Trisomy 21 causes changes in the circulating proteome indicative of chronic autoinflammation

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Supplementary Files.

Supplementary File 1. (a) Discovery Study. Kolmogorov Smirnoff analysis of T21 versus D21 cohort. Columns include: (A) Gene ID, (B) Chromosome, (C) Chromosome Start Position, (D) Chromosome End Position, (E) Chromosome Strand, (F) Gene Length, (G) Uniprot ID, (H) Soma ID, (I) Seq ID, (J) Average RFU, (K) Euploid (D21) average RFU, (L) Trisomy 21 (T21) average RFU, (M) Fold change (T21/D21), (N) Log_2 fold change, (O) p-value, (P) Bonferroni (Bon) adjusted p-value. **(b) Validation Study #1.** Kolmogorov Smirnoff analysis of T21 versus D21 cohort. Columns include: (A) Gene ID, (B) Uniprot ID, (C) Soma ID, (D) Seq ID, (E) Average RFU, (F) D21 average RFU, (G) T21 average RFU, (H) Fold change (T21/D21), (I) Log_2 fold change, (J) p-value, (K) Benjamini-Hochberg (BH) adjusted p-value. **(c) Validation Study #2.** Kolmogorov Smirnoff analysis of T21 versus D21 cohort. Columns include: (A) Gene ID, (B) Uniprot ID, (C) Soma ID, (D) Seq ID, (E) Average RFU, (F) D21 average RFU, (G) T21 average RFU, (H) Fold change (T21/D21), (I) Log_2 fold change, (J) p-value, (K) Benjamini-Hochberg (BH) adjusted p-value. **(d) Study overlaps.** Lists of differential proteins common among the three SOMAscan® studies. **(e) Meso Scale Discovery Assay.** Kolmogorov Smirnoff analysis of T21 versus D21 cohort. Columns include: (A) Cytokine, (B) D21 median protein concentration (ng/mL), (C) T21 median protein concentration (ng/mL), (D) Fold change (T21/D21), (E) Log_2 fold change, (F) p-value, (G) Benjamini-Hochberg adjusted p-value. **(f) Cohort information.** Statistics on individual cohorts including sex and age.

Supplementary File 2. Metascape Analysis. (a) Annotation of gene list used for analysis. **(b)** Enrichment analysis. **(c)** Random permutation analysis with 10 list of 300 randomly selected proteins.

