0006091 | 505 | 6.0E-03 | 2.22 | 114 | 13 | | | | |
| GO:BP | cellular carbohydrate metabolic process | GO:0044262 | 291 | 7.1E-03 | 2.15 | 114 | 10 | | | | |

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|-------|---|------------|-------|---------|-------|-----|-----|---------|------|----|----|
| GO:BP | negative regulation of molecular function | GO:0044092 | 1111 | 1.3E-02 | 1.88 | 114 | 19 | | | | |
| GO:BP | purine-containing compound biosynthetic process | GO:0072522 | 251 | 1.6E-02 | 1.81 | 114 | 9 | | | | |
| GO:BP | cellular macromolecule metabolic process | GO:0044260 | 3234 | 1.7E-02 | 1.76 | 114 | 36 | | | | |
| GO:BP | purine-containing compound metabolic process | GO:0072521 | 506 | 3.3E-02 | 1.48 | 114 | 12 | | | | |
| GO:BP | nucleotide metabolic process | GO:0009117 | 597 | 3.6E-02 | 1.44 | 114 | 13 | | | | |
| GO:BP | proteolysis | GO:0006508 | 1783 | 4.1E-02 | 1.38 | 114 | 24 | | | | |
| GO:BP | nucleoside phosphate metabolic process | GO:0006753 | 607 | 4.3E-02 | 1.37 | 114 | 13 | | | | |
| GO:BP | organic acid metabolic process | GO:0006082 | 958 | | | | | 8.8E-03 | 2.06 | 54 | 12 |
| GO:BP | response to stress | GO:0006950 | 3938 | | | | | 2.5E-02 | 1.60 | 54 | 24 |
| GO:BP | response to chemical | GO:0042221 | 4383 | | | | | 4.9E-02 | 1.31 | 54 | 25 |
| GO:BP | oxoacid metabolic process | GO:0043436 | 951 | | | | | 5.0E-02 | 1.30 | 54 | 11 |
| GO:BP | regulation of biological quality | GO:0065008 | 3726 | | | | | 9.5E-03 | 2.02 | 54 | 24 |
| GO:BP | cellular response to chemical stimulus | GO:0070887 | 3049 | | | | | 1.9E-02 | 1.73 | 54 | 21 |
| GO:CC | cytoplasm | GO:0005737 | 12270 | 6.6E-19 | 18.18 | 114 | 109 | 6.5E-05 | 4.19 | 54 | 48 |
| GO:CC | cytosol | GO:0005829 | 5419 | 5.3E-12 | 11.28 | 114 | 67 | | | | |
| GO:CC | extracellular exosome | GO:0070062 | 2108 | 9.0E-12 | 11.05 | 114 | 41 | 1.6E-05 | 4.80 | 54 | 20 |
| GO:CC | extracellular vesicle | GO:1903561 | 2132 | 1.3E-11 | 10.88 | 114 | 41 | 3.3E-06 | 5.48 | 54 | 21 |
| GO:CC | extracellular organelle | GO:0043230 | 2133 | 1.3E-11 | 10.87 | 114 | 41 | 3.3E-06 | 5.48 | 54 | 21 |
| GO:CC | extracellular membrane-bounded organelle | GO:0065010 | 2133 | 1.3E-11 | 10.87 | 114 | 41 | 3.3E-06 | 5.48 | 54 | 21 |
| GO:CC | vesicle | GO:0031982 | 3973 | 1.5E-09 | 8.82 | 114 | 53 | 7.6E-03 | 2.12 | 54 | 23 |
| GO:CC | secretory granule lumen | GO:0034774 | 320 | 4.4E-07 | 6.36 | 114 | 14 | | | | |
| GO:CC | cytoplasmic vesicle lumen | GO:0060205 | 323 | 5.0E-07 | 6.30 | 114 | 14 | | | | |

