Package 'NAM'

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Description Designed for association studies in nested association mapping (NAM) panels, also handling biparental and random panels. It includes functions for genome-wide associations mapping of multiple populations, marker quality control, solving mixed models and finding variance components through REML and Gibbs sampling.

License GPL-3

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NAM-package *Nested Association Mapping Analysis*

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See Also

Functions: gibbs, reml, Fst, gwas/gwas2, reference, snpQC, snpH2, cleanREP, wgr

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BMM *Bayesian Mixed Model*

Description

Univariate mixed model solver through Bayesian Gibbs Sampling or iterative solution.

Usage

```
gibbs(y,Z=NULL,X=NULL,iK=NULL,iR=NULL,Iter=1500,Burn=500,Thin=4,DF=5,S=1,GSRU=FALSE)
ml(y,Z=NULL,X=NULL,iK=NULL,iR=NULL,DF=-2,S=0)
```

Arguments

y	Numeric vector of observations (n) describing the trait to be analyzed. NA is allowed.
Z	Right-hand side formula or list of numeric matrices (n by p) with incidence matrices for random effect. NA is not allowed.
X	Right-hand side formula or incidence matrices (n by p) for fixed effect. NA is not allowed.
iK	Numeric matrix or list of numeric matrices (p by p) corresponding to the the inverse kinship matrix of each random effect with p parameters.
iR	Numeric matrix (n by n) corresponding to the inverse residual correlation matrix.
Iter	Integer. Number of iterations or samples to be generated.
Burn	Integer. Number of iterations or samples to be discarted.
Thin	Integer. Thinning parameter, used to save memory by storing only one every 'Thin' samples.
DF	Integer. Hyperprior degrees of freedom of variance components.
S	Integer or NULL. Hyperprior shape of variance components. If NULL, the hyperprior solution for the scale parameter is calculated as proposed by de los Campos et al. (2013).
GSRU	Logical. If TRUE, it updates the regression coefficient using Gauss-Seidel Residual Update (Legarra and Misztal 2008). Useful for p»n, but does not work when iK or iR are provided.

Details

Solve Gaussian mixed models in the Bayesian framework as described by Garcia-Cortes and Sorensen (1996) and Sorensen and Gianola, D. (2002) with conjugated priors. The alternative, ml, finds the solution iteratively using the full-conditional expectation.

Value

The function gibbs returns a list with variance components distribution a posteriori (Posterior.VC) and mode estimated (VC.estimate), a list with the posterior distribution of regression coefficients (Posterior.Coef) and the posterior mean (Coef.estimate), and the fitted values using the mean (Fit.mean) of posterior coefficients.

Author(s)

Alencar Xavier

References

de los Campos, G., Hickey, J. M., Pong-Wong, R., Daetwyler, H. D., and Calus, M. P. (2013). Whole-genome regression and prediction methods applied to plant and animal breeding. Genetics, 193(2), 327-345.

Legarra, A., & Misztal, I. (2008). Technical note: Computing strategies in genome-wide selection. Journal of dairy science, 91(1), 360-366.

Garcia-Cortes, L. A., and Sorensen, D. (1996). On a multivariate implementation of the Gibbs sampler. Genetics Selection Evolution, 28(1), 121-126.

Sorensen, D., & Gianola, D. (2002). Likelihood, Bayesian, and MCMC methods in quantitative genetics. Springer Science & Business Media.

Examples

```
# Fitting Mixed Model
data(tpod)
S = seq(1,350,50)
test1=gibbs(y,X=~factor(fam),Z=gen[,S],S=0.5)
plot(test1)

# Fitting GBLUP
K = tcrossprod(gen)
K = K/mean(diag(K))
iK = chol2inv(K)
test2=gibbs(y,iK=iK)
plot(test2)

# Fitting RKHS
E = dist(gen)
G = exp(-E^2/mean(E^2))
EIG = eigen(G,symmetric = TRUE)
ev = 20
U = EIG$vectors[,1:ev]
iV = diag(1/EIG$values[1:ev])
test3=gibbs(y,Z=U,iK=iV,S=NULL)
plot(test3)
```

Dataset *Tetra-seed Pods*

Description

Two biparental crosses phenotyped for the percentage of pods containing four seeds

Usage

```
data(tpod)
```

Details

Soybean nested association panel with 2 families (fam) containing 196 indiviuals. Genotypic matrix (gen) have 376 SNP across 20 chromosome (chr). Phenotypic information (y) regards the proportion of tetra-seed pods. Data provided by Rainey Lab for Soybean Breeding and Genetics, Purdue University.

