# pQTLs on inflammation – findings from the SCALLOP consortium

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## Abstract

BACKGROUND. METHODS. FINDINGS. INTERPRETATION.

## Introduction

We report pQTLs associated with OLINK/INF1 panel, using cohorts in the SCAndinavian coLLaboration for Olink plasma Protein genetics (SCALLOP) consortium, https://www.olink.com/scallop/, a collaborative framework for discovery and follow-up of genetic associations with proteins on the Olink Proteomics platform.

The objectives of the study are as follows

* Identification of pQTLs in SCALLOP discovery cohorts
* Study of pQTLs in replication cohorts
* Investigation of the mechanistic basis of identified cis- and trans-pQTL by functional annotation
* Examination of pQTL pleiotropic effects
* Evaluation over the causal role of INF proteins disease outcomes such as CHD and stroke
* Other downstream analysis

## Data and analysis

### Proteins

The Olink INFlammation panel of 92 proteins, e.g, https://github.com/jinghuazhao/INF/blob/master/doc/olink.inf.panel.annot.tsv.

### SNPs

* 1000 genomes imputation, build 37 (hg19) positions.
* SNPs filtering on imputation quality at time of meta-analysis.
* Quality control on aspects such as SNP/sample call rates, gender mismatch, abnormal inbreeding coefficient, failed cryptic relatedness test, ancestry outlier, heterozygosity and Hardy-Weinberg equilibrium test.

### Association analysis

* Rank-based inverse normal transformation on the raw measurement of proteins including those below lower limit of detection, e.g., via invnormal function,

invnormal <- function(x) qnorm((rank(x,na.last="keep")-0.5)/sum(!is.na(x)))

* Multiple linear regression for all samples including sex, age, principal components and other cohort specific covariates.
* Additive genetic model
* For case-control data, cases and controls are analysed separately – results will be merged at meta-analysis stage

### Software

It is preferable to use software which account for genotype uncertainty, such as SNPTEST, QUICKTEST, and BOLT-LMM.

### SNP table for GWAS results

Please include the following columns. Missing values are coded as “NA”.

|  |  |  |
| --- | --- | --- |
| No | Variable name | Description of variable |
| 1 | SNPID | CHR:POS\_A1\_A2 (such that A1<A2) or rsid |
| 2 | CHR | Chromosome number |
| 3 | POS | Physical position for the reference sequence (please indicate NCBI build in descriptive file) |
| 4 | STRAND | Indicator of strand direction. Please specify “+” if positive or forward strand and “-” if negative or reverse strand. |
| 5 | N | Number of non-missing observations |
| 6 | EFFECT\_ALLELE | Allele for which the effect (beta coefficient) is reported. For example, in an A/G SNP in which AA = 0, AG=1, and GG=2, the coded allele is G. |
| 7 | REFERENCE\_ALLELE | Second allele at the SNP (the other allele). In the example above, the non-coded allele is A. |
| 8 | CODE\_ALL\_FQ | Allele frequency for the coded allele – “NA” if not available |
| 9 | BETA | Effect size for the coded allele, beta estimate from the genotype-phenotype association, with at least 5 decimal places. Note: if not available, please report “NA” for this variable. |
| 10 | SE | Standard error of the beta estimate, to at least 5 decimal places - “NA” if not available. |
| 11 | PVAL | p-value of Wald test statistic – “NA” if not available |
| 12 | RSQ | Residual phenotypic variance explained by SNP. “NA” if not available |
| 13 | RSQ\_IMP | Observed divided by expected variance for imputed allele dosage. |
| 14 | IMP | Please specify whether the SNP was imputed or genotyped: 1: imputed SNP, 0: directly genotyped SNP |

### File-naming convention

It is recommended to use format STUDY\_analyst\_inf1\_protein\_UniProtID\_date.gz, see https://www.uniprot.org/ for additional information on UniProt IDs.

### Notes on PLINK

Due possibly to the large number of proteins for GWAS, some cohorts employed PLINK to expedite analysis with which one may see the following information:

|  |  |  |  |
| --- | --- | --- | --- |
| No | Name | Description | Additional comment |
| 1 | BP | Position in base pairs |
| 2 | CHR | Chromosome |
| 3 | SNP | CHR:POS\_A1\_A2 or rsid |
| 4\* | HWE | Hardy-Weinberg equilibrium P-value |
| 5\* | MAF | Minor allele frequency | Please indicate if this is the effect allele frequency |
| 6 | A1 | Allele 1 | Please indicate if this is the effect/reference allele |
| 7\* | A2 | Allele 2 | Please indicate if this is the effect/reference allele |
| 8 | NMISS | Sample size |
| 9 | BETA | Regression coefficient |
| 10 | STAT | Regression test statistic |
| 11 | P | P value |

\* may be taken from the PLINK –hardy option and .bim file, see http://zzz.bwh.harvard.edu/plink/anal.shtml#glm.

In this case, please provide for each SNP information on strand, effect allele, effect allele frequency, and the information measures for imputation – the information measure can be on the genotype level obtained once for a cohort rather than from phenotype-genotype regression through software such as SNPTEST. SNP and sample based statistics can be greatly facilitated with software qctool, http://www.well.ox.ac.uk/~gav/qctool\_v2/. As is the case with INTERVAL.bgen and INTERVAL.sample, one can obtain the SNP-based statistics as follows,

qctool -g INTERVAL.bgen -s INTERVAL.sample -snp-stats -osnp INTERVAL.snp-stats -sample-stats -osample INTERVAL.sample-stats

See also the full SLURM sbatch script in the Appendix.

When a dosage format is used, PLINK also gives an INFO measure; see http://zzz.bwh.harvard.edu/plink/dosage.shtml.

## Meta-analysis

Meta-analysis will be performed centrally using the inverse-N weighted analysis of regression betas and standard errors, as implemented in the software METAL (https://github.com/statgen/METAL).

Genomic control and appropriate marker filters will be applied at this stage.

* **Marker exclusion filters**: we will apply imputation quality filters at the meta-analysis stage, so provide unfiltered results.
* **Genomic control (GC)**: genomic control will be applied at the meta-analysis stage (single GC), so GC-correction is not needed for each cohort.
* **Significance**: the Bonferroni threshold for the genome-wide analyses will be set at 5 x 10-10. The results will be replicated in independent cohorts.

## Contact information

For general questions about SCALLOP, please contact Anders Malarstig (anders.malarstig@ki.se). For technical issues about TRYGGVE, please contact Lasse Folkersen (lasfol@cbs.dtu.dk).

For questions regarding SCALLOP/INF, please contact Jing Hua Zhao (jhz22@medschl.cam.ac.uk) and James Peters ([jp549@medschl.cam.ac.uk](mailto:jp549@medschl.cam.ac.uk)).

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## References

Folkersen L, et al. (2017). Mapping of 79 loci for 83 plasma protein biomarkers in cardiovascular disease. PLoS Genetics 13(4), doi.org/10.1371/journal.pgen.1006706.

Kwan JSH, et al. (2014). Meta-analysis of genome-wide association studies identiﬁes two loci associated with circulating osteoprotegerin levels. Hum Mol Genet 23(24): 6684–669.

Sun BB, et al. (2018). Genomic atlas of the human plasma proteome. Nature 558: 73–79.

## Supplementary information

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