BLUPf90 & PreGS and Quality Control

PreGSf90

- Interface program to the genomic module to process the genomic information for the BLUPF90 family of programs

- Efficient methods
  - creation of the genomic relationship matrix, relationship based on pedigree
  - Inverse of relationship matrices

- Performs Quality Control of SNP information

BLUPF90 programs using Genomic

- Genomic programs
  - controled by adding OPTIONS commands to the parameter file
  - `OPTION SNP_file` *`marker.geno.clean`*
  - Read 2 files:
    - `marker.geno.clean`
    - `marker.geno.clean_XrefID`

Output Files

- GimA22i
  - Store the content of the inv(G) – inv(A22)
  - Only if preGSf90 for runs, not in applications programs

- freqdata.count
  - Contains the estimated allele frequency before QC

- freqdata.count.after.clean
  - Contains allele frequencies as used in calculations, remove code
  - For removed SNP these will be zero

- Gen_call_rate
  - List of animals removed by low call rate

- Gen_conflicts
  - Report of animals with Mendelian conflicts

Quality control
By default exclude

- MAF
  - SNP with MAF < 0.05
- Call rate
  - SNP with call rate < 0.90
  - Individuals with call rate < 0.90
- Monomorphic
  - Exclude monomorphic SNP. ONLY when MAF <> 0

Quality control
By default exclude (cont)

- Parent-progeny conflicts (SNP & Individuals)
  - Exclusion -> opposite homozygous
  - For SNP: >10 % of parent-progeny exclusion from the total of pairs evaluated
  - For Individuals: > 1% of parent-progeny from total number of SNP

Control default values

- For MAF
  - OPTION minfreq x
- Call rate
  - OPTION callrate x
  - OPTION callrateAnim x
- Mendelian conflicts
  - OPTION exclusion_threshold x
  - OPTION exclusion_threshold_snp x

Parent-progeny conflicts

- Presence of these conflicts results in a negative H matrix !!!
- Problems in estimation of variance component by REML, programs do not converge, etc.
- Solution:
  - Report all conflicts, with counts for each individual as parent or progeny to trace the conflicts
  - Remove progeny genotype
    - maybe not the best option
    - But results in a positive-definite H matrix !!!

Parent-progeny conflicts

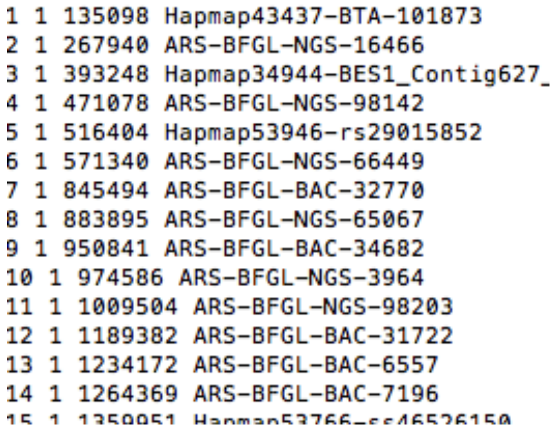
- OPTION verify_parentage x
  - 0: no action
  - 1: only detect
  - 2: detect and search for an alternate parent; no change to any file. Not implemented
    - implemented **in seekparentf90** program
  - 3: detect and eliminate progenies with conflicts (default)

Other Options

- Exclusion of selected chromosomes:
  - OPTION excludeCHR n1 n2 n3 ...
- Inclusion of selected chromosomes:
  - OPTION includeCHR n1 n2 n3 ...
- Exclude samples from analyses
  - OPTION excludeSample n1 n2 n3
- Inform which are sex chromosomes:
  - OPTION sex_chr n
  - Chromosome # > n will be excluded only for HWE and parent-progeny checks, but not in calculations