Author(s)

Alencar Xavier and Katy Rainey

`Duplicates` *Genotype Duplicates*

Description

Identify and merge duplicate genotypes

Usage

```
cleanREP(y,fam,gen,thr=0.95)
```

Arguments

`y`	Numeric vector (n) or numeric matrix (nxt) of observations describing the trait to be analyzed. `NA` is allowed.
`fam`	Numeric vector of length (n) indicating which subpopulations ($i.e.$ family) each observation comes from. Default assumes that all observations are from the same populations.
`gen`	Numeric matrix containing the genotypic data. A matrix with n rows of observations and (m) columns of molecular markers. SNPs must be coded as `0, 1, 2`, for founder homozygous, heterozygous and reference homozigous. `NA` not allowed.
`thr`	Threshold above which genotypes are considered identical. Default is 0.95, merging genotypes >95

Details

The algorithm start from generating a genomic identity matrix (IBS), with pairwise percentage of identical loci among individuals. Individuals above the threshold have the phenotypes merged while keeping only one genotype.

Value

List containing the inputs without replicates. Groups of replicates are replaced by a single observation with the phenotypic expected value. The algorithm keeps the genotypic information of the first individual (genotypic matrix order).

Author(s)

Alencar Xavier

Examples

```
data(tpod)
test = cleanREP(y,fam,gen)
```

Fst *Fixation Index*

Description

Genetic variation associated with markers distributed among subpopulations. The function generates a plot for structure diagnosis.

Usage

```
Fst(gen,fam)
```

Arguments

gen	Numeric matrix containing the genotypic data. A matrix with n rows of observations and (m) columns of molecular markers. SNPs must be coded as `0, 1, 2`, for founder homozigous, heterozygous and reference homozygous. `NA` is allowed.
fam	Numeric vector of length (n) indicating which subpopulations ($i.e.$ family) each observation comes from. `NA` is not allowed.

Details

F-statistics (Wright 1965) represent the differentiation among populations for a given locus. Weir and Cockerham (1984) provided an unbiased version for molecular analysis.

FIT is the correlation between gametes that unite to produce the individuals, relative to the gametes of the total population. FIS is the average over all subdivisions of the correlation between uniting gametes relative to those of their own subdivision. FST is the correlation between random gametes within subdivisions, relative to gametes of the total population. Neutral markers have an expected FST 0.05.

Value

List with values of FST, FIS and FIT. Unbiased F-statistics from weighted AOV (Weir and Cockerham 1984).

Author(s)

Alencar Xavier and William Muir

References

Weir, B. S., and Cockerham, C. C. (1984). Estimating F-statistics for the analysis of population structure. Evolution, 38(6), 1358-1370.

Wright, S. (1965). The interpretation of population structure by F-statistics with special regard to systems of mating. Evolution, 19(3), 395-420.

Examples

```
data(tpod)
Fstat = Fst(gen=gen,fam=fam)
plot(Fstat,p=0.05,chr=chr)
```

GWAS *Empirical Bayes Genome Wide Association Mapping*

Description

The gwas function calculates the likelihood ratio for each marker under the empirical Bayesian framework. The method also works with multiple populations.

Usage

```
gwas(y,gen,fam=NULL,chr=NULL,window=NULL,fixed=FALSE)
gwas2(y,gen,fam=NULL,chr=NULL,fixed=FALSE,EIG=NULL,cov=NULL)
gwasGE(Phe,gen,fam,chr=NULL,cov=NULL)
```