Sullivan et al., Figure 1 S1

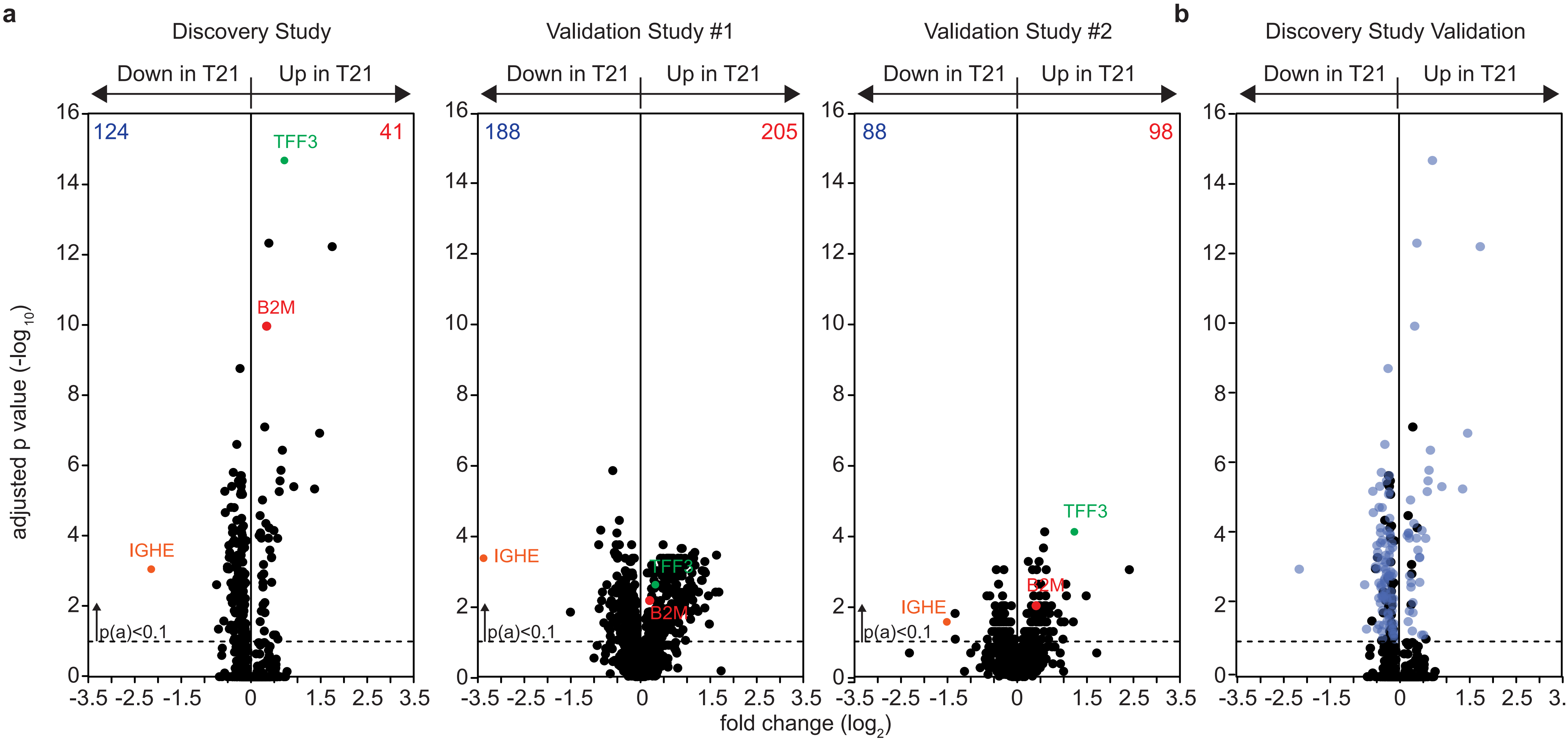


Figure 1 S1. (a) Volcano plots showing the 1047 proteins commonly detected in the samples unique to the Discovery Study (83 T21 and 52 D21), Validation Study #1 (38 T21 and 15 D21), and Validation Study #2 (22 T21 and 11 D21). Adjusted p values were calculated using KS test with Bonferroni correction for the Discovery Study and Benjamini-Hochberg correction for the Validation Studies. **(b)** Volcano plot of the Discovery study highlighting in blue those proteins with p(a)<0.1 (KS test with Bonferroni correction), that validated in one or both Validation Studies p(a)<0.1 (KS test with Benjamini-Hochberg correction).