SNP map file

- OPTION chrinfo *<file>*
- For some genomic analyses (GWAS) or QC
- Format:
  - SNP number
    - Index number of SNP in the sorted map by chromosome and position
  - chromosome number
  - Position
  - SNP name (Optional)



```
1 1 135098 Hapmap43437-BTA-101873
2 1 267940 ARS-BFGL-NGS-16466
3 1 393248 Hapmap34944-BES1_Contig627_
4 1 471078 ARS-BFGL-NGS-98142
5 1 516404 Hapmap53946-rs29015852
6 1 571340 ARS-BFGL-NGS-66449
7 1 845494 ARS-BFGL-BAC-32770
8 1 883895 ARS-BFGL-NGS-65067
9 1 950841 ARS-BFGL-BAC-34682
10 1 974586 ARS-BFGL-NGS-3964
11 1 1009504 ARS-BFGL-NGS-98203
12 1 1189382 ARS-BFGL-BAC-31722
13 1 1234172 ARS-BFGL-BAC-6557
14 1 1264369 ARS-BFGL-BAC-7196
15 1 1359951 Hapmap53766-ss46526150
```

- First column corresponds to first row SNP in genotype file !!!

Saving ‘clean’ files

- SNP excluded from QC are set as missing (i.e. Code=5)
- Excluded Individuals are treated as unrealated in G and A22
  - For individual *i*

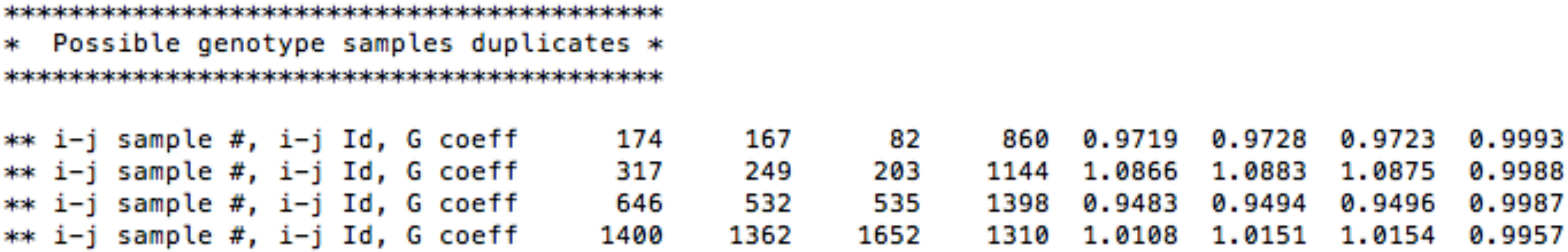
    G[i,:] = 0; G[:,i]=0; G[i,i]=1 ;   Same for A22

    so G-A22 will cancel out

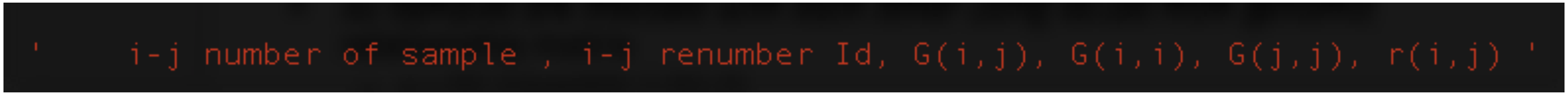
- OPTION saveCleanSNPs
- Save clean genotype data with excluded SNP and individuals
  - For example for a SNP_file *gt*
  - Clean fles will be:
    - *gt*_clean
    - *gt*_clean_XrefID
  - Removed will be output in files:
    - *gt*_SNPs_removed
    - *gt*_Animals_removed

Potential duplicate samples

- All samples are checked with each other using values from genomic relationship matrix
  - x = G(i,j)/sqrt(G(i,i),G(j,j))
  - Values of x > 0.90 are printed in the output



```
*****************************************
*  Possible genotype samples duplicates *
*****************************************

** i-j sample #, i-j Id, G coeff      174     167      82     860  0.9719  0.9728  0.9723  0.9993
** i-j sample #, i-j Id, G coeff      317     249     203    1144  1.0866  1.0883  1.0875  0.9988
** i-j sample #, i-j Id, G coeff      646     532     535    1398  0.9483  0.9494  0.9496  0.9987
** i-j sample #, i-j Id, G coeff     1400    1362    1652    1310  1.0108  1.0151  1.0154  0.9957
```