Arguments

y	Numeric vector of observations (n) describing the trait to be analyzed. NA is allowed.
gen	Numeric matrix containing the genotypic data. A matrix with n rows of observations and (m) columns of molecular markers. SNPs must be coded as 0, 1, 2, for founder homozygous, heterozygous and reference homozigous. NA is allowed.
fam	Numeric vector of length n indicating which subpopulations ($i.e.$ family) each observation comes from. Default assumes that all observations are from the same populations.
chr	Numeric vector indicating the number of markers in each chromosome. The sum of chr must be equal to the number of columns in gen. Default assumes that all markers are from the same chromosome.
window	Numeric. If specified, genetic distance between markers is used for moving window strategy (Wang 2015). Window must be specified in Morgans ($e.g.$ 0.05 would represent 5cM). Genetic distance is calculated assuming that individuals are RILs.

fixed	Logical. If TRUE, markers are treated as fixed effect and hence, evaluated through Wald statistics. If markers are specief as fixed, the argument 'window' is not applicable.
EIG	Output of the R function 'eigen'. It is used for user-defined kinship matrix.
cov	Numeric vector of length n to be used as covariate in the association analysis.
Phe	Numeric matrix of observations ($n * e$) where rows represent genotypes and columns represent environments. NA is allowed.

Details

Special incidence matrix is recreated to optimize the information provided by the subpopulations. Each locus is recoded as a vector with length f equal to number of subpopulations, or NAM families, as the interaction locus by family. For example, a locus heterozigous from an individual from subpopulation 2 is coded as [1, 0, 1 , ... ,f], a locus homozigous for the reference allele from any subpopulation is coded as [2, 0, 0, ... , f] and a locus homozigous for the founder allele from an individual from subpopulation 1 is coded as [0, 2, 0, ... ,f].

The base model for genome scanning includes the fixed effect (Xb), the marker (Zu), the polygene (g) and the residuals (e). If the $window$ term is specified, the model for genome scanning includes three extra terms, the left side genome ($Zu[k-1]$), the right side genome ($Zu[k+1]$) and window polygene ($-g[k]$). The genetic distance between markers is computed using Kosambi map function, where the recombination rate is estimated under the assuption that genotypes are recombinant inbred lines.

The polygenic term is calculated only once (Zhang et al 2010) using eigendecomposition with a GEMMA-like algorithm (Zhou ans Stephens 2012). Efficient inversion of capacitance matrix is obtained through the Woodbury matrix identities. Models and algorithms are described with more detail by Xavier et al (2015).

In order to analyze large dateset, one can avoid memory issues by using the function "gwas2", but that the argument 'window' is not implemented for "gwas2". This function also allows used-defined kindship through the argument EIG, and the use of a numeric covariate vector through the argument cov.

Value

The function gwas returns a list containing the method deployed ($Method$), a summary of predicted parameters and statistical tests ($PolyTest$), estimated genetic map for NAM panels (MAP) and the marker names ($SNPs$).

Author(s)

Alencar Xavier, Tiago Pimenta, Qishan Wang and Shizhong Xu

References

Wang, Q. An Empirical Bayes Method for Genome-Wide Association Studies. W799/Statistical Genomics. PAG XXXII. 2015.

Xavier, A., Xu, S., Muir, W. M., & Rainey, K. M. (2015). NAM: Association Studies in Multiple Populations. Bioinformatics, btv448.

Zhang et al. 2010. Mixed linear model approach adapted for genome-wide association studies. Nat. Genet. 42:355-360.

Zhou, X., & Stephens, M. (2012). Genome-wide efficient mixed-model analysis for association studies. Nature genetics, 44(7), 821-824.

Examples

```
data(tpod)
test=gwas(y=y,gen=gen[,1:240],fam=fam,chr=chr[1:12])
par(mfrow=c(2,1))

# Example Manhattan 1
SIGNIF = 1+(2*test$PolyTest$lrt>4.9)
plot(x=test,pch=SIGNIF+3,lwd=SIGNIF,main="Example 2")

# Example Manhattan 2
plot(x=test,main="Example 3",pch=20,lwd=2)
Kern = ksmooth(1:240,test$PolyTest$lrt,bandwidth=1)
lines(Kern,type="l",lwd=2)
```

Internals *Internal functions*

Description

Complimentary statistics and functions written in C++ to speed up $gwas$, $gibbs$ and wgr.