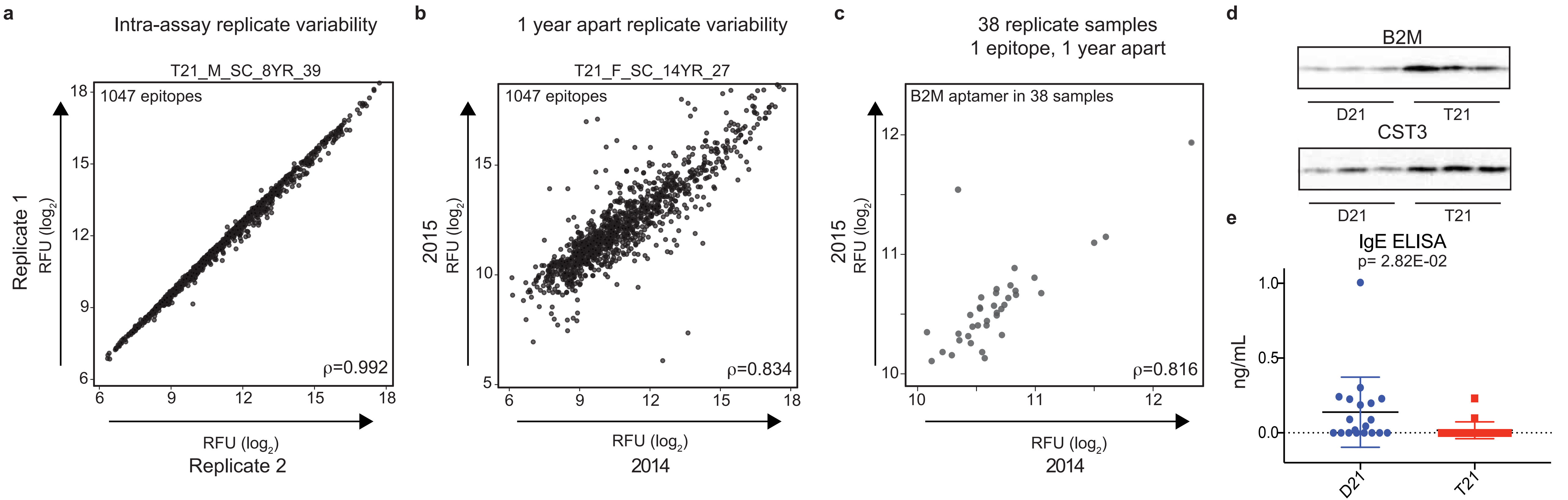


Figure 1 S2. (a). Scatter plot showing the 1047 epitopes measured twice during the same assay for sample T21_M_SC_8YR_39. **(b).** Scatter plot showing 1047 epitopes measured twice, in two different assays a year apart, for sample T21_F_SCP_14YR_27. **(c).** Scatter plot showing the values for the aptamer recognizing B2M in 38 samples that were measured twice, in two different assays a year apart. **(d)** Western blots for B2M and CST3 on select plasma samples from euploid (D21) and T21 individuals that showed low and high signals in the SOMAscan assay. **(e)** ELISA of IgE levels from 19 individuals with T21 and 19 euploid controls. Difference between the groups was calculated using the Kolmogorov-Smirnoff test.