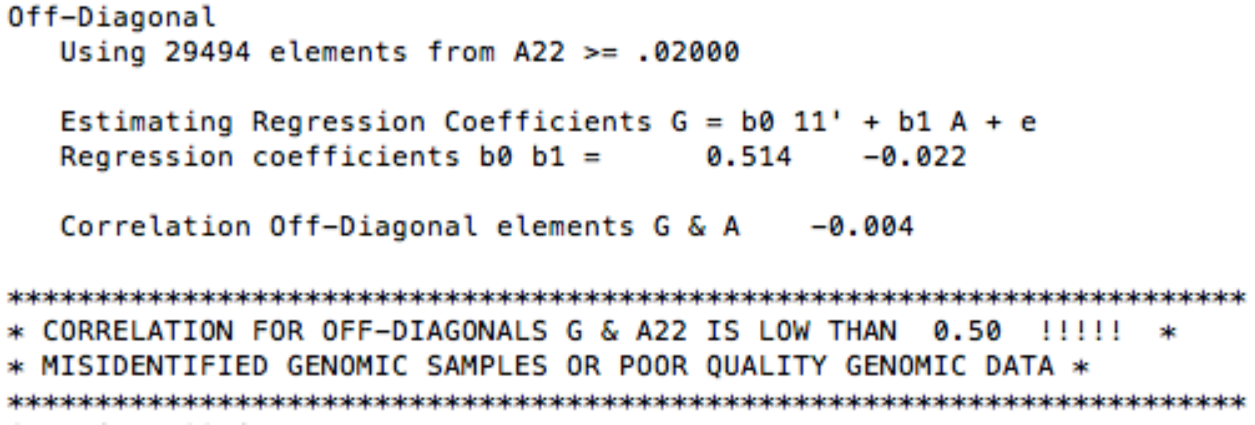


```
'    i-j number of sample , i-j renumber Id, G(i,j), G(i,i), G(j,j), r(i,j) '
```

- Threshold to identify potential duplicates
  - OPTION threshold_duplicate_samples x
- Exclude specific samples
  - OPTION excludeSample n1 n2….

Correlation off-diagonal G vs A

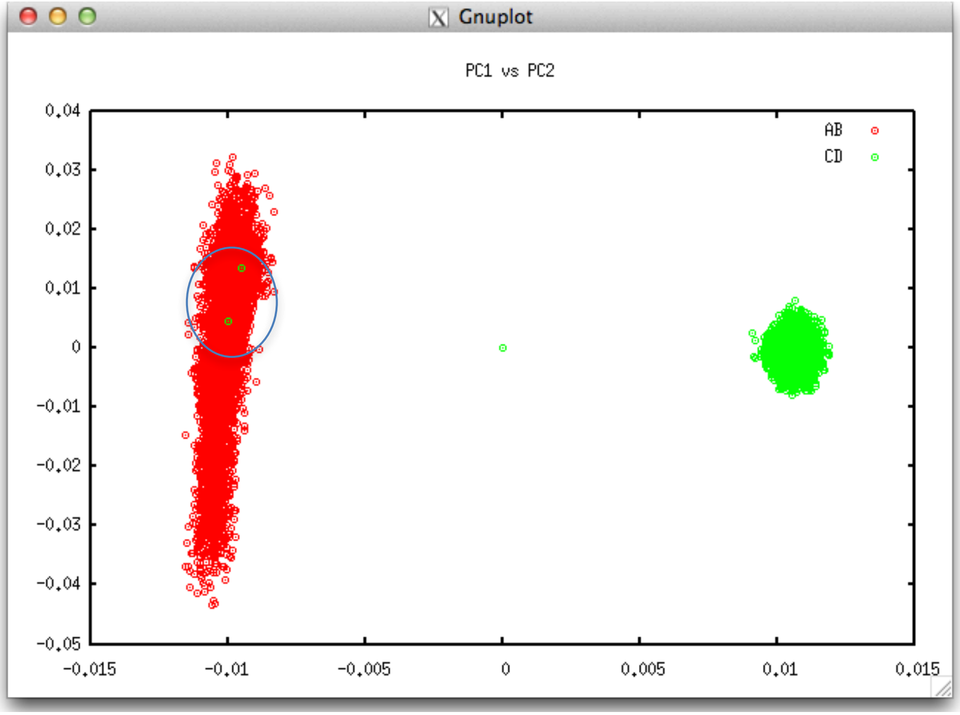
- Compute correlation for all elements of A > 0.02
- Potential problems with matching genotype and pedigree files
- For low values (<0.5) => print a warning !!!!
- For low values (<0.3) => program stop !!!
- If still you want to go …
  - OPTION thrStopCorAG -1