Author(s)

Alencar Xavier and Tiago Pimenta

Examples

```
# Forward gen imputation
data(tpod)
fast.impute.gen = markov(gen,chr)

# A matrix
PedMat()

# Pairwise LD
ld = LD(gen[,1:3])
ld = LD(gen[,1:10])
heatmap(ld$r2)

# Spatial correlation kernel
covar()

# Genetic distance
```

```
round(Gdist(gen[1:10,],method=1),2)

# PCs of a NAM kinship
eG = eigX(gen,fam)
plot(eG[[2]],col=fam)
```

Manhattan *Manhattan plot for Association Studies*

Description

Generates a graphical visualization for the output of the function gwas/gwas2.

Usage

```
## S3 method for class 'NAM'
plot(x,...,alpha=0.05,colA=2,colB=4,find=NULL,FDR=NULL,gtz=FALSE,phys=NULL)
```

Arguments

x	Output of the gwas/gwas2 function.
...	Further arguments passed to or from other methods.
alpha	Numberic. Significance threshold to display in the Manhattan plot.
colA	Color of odd chromosomes in the Manhattan plot.
colB	Color of even chromosomes in the Manhattan plot.
find	Integer. If provided, you can click on the specified number of hits in the Manhattan plot to obtain the name of the markers.
FDR	Null or numeric between zero and one. If provided, it will display the Manhattan plot with Bonferroni threshold by chromosome, adjusted for the specified false discovery rate (FDR). Thus, zero provides the Bonferroni correction.
gtz	Logical. If TRUE, the argument FDR will just take into account markers with p-value Greater Than Zero (GTZ).
phys	Numeric vector with length equal to the number of markers. If provided, the Manhattan plot is generated using the physical distance in the x axis.

Author(s)

Alencar Xavier and William Beavis

Examples

```
data(tpod)
test=gwas2(y=y,gen=gen,fam=fam,chr=chr)

par(mfrow=c(2,1))

# Example Manhattan 1
plot(x=test,colA=3,colB=1,alpha=0.05/ncol(gen),type="h",main="Example 1: Genome-Wide Bonferroni")

# Example Manhattan 2
Title = "Example 2: FDR 0.25 by Chromosome"
plot(x=test,alpha=0.05,FDR=0.25,gtz=TRUE,pch=20,main=Title)
```

Reference *Changing the Reference Genotype*

Description

Function changes the reference genotype. For NAM populations, this function must be used when genotypes are coded according to the reference genome instead of the standard parent.

Usage

```
reference(gen, ref = NULL)
```

Arguments

`gen`	Numeric matrix containing the genotypic data. A matrix with n rows of observations and (m) columns of molecular markers. SNPs must be coded as `0, 1, 2`.
`ref`	Numeric vector of length n with elements coded as `0, 1, 2`, it represents the genotypic information of a new reference genotype. Default assumes that more frequent allele represents the reference genome.

Details

If genotypes are coded based on the refence genome, NAM analysis are optimized by using the standard parent as reference to allele coding.

Value

Returns a recoded gen matrix

Author(s)

Alencar Xavier

Examples

```
data(tpod)
gen=reference(gen=gen,ref=NULL)
```

REML *Restricted Maximum Likelihood*

Description

Univariate REML estimators and variance components for a single random variable fitted by an EMMA-like algorithm.

Usage

```
reml(y,X=NULL,Z=NULL,K=NULL)
MCreml(y,K,X=NULL,MC=300,samp=300)
```

Arguments

y	Numeric vector of observations (n) describing the trait to be analyzed. NA is allowed.
X	Formula or incidence matrix (n by p) for fixed effect. NA is not allowed.
Z	Formula or numeric matrix (n by p) that corresponds to the incidence matrix of random effect. NA is not allowed.
K	Numeric matrix (p by p). Kinship matrix for random effect with p parameters. NA is not allowed.
MC	Number of sampling procedures to estimate variance components using MCreml.
samp	Sample size of the sampling procedure to estimate variance components using MCreml.

Details

Solve mixed models with a single random effects minizing the log restricted maximum likelihood (REML) using the EMMA algorithm (Kang et al 2008). Prediction of random coefficients are performed according to VanRaden (2008). If y is a matrix with multiple traits, the fuctions solves the mixed model via an ECM algorithm adapted from the EMMREML package (Akdemir and Godfrey 2014). MCreml is a resampling strategy to find variance components for large datasets.