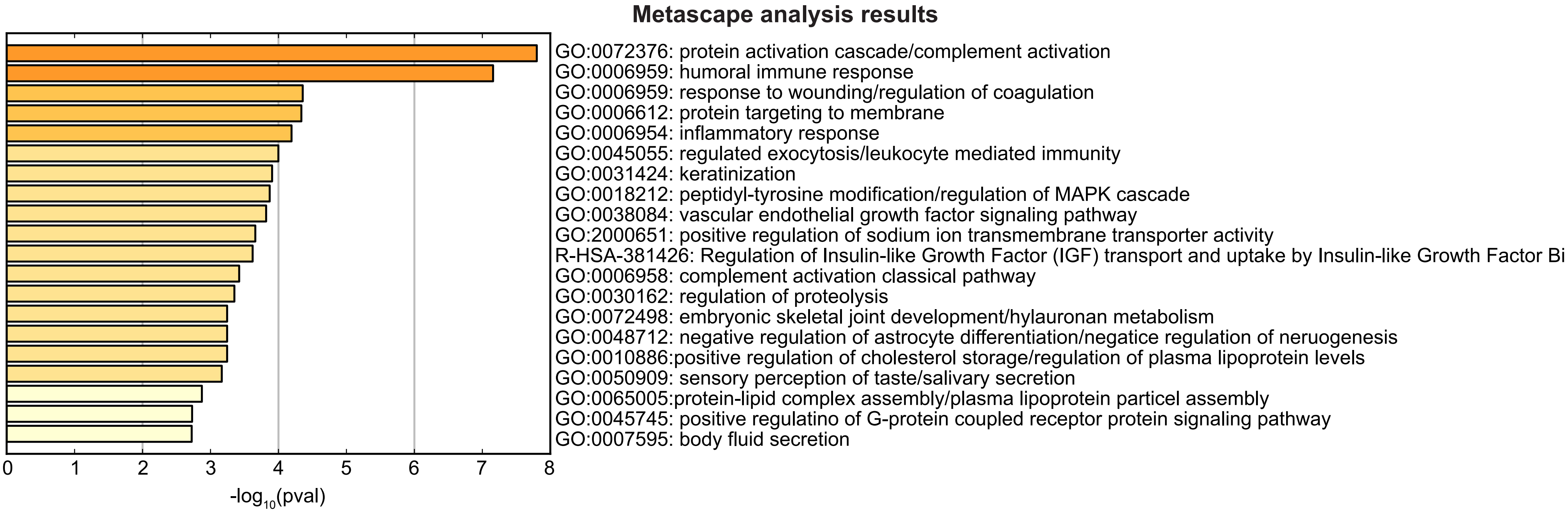
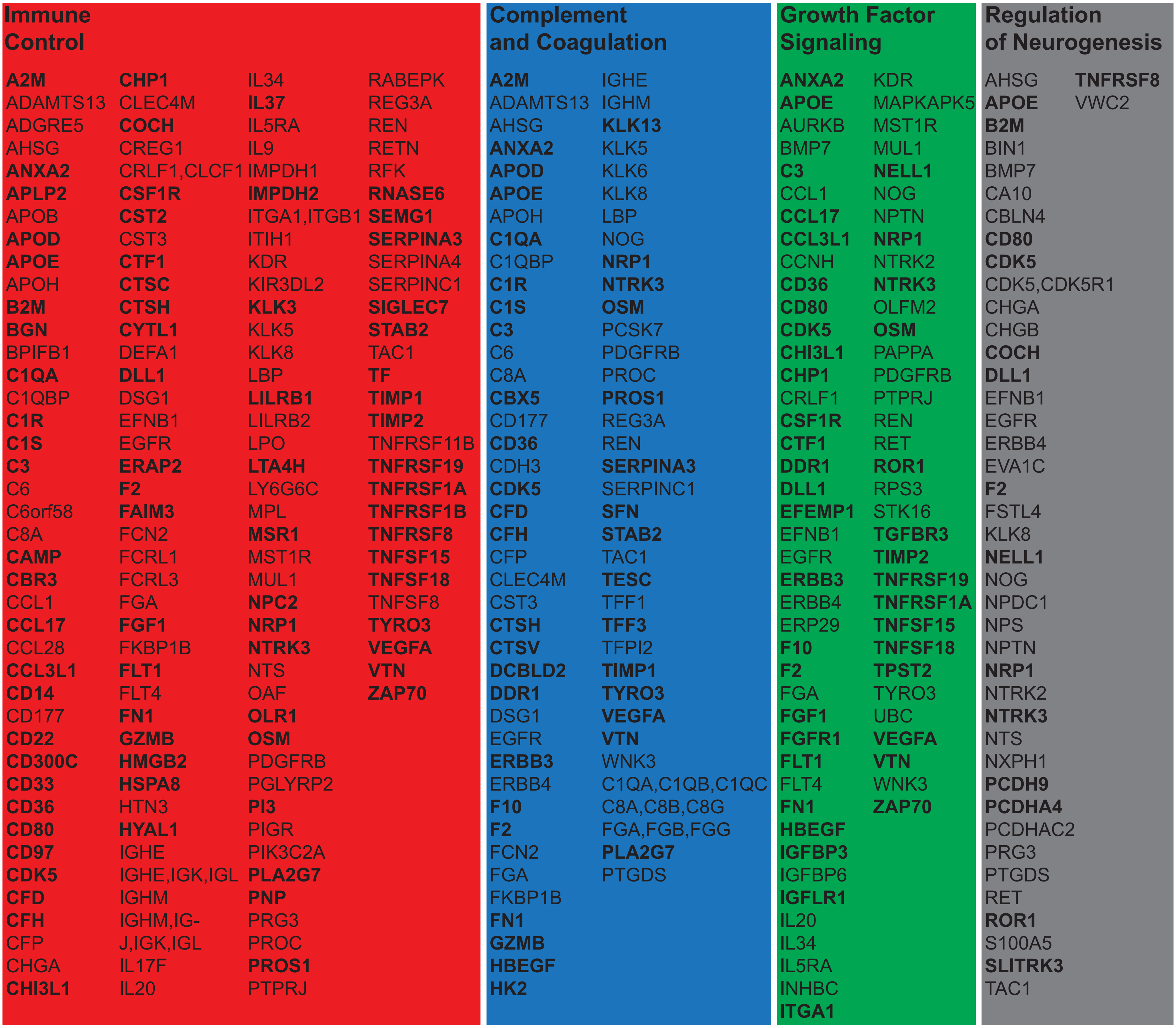


Figure 1 S3. Metascape heatmap of terms enriched in the 299 most significantly different proteins from the Discovery Study, colored by p-values.



