iMKT Pipeline

Brief intro about why we developed the package. The importance of detecting adaptation, and the many MKT derived methodologies. Take from paper.

Loading the package and checking test data

First of all, install (if this is not done yet) and load the package. Notice that iMKT package includes two sample dataframes named mydafdata and mydivergencedata which are the ones used in this tutorial. This makes easier to replicate the vignettes to understand better all the package functionalities.

```
## Load package
#devtools::install_github("sergihervas/iMKT")
library(iMKT)

## Sample daf data
head(mydafdata)
#>     daf    Pi    P0
#> 1 0.025 22490 17189
#> 2 0.075  3217  4780
#> 3 0.125  1616  2874
#> 4 0.175   999  2088
#> 5 0.225   754  1685
#> 6 0.275   679  1443

## Sample divergence data
mydivergencedata
#>        mi    Di     m0    D0
#> 1 2598805 54641 620019 52537
```

The package includes several funcions, classified as follows:

- MKT derived methodologies calculation
  - standard(): Standard MKT
  - FWW(): FWW correction
  - DGRP(): DGRP correction
  - asymptoticMK(): Asymptotic MKT
  - iMK(): integrative MKT
  - completeMKT(): perform all previous tests
- iMK using PopFly and PopHuman data
  - loadPopFly(): load PopFlyData
  - loadPopHuman(): load PopHumanData
  - PopFlyAnalisys(): perform any test using PopFlyData
  - PopHumanAnalysis(): perform any test using PopHumanData

- Miscelanious
  - multipleDatasets(): analyze all datasets in your working directory
  - checkInput(): check data before performing analyses
  - themePublication(): output plots and tables styling

Rembember you always can access to the help, to check more examples or the passing arguments writting `{r}??` and the function in your console.

This vignette focuses on the first category of functions: MKT derived methodologies calculation. For details about how to use the functions from iMKT using PopFly and PopHuman data category, check the corresponding vignette.

Performing MKT derived methodologies calculation

Standard MKT

```
standard(daf=mydafdata, divergence=mydivergencedata)
#> $alpha.symbol
#> [1] 0.2364499
#>
#> $`Fishers exact test P-value`
#> [1] 1.480943e-183
#>
#> $`MKT table`
#>
#>
#>                   Polymorphism   Divergence
#> ---------------  -------------  -----------
#> Neutral class            45101        52537
#> Selected class           35816        54641
#>
#> $`Divergence metrics`
#>
#>
#>          Ka           Ks       omega      omegaA      omegaD
#> ----------  -----------  ----------  ---------  ---------
#>  0.0210254   0.0847345   0.2481331   0.058671   0.189462
```

FWW correction

Alpha estimates can be biased by the segregation of slighlty deleterious substitutions. One method to partially controlled its effects is to remove low frequency polymorphisms from the analysis, as proposed by Fay et al. (2001).