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|--|------------|-------|---------|------|-----|-----|---------|------|----|----|--|
| GO:CC vesicle lumen | GO:0031983 | 325 | 5.4E-07 | 6.27 | 114 | 14 | | | | | |
| GO:CC extracellular space | GO:0005615 | 3368 | 2.0E-06 | 5.70 | 114 | 43 | 2.3E-02 | 1.63 | 54 | 20 | |
| GO:CC extracellular region | GO:0005576 | 4302 | 4.4E-06 | 5.36 | 114 | 49 | 8.5E-03 | 2.07 | 54 | 24 | |
| GO:CC catalytic complex | GO:1902494 | 1707 | 1.3E-05 | 4.88 | 114 | 28 | | | | | |
| GO:CC proteasome complex | GO:0000502 | 66 | 1.7E-05 | 4.77 | 114 | 7 | | | | | |
| GO:CC ficolin-1-rich granule lumen | GO:1904813 | 124 | 9.2E-05 | 4.04 | 114 | 8 | | | | | |
| GO:CC endopeptidase complex | GO:1905369 | 88 | 1.3E-04 | 3.90 | 114 | 7 | | | | | |
| GO:CC intracellular anatomical structure | GO:0005622 | 17820 | 1.5E-04 | 3.82 | 114 | 111 | | | | | |
| GO:CC contractile fiber | GO:0043292 | 247 | 2.2E-04 | 3.65 | 114 | 10 | | | | | |
| GO:CC ribose phosphate diphosphokinase complex | GO:0002189 | 5 | 4.3E-04 | 3.37 | 114 | 3 | | | | | |
| GO:CC peptidase complex | GO:1905368 | 122 | 1.1E-03 | 2.94 | 114 | 7 | | | | | |
| GO:CC myofibril | GO:0030016 | 238 | 1.4E-03 | 2.84 | 114 | 9 | | | | | |
| GO:CC ficolin-1-rich granule | GO:0101002 | 184 | 1.8E-03 | 2.75 | 114 | 8 | | | | | |
| GO:CC secretory vesicle | GO:0099503 | 1043 | 2.5E-03 | 2.60 | 114 | 18 | | | | | |
| GO:CC membrane-enclosed lumen | GO:0031974 | 6637 | 3.4E-03 | 2.47 | 114 | 57 | | | | | |
| GO:CC organelle lumen | GO:0043233 | 6637 | 3.4E-03 | 2.47 | 114 | 57 | | | | | |
| GO:CC intracellular organelle lumen | GO:0070013 | 6637 | 3.4E-03 | 2.47 | 114 | 57 | | | | | |
| GO:CC sarcomere | GO:0030017 | 217 | 6.0E-03 | 2.22 | 114 | 8 | | | | | |
| GO:CC supramolecular fiber | GO:0099512 | 1017 | 6.9E-03 | 2.16 | 114 | 17 | | | | | |
| GO:CC supramolecular polymer | GO:0099081 | 1026 | 7.8E-03 | 2.11 | 114 | 17 | | | | | |
| GO:CC secretory granule | GO:0030141 | 870 | 1.6E-02 | 1.81 | 114 | 15 | | | | | |
| GO:CC organelle | GO:0043226 | 16770 | 2.5E-02 | 1.60 | 114 | 104 | | | | | |
| GO:CC cytoskeleton | GO:0005856 | 2381 | 2.6E-02 | 1.59 | 114 | 27 | | | | | |
| GO:CC supramolecular complex | GO:0099080 | 1377 | 3.0E-02 | 1.52 | 114 | 19 | | | | | |
| GO:CC azurophil granule lumen | GO:0035578 | 91 | 3.6E-02 | 1.44 | 114 | 5 | | | | | |
| GO:CC I band | GO:0031674 | 146 | 3.8E-02 | 1.43 | 114 | 6 | | | | | |