Immune Control

- **A2M**
- ADAMTS13
- ADGRE5
- AHSG
- **ANXA2**
- **APLP2**
- APOB
- **APOD**
- **APOE**
- APOH
- **B2M**
- **BGN**
- BPIFB1
- **C1QA**
- C1QBP
- **C1R**
- **C1S**
- **C3**
- C6
- C6orf58
- C8A
- **CAMP**
- **CBR3**
- CCL1
- **CCL17**
- CCL28
- **CCL3L1**
- **CD14**
- CD177
- **CD22**
- **CD300C**
- **CD33**
- **CD36**
- **CD80**
- **CD97**
- **CDK5**
- **CFD**
- **CFH**
- CFP
- CHGA
- **CHI3L1**
- **CHP1**
- CLEC4M
- **COCH**
- CREG1
- CRLF1,CLCF1
- **CSF1R**
- **CST2**
- CST3
- **CTF1**
- **CTSC**
- **CTSH**
- **CYTL1**
- DEFA1
- **DLL1**
- DSG1
- EFNB1
- EGFR
- **ERAP2**
- **F2**
- **FAIM3**
- FCN2
- FCRL1
- FCRL3
- FGA
- **FGF1**
- FKBP1B
- **FLT1**
- FLT4
- **FN1**
- **GZMB**
- **HMGB2**
- **HSPA8**
- HTN3
- **HYAL1**
- IGHE
- IGHE,IGK,IGL
- IGHM
- IGHM,IGJ,IGK,IGL
- IL17F
- IL20
- IL34
- **IL37**
- IL5RA
- IL9
- IMPDH1
- **IMPDH2**
- ITGA1,ITGB1
- ITIH1
- KDR
- KIR3DL2
- **KLK3**
- KLK5
- KLK8
- LBP
- **LILRB1**
- LILRB2
- LPO
- **LTA4H**
- LY6G6C
- MPL
- **MSR1**
- MST1R
- MUL1
- **NPC2**
- **NRP1**
- **NTRK3**
- NTS
- OAF
- **OLR1**
- **OSM**
- PDGFRB
- PGLYRP2
- **PI3**
- PIGR
- PIK3C2A
- **PLA2G7**
- **PNP**
- PRG3
- PROC
- **PROS1**
- PTPRJ
- RABEPK
- REG3A
- REN
- RETN
- RFK
- **RNASE6**
- **SEMG1**
- **SERPINA3**
- SERPINA4
- SERPINC1
- **SIGLEC7**
- **STAB2**
- TAC1
- **TF**
- **TIMP1**
- **TIMP2**
- TNFRSF11B
- **TNFRSF19**
- **TNFRSF1A**
- **TNFRSF1B**
- **TNFRSF8**
- **TNFSF15**
- **TNFSF18**
- TNFSF8
- **TYRO3**
- **VEGFA**
- **VTN**
- **ZAP70**

Complement and Coagulation

- **A2M**
- ADAMTS13
- AHSG
- **ANXA2**
- **APOD**
- **APOE**
- APOH
- **C1QA**
- C1QBP
- **C1R**
- **C1S**
- **C3**
- C6
- C8A
- **CBX5**
- CD177
- **CD36**
- CDH3
- **CDK5**
- **CFD**
- **CFH**
- CFP
- CLEC4M
- CST3
- **CTSH**
- **CTSV**
- **DCBLD2**
- **DDR1**
- DSG1
- EGFR
- **ERBB3**
- ERBB4
- **F10**
- **F2**
- FCN2
- FGA
- FKBP1B
- **FN1**
- **GZMB**
- **HBEGF**
- **HK2**
- IGHE
- IGHM
- **KLK13**
- KLK5
- KLK6
- KLK8
- LBP
- NOG
- **NRP1**
- **NTRK3**
- **OSM**
- PCSK7
- PDGFRB
- PROC
- **PROS1**
- REG3A
- REN
- **SERPINA3**
- SERPINC1
- **SFN**
- **STAB2**
- TAC1
- **TESC**
- TFF1
- **TFF3**
- TFPI2
- **TIMP1**
- **TYRO3**
- **VEGFA**
- **VTN**
- WNK3
- C1QA,C1QB,C1QC
- C8A,C8B,C8G
- FGA,FGB,FGG
- **PLA2G7**
- PTGDS