Value

The function reml returns a list with variance components and heritability (VC), fixed effect coefficients and standard variations (Fixed) and estimated breeding values (EBV).

Author(s)

Alencar Xavier, Tiago Pimenta and Shizhong Xu

References

Akdemir, D., and O. U. Godfrey (2014) EMMREML: Fitting Mixed Models with Known Covariance Structures. R Package Version 2.0. Available at: http://CRAN.R-project.org/package=EMMREML.

Kang, H. M., Zaitlen, N. A., Wade, C. M., Kirby, A., Heckerman, D., Daly, M. J., & Eskin, E. (2008). Efficient control of population structure in model organism association mapping. Genetics, 178(3), 1709-1723.

VanRaden, P. M. (2008). Efficient methods to compute genomic predictions. Journal of dairy science, 91(11), 4414-4423.

Examples

```
# Fitting a random model
data(tpod)
FIT = reml(y=y,Z=~as.factor(fam))

# Fitting GBLUP
G = tcrossprod(gen)
G = G/mean(diag(G))
GBLUP = reml(y=y,K=G)
```

`SNP H2` *SNP heritability*

Description

Calculates the ability of markers to carry a gene.

Usage

```
snpH2(gen)
```

Arguments

`gen`	Numeric matrix containing the genotypic data. A matrix with n rows of observations and (m) columns of molecular markers. Missing values not allowed.

Value

Numeric vector containing the heritability of each markers.

Author(s)

Alencar Xavier

References

Forneris, N. S., Legarra, A., Vitezica, Z. G., Tsuruta, S., Aguilar, I., Misztal, I., & Cantet, R. J. (2015). Quality Control of Genotypes Using Heritability Estimates of Gene Content at the Marker. Genetics, genetics-114.

Examples

```
data(tpod)
Heritability=snpH2(gen)
plot(Heritability,chr=chr)
```

`SNP QC` *SNP Quality Control*

Description

A function for quality control. It may be used to count/remove neighbor repeated SNPs and markers with MAF lower than a given threshold. This function is also used for imputations.

Usage

```
snpQC(gen,psy=1,MAF=0.05,remove=TRUE,impute=FALSE)
```

Arguments

`gen`	Numeric matrix containing the genotypic data. A matrix with n rows of observations and (m) columns of molecular markers. SNPs must be coded as `0, 1, 2`, for founder homozigous, heterozigous and reference homozigous. `NA` is allowed.
`psy`	Tolerance parameter for repeated markers. Default is 1, which removes only SNPs 100% equal to its following neighbor.
`MAF`	Minor Allele Frequency. Default is 0.05. Useful to inform or remove markers below the MAF threshold.
`remove`	Remove SNPs that are redundant or pursue low MAF: TRUE/FALSE.
`impute`	If TRUE, impute missing values using Random Forest adapted from the package missForest (Stekhoven and Buhlmann 2012) as suggested by Rutkoski et al (2013).

Value

Returns the genomic matrix without missing, redundant or low MAF markers.

Author(s)

Alencar Xavier, Katy Rainey and William Muir

References

Rutkoski, J. E., Poland, J., Jannink, J. L., & Sorrells, M. E. (2013). Imputation of unordered markers and the impact on genomic selection accuracy. G3: Genes| Genomes| Genetics, 3(3), 427-439.

Stekhoven, D. J. and Buhlmann, P. 2012. MissForest - nonparametric missing value imputation for mixed-type data. Bioinformatics, 28(1), 112-118.

Examples

```
data(tpod)
gen=snpQC(gen=gen,psy=1,MAF=0.05,remove=TRUE,impute=FALSE)
```

WGR *Whole-genome Regression*

Description

Univariate model to find breeding values through regression with optional resampling techniques.