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|-------|---|--------------------|-------|---------|------|-----|-----|---------|------|----|----|
| GO:CC | mitochondrion | GO:0005739 | 1684 | | | | | 4.7E-02 | 1.32 | 54 | 13 |
| GO:CC | mitochondrial envelope | GO:0005740 | 807 | | | | | 3.8E-02 | 1.42 | 54 | 9 |
| GO:CC | mitochondrial outer membrane | GO:0005741 | 211 | | | | | 4.4E-02 | 1.36 | 54 | 5 |
| GO:CC | membrane | GO:0016020 | 9856 | | | | | 1.5E-02 | 1.83 | 54 | 39 |
| GO:CC | mitochondrial membrane | GO:0031966 | 756 | | | | | 2.3E-02 | 1.64 | 54 | 9 |
| GO:CC | phagocytic vesicle lumen | GO:0097013 | 5 | | | | | 1.5E-02 | 1.83 | 54 | 2 |
| GO:MF | protein binding | GO:0005515 | 14832 | 5.3E-06 | 5.28 | 117 | 110 | | | | |
| GO:MF | identical protein binding | GO:0042802 | 2121 | 4.5E-04 | 3.35 | 117 | 31 | | | | |
| GO:MF | ribose phosphate diphosphokinase activity | GO:0004749 | 5 | 9.4E-04 | 3.03 | 117 | 3 | | | | |
| GO:MF | diphosphotransferase activity | GO:0016778 | 6 | 1.9E-03 | 2.73 | 117 | 3 | | | | |
| GO:MF | protein homodimerization activity | GO:0042803 | 701 | 6.6E-03 | 2.18 | 117 | 15 | | | | |
| GO:MF | UDP-glucosyltransferase activity | GO:0035251 | 12 | 2.0E-02 | 1.70 | 117 | 3 | | | | |
| GO:MF | protein dimerization activity | GO:0046983 | 1088 | 2.7E-02 | 1.57 | 117 | 18 | | | | |
| KEGG | Proteasome | KEGG:03050 | 46 | 2.1E-03 | 2.67 | 63 | 5 | | | | |
| KEGG | Starch and sucrose metabolism | KEGG:00500 | 36 | 1.3E-02 | 1.88 | 63 | 4 | | | | |
| KEGG | Metabolic pathways | KEGG:01100 | 1535 | 2.5E-02 | 1.60 | 63 | 24 | | | | |
| KEGG | Glycosaminoglycan degradation | KEGG:00531 | 19 | 3.2E-02 | 1.49 | 63 | 3 | | | | |
| REAC | Metabolism | REAC:R-HSA-1430728 | 2075 | 2.2E-06 | 5.66 | 86 | 41 | | | | |
| REAC | Metabolism of carbohydrates | REAC:R-HSA-71387 | 284 | 1.4E-03 | 2.87 | 86 | 12 | | | | |
| REAC | AUF1 (hnRNP D0) binds and destabilizes mRNA | REAC:R-HSA-450408 | 54 | 2.0E-03 | 2.69 | 86 | 6 | | | | |

| | | | | | | | |
|------|---|--------------------|-----|---------|------|----|---|
| REAC | Hh mutants are degraded by ERAD | REAC:R-HSA-5362768 | 56 | 2.5E-03 | 2.60 | 86 | 6 |
| REAC | Hh mutants abrogate ligand secretion | REAC:R-HSA-5387390 | 59 | 3.5E-03 | 2.46 | 86 | 6 |
| REAC | Defective CFTR causes cystic fibrosis | REAC:R-HSA-5678895 | 61 | 4.2E-03 | 2.38 | 86 | 6 |
| REAC | Hedgehog ligand biogenesis | REAC:R-HSA-5358346 | 65 | 6.1E-03 | 2.22 | 86 | 6 |
| REAC | ABC-family proteins mediated transport | REAC:R-HSA-382556 | 103 | 8.7E-03 | 2.06 | 86 | 7 |
| REAC | ABC transporter disorders | REAC:R-HSA-5619084 | 78 | 1.7E-02 | 1.76 | 86 | 6 |
| REAC | Cross-presentation of soluble exogenous antigens (endosomes) | REAC:R-HSA-1236978 | 49 | 2.0E-02 | 1.70 | 86 | 5 |
| REAC | Regulation of activated PAK-2p34 by proteasome mediated degradation | REAC:R-HSA-211733 | 50 | 2.2E-02 | 1.65 | 86 | 5 |
| REAC | Regulation of ornithine decarboxylase (ODC) | REAC:R-HSA-350562 | 51 | 2.4E-02 | 1.61 | 86 | 5 |
| REAC | Degradation of beta-catenin by the destruction complex | REAC:R-HSA-195253 | 83 | 2.5E-02 | 1.61 | 86 | 6 |