Growth Factor Signaling

- **ANXA2**
- **APOE**
- AURKB
- BMP7
- **C3**
- CCL1
- **CCL17**
- **CCL3L1**
- CCNH
- **CD36**
- **CD80**
- **CDK5**
- **CHI3L1**
- **CHP1**
- CRLF1
- **CSF1R**
- **CTF1**
- **DDR1**
- **DLL1**
- **EFEMP1**
- EFNB1
- EGFR
- **ERBB3**
- ERBB4
- ERP29
- **F10**
- **F2**
- FGA
- **FGF1**
- **FGFR1**
- **FLT1**
- FLT4
- **FN1**
- **HBEGF**
- **IGFBP3**
- IGFBP6
- **IGFLR1**
- IL20
- IL34
- IL5RA
- INHBC
- **ITGA1**
- KDR
- MAPKAPK5
- MST1R
- MUL1
- **NELL1**
- NOG
- NPTN
- **NRP1**
- NTRK2
- **NTRK3**
- OLFM2
- **OSM**
- PAPPA
- PDGFRB
- PTPRJ
- REN
- RET
- **ROR1**
- RPS3
- STK16
- **TGFBR3**
- **TIMP2**
- **TNFRSF19**
- **TNFRSF1A**
- **TNFSF15**
- **TNFSF18**
- **TPST2**
- TYRO3
- UBC
- **VEGFA**
- **VTN**
- WNK3
- **ZAP70**

Regulation of Neurogenesis

- AHSG
- **APOE**
- **B2M**
- BIN1
- BMP7
- CA10
- CBLN4
- **CD80**
- **CDK5**
- CDK5,CDK5R1
- CHGA
- CHGB
- **COCH**
- **DLL1**
- EFNB1
- EGFR
- ERBB4
- EVA1C
- **F2**
- FSTL4
- KLK8
- **NELL1**
- NOG
- NPDC1
- NPS
- NPTN
- **NRP1**
- NTRK2
- **NTRK3**
- NTS
- NXPH1
- **PCDH9**
- **PCDHA4**
- PCDHAC2
- PRG3
- PTGDS
- RET
- **ROR1**
- S100A5
- **SLITRK3**
- TAC1
- **TNFRSF8**
- VWC2

Figure 1 S4. Table of differentially abundant proteins in each of the four largest functional categories. Protein identified as interferon-related in the Interferome database are highlighted in bold.