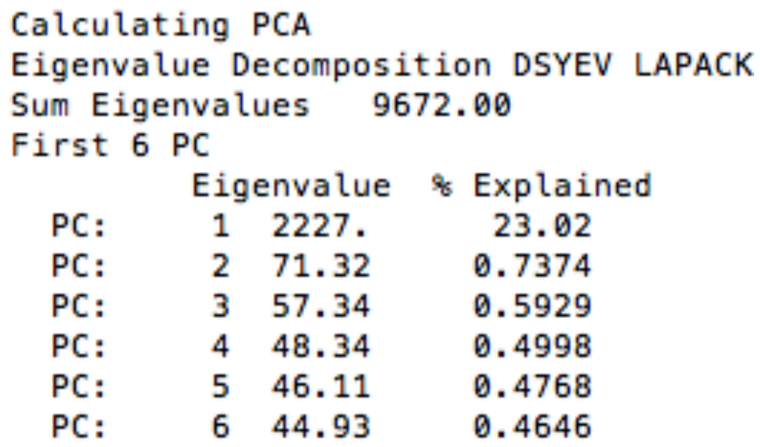


```
Off-Diagonal
   Using 29494 elements from A22 >= .02000

   Estimating Regression Coefficients G = b0 11' + b1 A + e
   Regression coefficients b0 b1 =      0.514    -0.022

   Correlation Off-Diagonal elements G & A    -0.004

***********************************************************************
* CORRELATION FOR OFF-DIAGONALS G & A22 IS LOW THAN  0.50  !!!!!  *
* MISIDENTIFIED GENOMIC SAMPLES OR POOR QUALITY GENOMIC DATA *
***********************************************************************
```

Looking for stratification in populations

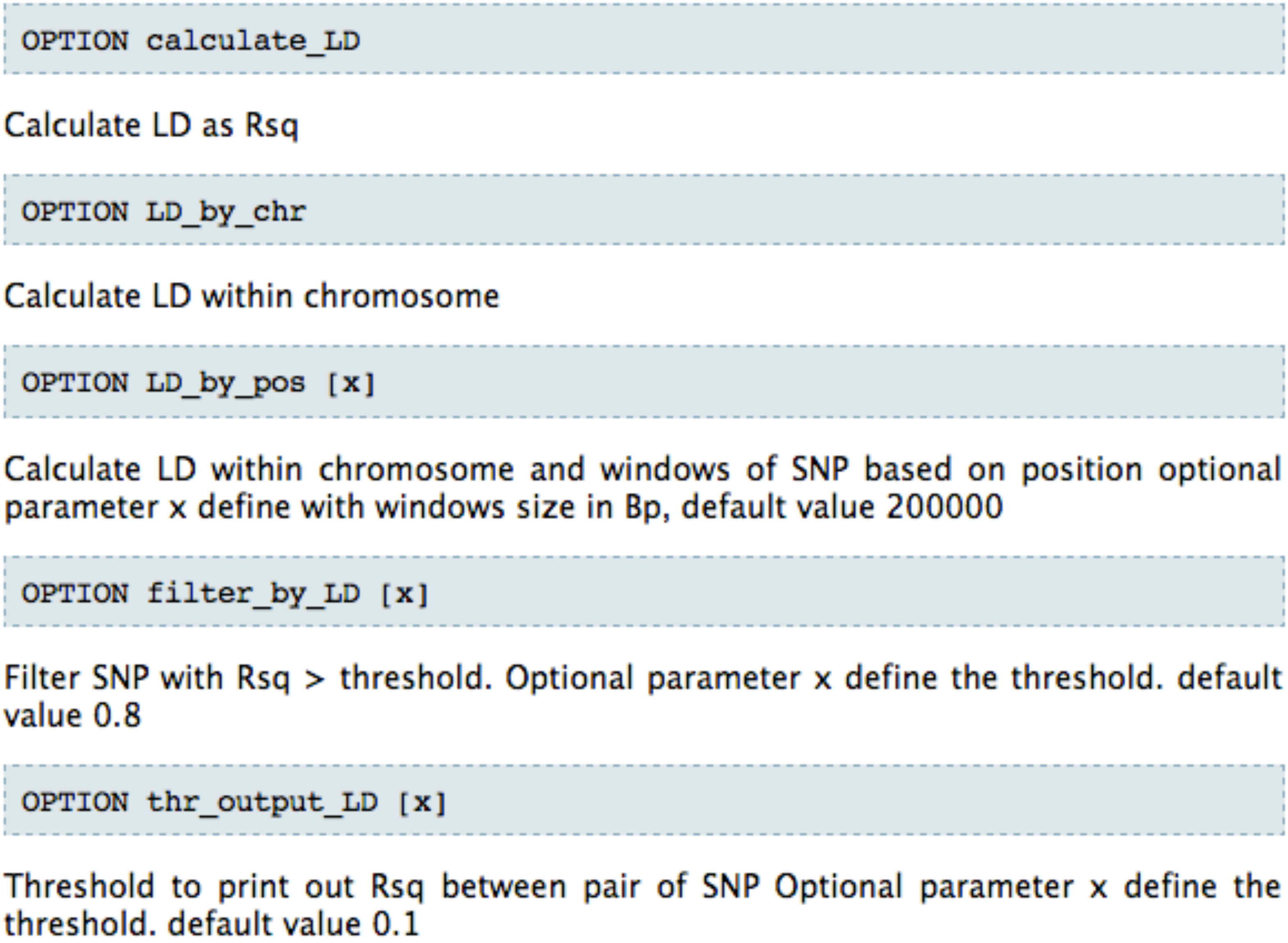
- OPTION plotpca
  - (only preGSf90 not in application programs)
  - Plot the first 2 PC
- OPTION extra_info_pca *filename col*
  - File with variables (alphanumeric) to plot PC with different colors for different classes
  - Same order as genotype file





```
Calculating PCA
Eigenvalue Decomposition DSYEV LAPACK
Sum Eigenvalues    9672.00
First 6 PC
             Eigenvalue   % Explained
  PC:        1   2227.        23.02
  PC:        2   71.32       0.7374
  PC:        3   57.34       0.5929
  PC:        4   48.34       0.4998
  PC:        5   46.11       0.4768
  PC:        6   44.93       0.4646
```