Usage

```
wgr(y,gen,it=1500,bi=500,th=1,bag=1,rp=TRUE,iv=FALSE,
    pi=0,df=5,R2=0.5,eigK=NULL,rankK=0.25,verb=FALSE)
```

Arguments

y	Numeric vector of observations (n) describing the trait to be analyzed. NA is allowed.
gen	Numeric matrix containing the genotypic data. A matrix with n rows of observations and (m) columns of molecular markers.
it	Integer. Number of iterations or samples to be generated.
bi	Integer. Burn-in, the number of iterations or samples to be discarted.
th	Integer. Thinning parameter, used to save memory by storing only one every 'th' samples.
bag	Proportion of data used to bag MCMC.
rp	Logical. Use replacement (bootstrap) samples when bag is different than one.
iv	Logical. Assign markers independent variance. If true, turns the default model BLUP into BayesA. For this model, the shape parameter is conjugated by a gamma with hyperpriors calculated based on the R2 rule.
pi	Value between 0 and 1. If greater than zero it activates variable selection, where markers have expected probability pi of having null effect. The model conjugates variable selection from a Beta-Binomial distribution.
df	Hyperprior degrees of freedom of variance components.
R2	Expected R2, used to calculate the prior shape as proposed by de los Campos et al. (2013).

eigK	Output of function 'eigen'. Spectral decomposition of the kernel used to compute the polygenic term.
rankK	Numeric between 0 and 1. Indicates the proportion of Eigenpairs used to fit the polygenic term.
verb	Logical. If verbose is TRUE, function displays MCMC progress bar.

Details

The model for the whole-genome regression is as follows:

$y = mu + Xg + u + e$,

where y is the response variable, mu is the intercept, X is the genotypic matrix, g is the product of two terms ($g = bg$), b is the effect of an allele substitution, d is an indicator variable that define whether or not the marker should be included into the model, u is the polygenic term and e is the residual term.

Users can obtain four WGR methods out of this function: BRR (pi=0,iv=F), BayesA (pi=0,iv=T), BayesB (pi=0.8,iv=T) and BayesC (pi=0.8,iv=F). The full theoretical basis of each model is described by de los Campos et al. (2013).

Gibbs sampler that updates regression coefficients is adapted from GSRU algorithm (Legarra and Misztal 2008). The variable selection works through the unconditional prior algorithm proposed by Kuo and Mallick (1998). The polygenic term is solved by Bayesian algorithm of reproducing kernel Hilbert Spaces proposed by de los Campos et al. (2010).

Value

The function wgr returns a list with expected value from the marker effect (b), probability of marker being in the model (d), regression coefficient (g), variance of each marker (Vb), the intercept (mu), the polygene (u) and polygenic variance (Vk), residual variance (Ve) and the fitted value (hat).

variance components distribution a posteriori (Posterior.VC) and mode estimated (VC.estimate), a list with the posterior distribution of regression coefficients (Posterior.Coef) and the posterior mean (Coef.estimate), and the fitted values using the mean (Fit.mean) of posterior coefficients.

Author(s)

Alencar Xavier

References

de los Campos, G., Hickey, J. M., Pong-Wong, R., Daetwyler, H. D., and Calus, M. P. (2013). Whole-genome regression and prediction methods applied to plant and animal breeding. Genetics, 193(2), 327-345.

de los Campos, G., Gianola, D., Rosa, G. J., Weigel, K. A., & Crossa, J. (2010). Semi-parametric genomic-enabled prediction of genetic values using reproducing kernel Hilbert spaces methods. Genetics Research, 92(04), 295-308.

Kuo, L., & Mallick, B. (1998). Variable selection for regression models. Sankhya: The Indian Journal of Statistics, Series B, 65-81.

Legarra, A., & Misztal, I. (2008). Technical note: Computing strategies in genome-wide selection. Journal of dairy science, 91(1), 360-366.

Examples

```
data(tpod)
gen = gen[,seq(1,376,5)]

# BLUP
BRR = wgr(y,gen,iv=FALSE,pi=0,bag=0.5,rp=TRUE,it=400,bi=50)
cor(y,BRR$hat)

# BayesA
BA = wgr(y,gen,iv=TRUE,pi=0,bag=0.5,rp=TRUE,it=400,bi=50)
cor(y,BA$hat)

# BayesB
BB = wgr(y,gen,iv=TRUE,pi=.5,bag=0.5,rp=TRUE,it=400,bi=50)
cor(y,BB$hat)

# BayesC
BC = wgr(y,gen,iv=FALSE,pi=.5,bag=0.5,rp=TRUE,it=400,bi=50)
cor(y,BC$hat)
```

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