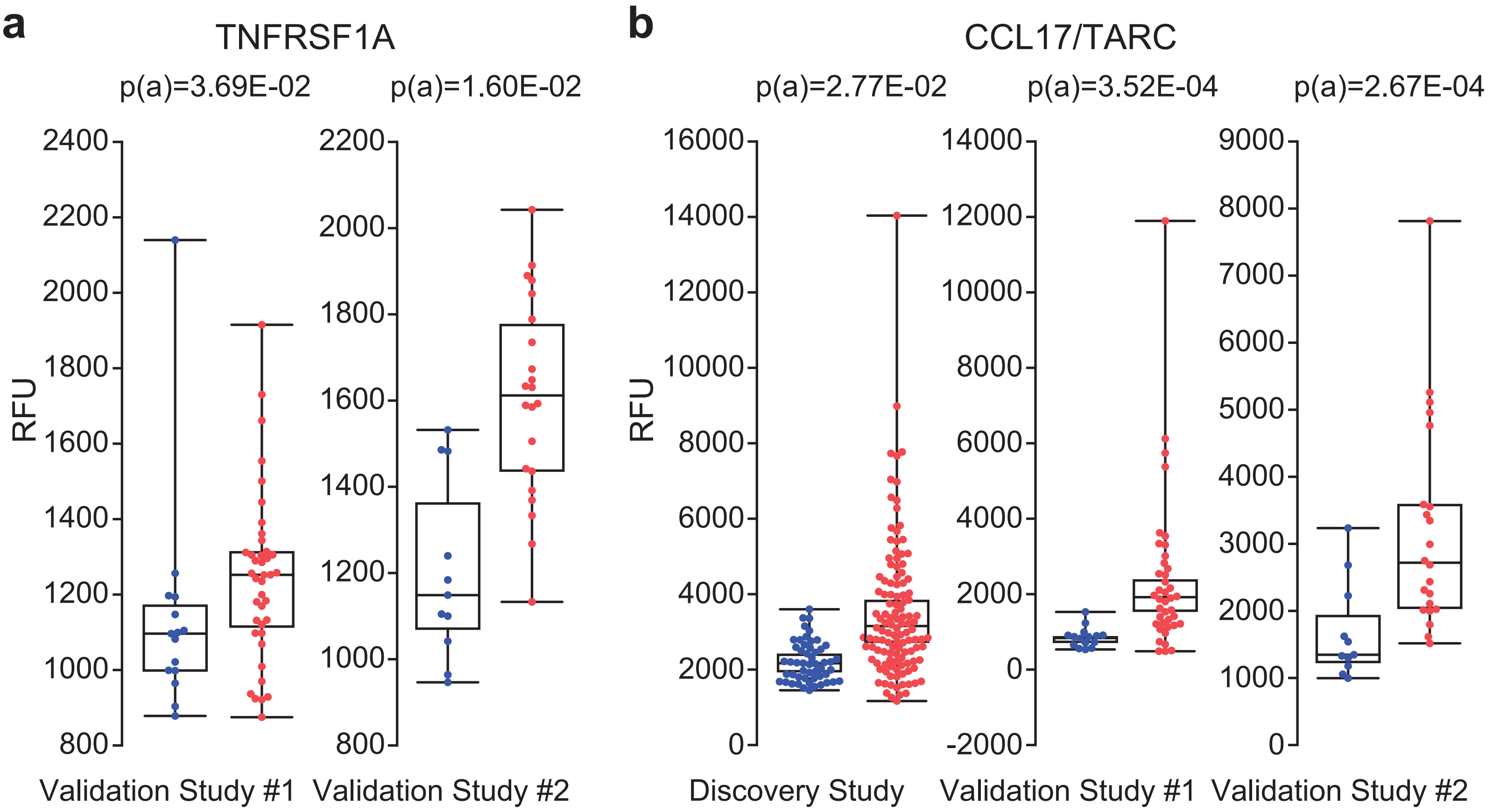


Figure 2 S1. Boxplots of raw data for TNFRSF1A from each validation study (**a**) or all three studies for CCL17/TARC (**b**).

