Analysis plan for GWAS of low muscle strength ("dynapenia") – 3rd August 2018

Garan Jones (gj286@exeter.ac.uk), Luke Pilling, David Melzer (University of Exeter)

On behalf of the CHARGE/GEFOS dynapenia GWAS working group

Rationale

Low grip strength (dynapenia, a component of sarcopenia) in older people is predictive of morbidity and mortality. A number of genome-wide association studies (GWAS) have discovered loci for musculoskeletal traits including maximum hand grip strength (1,2), lean body mass (3), and bone mineral density (4). However, none have investigated whether weakness (low grip strength / dynapenia) in older people has specific genetic susceptibility loci. It seems likely that the exceptionally low muscle strengths (dynapenia) seen in some older people may be influenced by specific genetic variants that differ from the influences on the normal distribution of strength. The former should be more influenced by aging factors, the latter by developmental and physical activity associated variants.

Hypothesis

Dynapenia is different to the "normal range" of functioning, especially in older people, and specific genetic loci are associated with low function.

Methods

GWAS meta-analysis of cohorts with both grip strength and genotype information in people aged 60+. Initial analysis to only include participants of European descent.

Additional trans-ethnic analysis to be considered based on numbers available (please provide this information)

Phenotype definitions

Baseline assessment should be used were possible. If the participant is younger than 60 at baseline then the first measurement when the participant is 60 or older should be used, in the case of multiple measurements over time.

Dynapenia (often referred to as sarcopenia) has two commonly used definitions: FNIH (Foundation National Institute of Health) (5) and EWGSOP (European Working Group on Sarcopenia in Older People) (6). Participants' age should be equal to or greater than 60 at time of Grip measurement.

FNIH: Grip strength < 26 Kg Men / < 16 Kg Women

EWGSOP: Grip strength < 30 Kg Men / < 20 Kg Women

Perform analyses using both the FNIH and EWGSOP grip definitions for comparison

Perform analyses in the whole sample (sex adjusted), and in males/females separately

Analysis set	Criteria
EWGSOP Both genders	Grip strength < 30 Kg Men / < 20 Kg Women; Age ≥ 60
EWGSOP Female only	Grip strength < 20 Kg Women only; Age ≥ 60
EWGSOP Male only	Grip strength < 30 Kg Men only; Age ≥ 60
FNIH Both genders	Grip strength < 26 Kg Men / < 16 Kg Women; Age ≥ 60
FNIH Female only	Grip strength < 16 Kg Women only; Age ≥ 60
FNIH Male only	Grip strength < 26 Kg Men only; Age ≥ 60

Quality control

Please include details of any QC steps applied to the genotyping prior to imputation. SNP QC on imputed data will be handled centrally, so upload of unfiltered data is preferred.

GWAS

We will use BOLT-LMM for GWAS of the UK Biobank (7), as the mixed-model approach mean we can include related participants in the analysis (suitable sample sizes > 5,000). We suggest using PLINK or SNPTEST if sample size is below 5,000, but individual studies may already have optimized workflows. Results files to include minor allele frequency and imputation quality of each variant. Please excluded or account for related participants in your analysis methods, as appropriate (and let us know).

Minimally adjust the models in the primary analysis phase: age, sex (where required), and technical covariates only – including population stratification e.g. PCs 1-10.

Model to run: Case/control~ SNP + age + sex + study specific covariates (Principal Components, study site, family structure)

Sensitivity analyses adjusting for height (etc.) can be performed later

Imputation

Please use the Haplotype reference consortium (HRC) v1.1 panel, if available. If not please advise us on the imputation panel used (see section 2.3.2, p.24 for methods used in UK Biobank imputation https://doi.org/10.1101/166298).

Include all 22 autosomal chromosomes. Include X / Y / MT chromosomes if data available (even if just directly genotyped i.e. not imputed).

Meta-Analysis

The METAL software (8) will be used to meta-analyse genetic variants across cohorts using fixed effect inverse-variance weighted approach.

Additional analyses

If numbers of participants from other ancestries great enough could also investigate trans-ethnic effects.

LD Score regression (9) will be used to investigate inflation/confounding in the GWAS results, to estimate the heritability of dynapenia, and also the genetic correlations with relevant traits, e.g. gait speed, lean mass, fat mass, height, etc.

Investigations of published candidates, especially in X, Y, and mitochondrial genomes.

Interpretation of GWAS results will include: DEPICT (10) analysis to identify independent signals and gene pathways, FUMA (11) analysis to investigate overlap with the GWAS catalog, and GTEx analysis of eQTLs to identify potential causal genes.

Follow-up analyses could include; utilizing cohorts where assessments of grip strength were made at multiple time points to model decline/prediction, and analysing overlap with muscle mass measures, where possible. Additional outcomes could include gait speed, balance tests, fall, fractures and medical records.

Sensitivity analyses could include adjustment for height, BMI, or lean mass.

Submission of results

Results should be sent for meta-analysis following the CHARGE protocol for result sharing.

http://depts.washington.edu/chargeco/wiki/ResultsSharing

http://depts.washington.edu/chargeco/wiki/ResultsSharingFormat

Details on the ShareSpace will be sent when the list of collaborators has been finalised.

Results should be named in the following format:

<cohort-name>_<phenotype>_<gender>_<submission-date>.txt.gz

- Date format should be YYYYMMDD.
- Gender can be **combined/male/female**
- Phenotype can be **ewgsop/fnih**
- For example; uk-biobank_ewgsop_combined_20181101.txt.gz

SNPs should be aligned with the forward (+) strand of the chromosome on HapMap.