LD calculation and options



```
OPTION calculate_LD
```

Calculate LD as Rsq

```
OPTION LD_by_chr
```

Calculate LD within chromosome

```
OPTION LD_by_pos [x]
```

Calculate LD within chromosome and windows of SNP based on position optional parameter x define with windows size in Bp, default value 200000

```
OPTION filter_by_LD [x]
```

Filter SNP with Rsq > threshold. Optional parameter x define the threshold. default value 0.8

```
OPTION thr_output_LD [x]
```

Threshold to print out Rsq between pair of SNP Optional parameter x define the threshold. default value 0.1

preGSf90 -Only Quality control

Shortcut…

```
OPTION SNP_file snp.dat
OPTION chrinfo angus_map
OPTION excludeCHR 30 31 32
OPTION saveCleanSNPs
OPTION createG 0
OPTION createGInverse 0
OPTION createA22 0
OPTION createA22Inverse 0
OPTION createGimA22i 0
```

No Quality control

- ONLY use:
  - If QC was performed in a previous run
  - and “clean” genotype file is used

- OPTION no_quality_control

Memory requirement

- Slow operations for quality control in PREGSF90
  - All data stored in memory as double precision
  - Designed for the computation of G-matrix
  - Required memory for 60k SNPs and

    500k genotyped animals = 224GB

Comparison preGSf90 and QCF90

- Holstein genotypes
  - Number of genotypes: 569,404
  - Number of SNP markers: 60,671
  - Number of Pedigree animals: 10,710,380
- Programs
  - QCF90: with pre-renumbered files
  - PREGSF90: with post-renumbered files

Masuda, 2017

QCF90: benchmark results

Step	QCF90 (sec.)	PREGSF90 (sec.)
Reading a SNP file	420	1407
MAF and call rate	150	245
HWE test	84	24
Call rate for animals	3	307
Mendelian tests for SNP	62	316
Mendelian tests for animals	62	248
Recalculation of MAF	136	161
Total	917	2708
Memory usage	9 GB	257 GB

Masuda, 2017