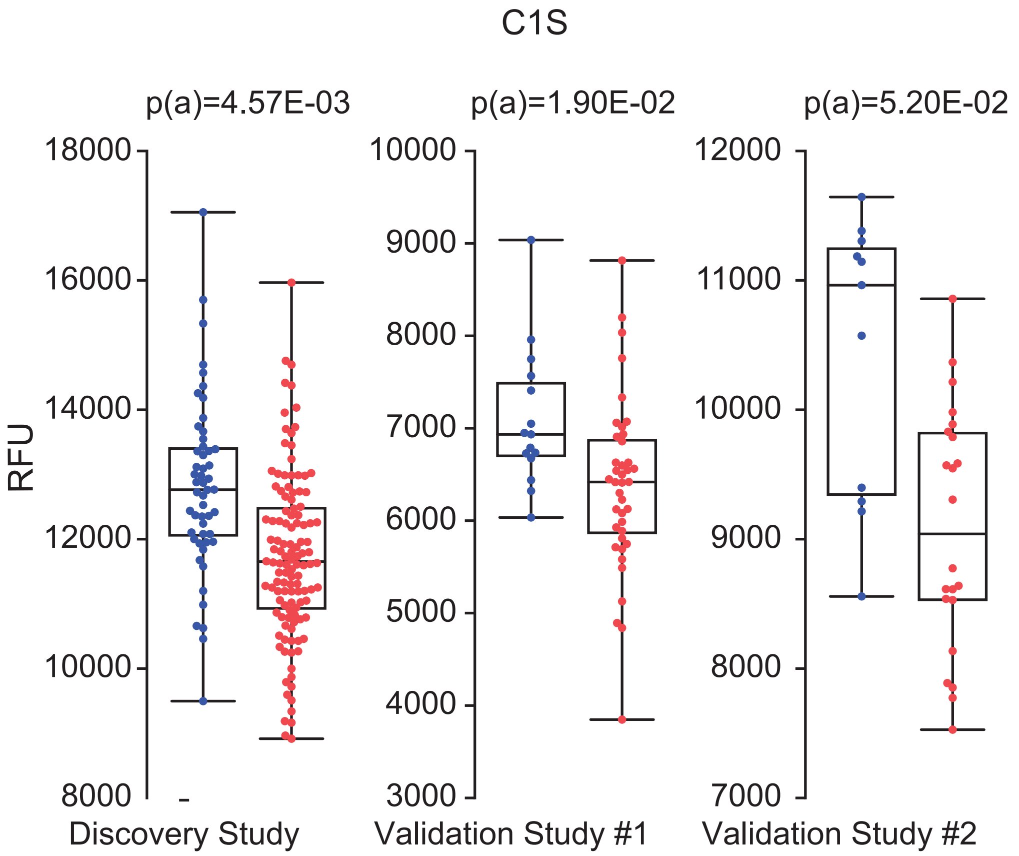


Figure 3 S1. Boxplots of raw data from all three studies for complement subunit C1S.

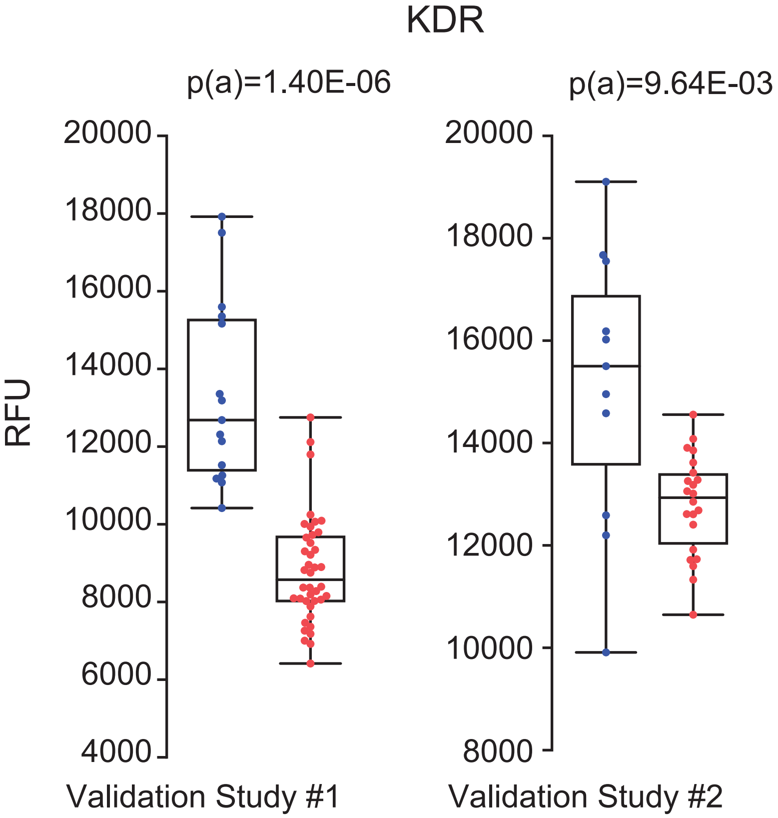


Figure 4 S1. Boxplots of raw data for KDR from each validation study.