Variable name	Description
SNPID	RS id number or "chr:position_effect-allele_noneffect-allele", for example 1:13289_CCT_C when an RSID is not available
chr	Chromosome number. X Chromosome should be coded as 23, Y chromosome as 24 and Mitochondrial as 25 (if available, see study description for more details)
position	Base position on GRCh.v.37
coded_all	Effect allele
noncoded_all	Alternate or non-effect allele
AF_coded_all	Effect allele frequency
strand_genome	+ or -, representing either the positive/forward strand or the negative/reverse strand of the human genome reference sequence; to clarify which strand the coded_all and noncoded_all are on
beta	Beta co-efficient from linear regression for the coded allele, at least 5 decimal places
lower_ci	Lower 95% confidence interval for outcome
upper_ci	Upper 95% confidence interval for outcome
odds_ratio	Point estimate of Odds-Ratio

se	Standard error of Beta estimate, to at least 5 decimal places – "NA" if not available
p_value	p-value of test statistic
HWE_pval	Exacy test Hardy-Weinberg equilibrium p-value – only directly typed SNPs, "NA" for imputed
callrate	genotyping callrate after exclusions
n_total	total sample with phenotype and genotype for SNP
imputed	1/0 coding; 1=imputed SNP, 0=if directly typed
used_for_imp	1/0 coding; 1=used for imputation, 0=not used for imputation
oevar_imp	Imputation quality (observed divided by expected variance for imputed allele dosage). Please report r2hat value for minimac and proper_info for IMPUTE2.

Cohorts included (so far)

- AGES
- BLSA
- CHS
- FHS
- Health ABC
- HRS
- InCHIANTI
- KORA
- LASA
- PLSAW
- Rotterdam studies
- SHIP
- UK Biobank
- WLS

Please fill in cohort details using the below Google Document (includes number of participants, methods for genotyping and grip strength measurement, etc.) https://docs.google.com/spreadsheets/d/1_IjJoumI1Af5KsQp0twdacfMX6mho63HRdgxZ7sMpjQ/edit?usp=sharing

References

1. Willems SM, Wright DJ, Day FR, Trajanoska K, Joshi PK, Morris JA, et al. Large-scale GWAS identifies multiple loci for hand grip strength providing biological insights into muscular fitness. Nat Commun. 2017 Jul;8(May):16015.

2. Matteini AM, Tanaka T, Karasik D, Atzmon G, Chou W-CC, Eicher JD, et al. GWAS analysis of handgrip and lower body strength in older adults in the CHARGE consortium. Aging Cell. 2016;15(5):792–800.

3. Zillikens MC, Demissie S, Hsu Y-H, Yerges-Armstrong LM, Chou W-C, Stolk L, et al. Large meta-

analysis of genome-wide association studies identifies five loci for lean body mass. Nat Commun. 2017;8(1):80.

4. Medina-Gomez C, Kemp JP, Trajanoska K, Luan J, Chesi A, Ahluwalia TS, et al. Life-Course Genome-wide Association Study Meta-analysis of Total Body BMD and Assessment of Age-Specific Effects. Am J Hum Genet. 2018;102(1):88–102.

5. McLean RR, Shardell MD, Alley DE, Cawthon PM, Fragala MS, Harris TB, et al. Criteria for Clinically Relevant Weakness and Low Lean Mass and Their Longitudinal Association With Incident Mobility Impairment and Mortality: The Foundation for the National Institutes of Health (FNIH) Sarcopenia Project. Journals Gerontol Ser A [Internet]. 2014 May 1 [cited 2018 Jun 11];69(5):576–83. Available from: https://academic.oup.com/biomedgerontology/article-lookup/doi/10.1093/gerona/glu012

6. Cruz-Jentoft a J, Baeyens JP, Bauer JM, Boirie Y, Cederholm T, Landi F, et al. Sarcopenia: European consensus on definition and diagnosis: Report of the European Working Group on Sarcopenia in Older People. Age Ageing. 2010;39(4):412–23.

7. Loh P-R, Tucker G, Bulik-Sullivan BK, Vilhjálmsson BJ, Finucane HK, Salem RM, et al. Efficient Bayesian mixed-model analysis increases association power in large cohorts. Nat Genet [Internet]. 2015 Feb 2 [cited 2018 Aug 1];47(3):284–90. Available from: http://www.nature.com/doifinder/10.1038/ng.3190

8. Willer CJ, Li Y, Abecasis GR. METAL: fast and efficient meta-analysis of genomewide association scans. Bioinformatics. 2010 Sep;26(17):2190–1.

9. Bulik-Sullivan BK, Loh P-R, Finucane HK, Ripke S, Yang J, Consortium SWG of the PG, et al. LD Score regression distinguishes confounding from polygenicity in genome-wide association studies. Nat Genet. 2015;advance on(3):291–5.

10. Pers TH, Karjalainen JM, Chan Y, Westra H-J, Wood AR, Yang J, et al. Biological interpretation of genome-wide association studies using predicted gene functions. Nat Commun. 2015 Jan;6:5890.

11. Watanabe K, Taskesen E, Bochoven A van, Posthuma D. FUMA: Functional mapping and annotation of genetic associations. bioRxiv. 2017;110023.