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|------|---|--------------------|----|---------|------|----|---|
| REAC | Vpu mediated degradation of CD4 | REAC:R-HSA-180534 | 52 | 2.7E-02 | 1.57 | 86 | 5 |
| REAC | Autodegradation of the E3 ubiquitin ligase COP1 | REAC:R-HSA-349425 | 52 | 2.7E-02 | 1.57 | 86 | 5 |
| REAC | Ubiquitin Mediated Degradation of Phosphorylated Cdc25A | REAC:R-HSA-69601 | 52 | 2.7E-02 | 1.57 | 86 | 5 |
| REAC | p53-Independent DNA Damage Response | REAC:R-HSA-69610 | 52 | 2.7E-02 | 1.57 | 86 | 5 |
| REAC | p53-Independent G1/S DNA damage checkpoint | REAC:R-HSA-69613 | 52 | 2.7E-02 | 1.57 | 86 | 5 |
| REAC | Ubiquitin-dependent degradation of Cyclin D | REAC:R-HSA-75815 | 52 | 2.7E-02 | 1.57 | 86 | 5 |
| REAC | Glycogen metabolism | REAC:R-HSA-8982491 | 27 | 2.7E-02 | 1.57 | 86 | 4 |
| REAC | Glycogen storage disease type XV (GYG1) | REAC:R-HSA-3814836 | 2 | 2.8E-02 | 1.55 | 86 | 2 |
| REAC | Glycogen storage disease type 0 (muscle GYS1) | REAC:R-HSA-3828062 | 2 | 2.8E-02 | 1.55 | 86 | 2 |
| REAC | Regulation of Apoptosis | REAC:R-HSA-169911 | 53 | 3.0E-02 | 1.53 | 86 | 5 |
| REAC | Vif-mediated degradation of APOBEC3G | REAC:R-HSA-180585 | 53 | 3.0E-02 | 1.53 | 86 | 5 |
| REAC | Negative regulation of NOTCH4 signaling | REAC:R-HSA-9604323 | 53 | 3.0E-02 | 1.53 | 86 | 5 |

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|------|--|--------------------|-----|---------|------|----|---|
| REAC | Regulation of mRNA stability by proteins that bind AU-rich elements | REAC:R-HSA-450531 | 86 | 3.0E-02 | 1.52 | 86 | 6 |
| REAC | Host Interactions of HIV factors | REAC:R-HSA-162909 | 125 | 3.0E-02 | 1.52 | 86 | 7 |
| REAC | Regulation of RUNX3 expression and activity | REAC:R-HSA-8941858 | 54 | 3.2E-02 | 1.49 | 86 | 5 |
| REAC | SCF-beta-TrCP mediated degradation of Emi1 | REAC:R-HSA-174113 | 55 | 3.5E-02 | 1.45 | 86 | 5 |
| REAC | Degradation of AXIN | REAC:R-HSA-4641257 | 55 | 3.5E-02 | 1.45 | 86 | 5 |
| REAC | FBXL7 down-regulates AURKA during mitotic entry and in early mitosis | REAC:R-HSA-8854050 | 55 | 3.5E-02 | 1.45 | 86 | 5 |
| REAC | MAPK6/MAPK4 signaling | REAC:R-HSA-5687128 | 89 | 3.7E-02 | 1.44 | 86 | 6 |
| REAC | Degradation of DVL | REAC:R-HSA-4641258 | 57 | 4.2E-02 | 1.38 | 86 | 5 |
| REAC | Stabilization of p53 | REAC:R-HSA-69541 | 57 | 4.2E-02 | 1.38 | 86 | 5 |
| REAC | Metabolism of polyamines | REAC:R-HSA-351202 | 58 | 4.6E-02 | 1.34 | 86 | 5 |

| | | | | | | | | |
|------|------------------------------------|----------------------------|----|---------|------|----|---|--|
| REAC | NIK-->noncanonical NF-kB signaling | REAC:R- HSA- 5676590 | 59 | 5.0E-02 | 1.30 | 86 | 5 | |
|------|------------------------------------|----------------------------|----|---------|------|----|---|--|