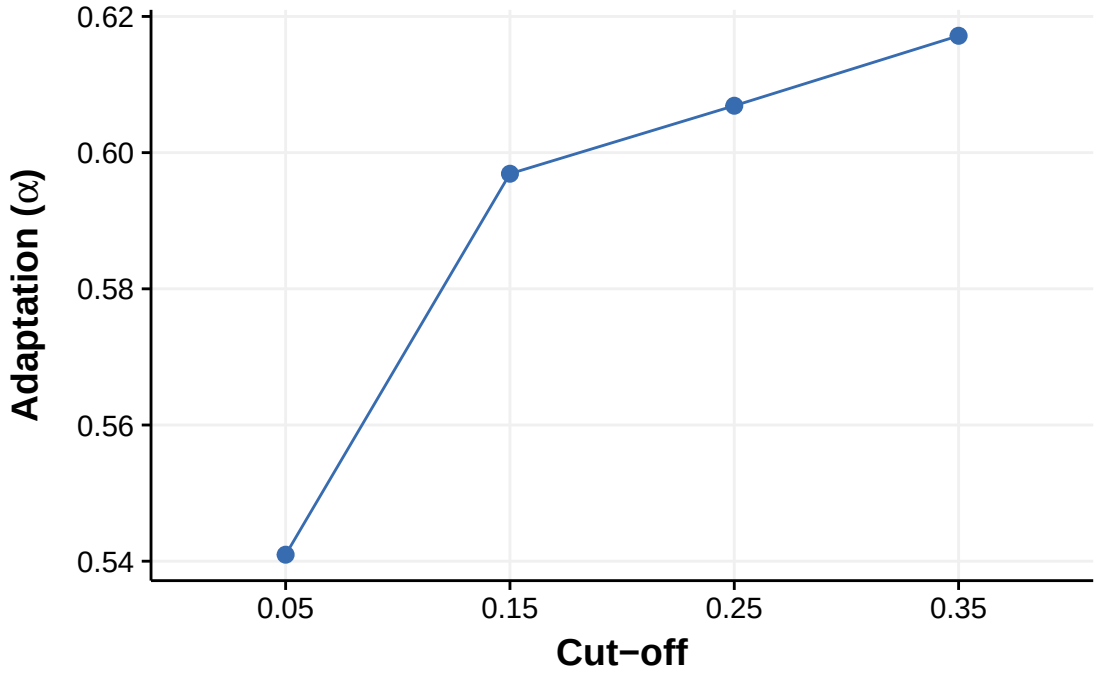
```
FWW(daf=mydafdata, divergence=mydivergencedata)
#> $Results
```

```
#>                alpha.symbol Fishers exact test P-value
#> Cutoff =  0       0.2364499              1.480943e-183
#> Cutoff =  0.05    0.5409548               0.000000e+00
#> Cutoff =  0.1     0.5798139               0.000000e+00
#>
#> $`Divergence metrics`
#> $`Divergence metrics`$`Global metrics`
#>           Ka        Ks     omega
#> 1 0.02102543 0.0847345 0.2481331
#>
#> $`Divergence metrics`$`Estimates by cutoff`
#>                omegaA.symbol omegaD.symbol
#> Cutoff =  0       0.05867104     0.1894620
#> Cutoff =  0.05    0.13422877     0.1139043
#> Cutoff =  0.1     0.14387102     0.1042621
#>
#>
#> $`MKT tables`
#> $`MKT tables`$`Cutoff =  0`
#>
#>
#> Table: cutoff
#>
#>                   Polymorphism   Divergence
#> ----------------  -------------  -----------
#> Neutral class            45101        52537
#> Selected class           35816        54641
#>
#> $`MKT tables`$`Cutoff =  0.05`
#>
#>
#> Table: cutoff
#>
#>                   Polymorphism   Divergence
#> ----------------  -------------  -----------
#> Neutral class            27912        52537
#> Selected class           13326        54641
#>
#> $`MKT tables`$`Cutoff =  0.1`
#>
#>
#> Table: cutoff
#>
#>                   Polymorphism   Divergence
#> ----------------  -------------  -----------
#> Neutral class            23132        52537
#> Selected class           10109        54641
```

By default the argument **list_cutoff** uses a list of cutoffs with the following values: 0, 0.05, 0.1, and the argument **plot** is set to **FALSE**. This parameters can be customized, like in the following example:

```
FWW(daf=mydafdata, divergence=mydivergencedata, list_cutoff=c(0.05, 0.15,0.25,0.35), plot=TRUE)
```

```
#> $Results
#>                 alpha.symbol Fishers exact test P-value
#> Cutoff =  0.05     0.5409548                           0
#> Cutoff =  0.15     0.5969015                           0
#> Cutoff =  0.25     0.6068868                           0
#> Cutoff =  0.35     0.6171609                           0
#>
#> $Graph
```




```
#>
#> $`Divergence metrics`
#> $`Divergence metrics`$`Global metrics`
#>           Ka        Ks     omega
#> 1 0.02102543 0.0847345 0.2481331
#>
#> $`Divergence metrics`$`Estimates by cutoff`
#>                 omegaA.symbol omegaD.symbol
#> Cutoff =  0.05     0.1342288    0.11390431
#> Cutoff =  0.15     0.1481110    0.10002208
#> Cutoff =  0.25     0.1505887    0.09754438
#> Cutoff =  0.35     0.1531380    0.09499504
#>
#>
#> $`MKT tables`
#> $`MKT tables`$`Cutoff =  0.05`
#>
#>
#> Table: cutoff
#>
#>                   Polymorphism   Divergence
#> ---------------  -------------  -----------
#> Neutral class            27912        52537
#> Selected class           13326        54641
```

```
#>
#> $`MKT tables`$`Cutoff =  0.15`
#>
#>
#> Table: cutoff
#>
#>                   Polymorphism   Divergence
#> ---------------  -------------  -----------
#> Neutral class            20258        52537
#> Selected class            8493        54641
#>
#> $`MKT tables`$`Cutoff =  0.25`
#>
#>
#> Table: cutoff
#>
#>                   Polymorphism   Divergence
#> ---------------  -------------  -----------
#> Neutral class            16485        52537
#> Selected class            6740        54641
#>
#> $`MKT tables`$`Cutoff =  0.35`
#>
#>
#> Table: cutoff
#>
#>                   Polymorphism   Divergence
#> ---------------  -------------  -----------
#> Neutral class            13778        52537
#> Selected class            5486        54641
```

DGRP correction

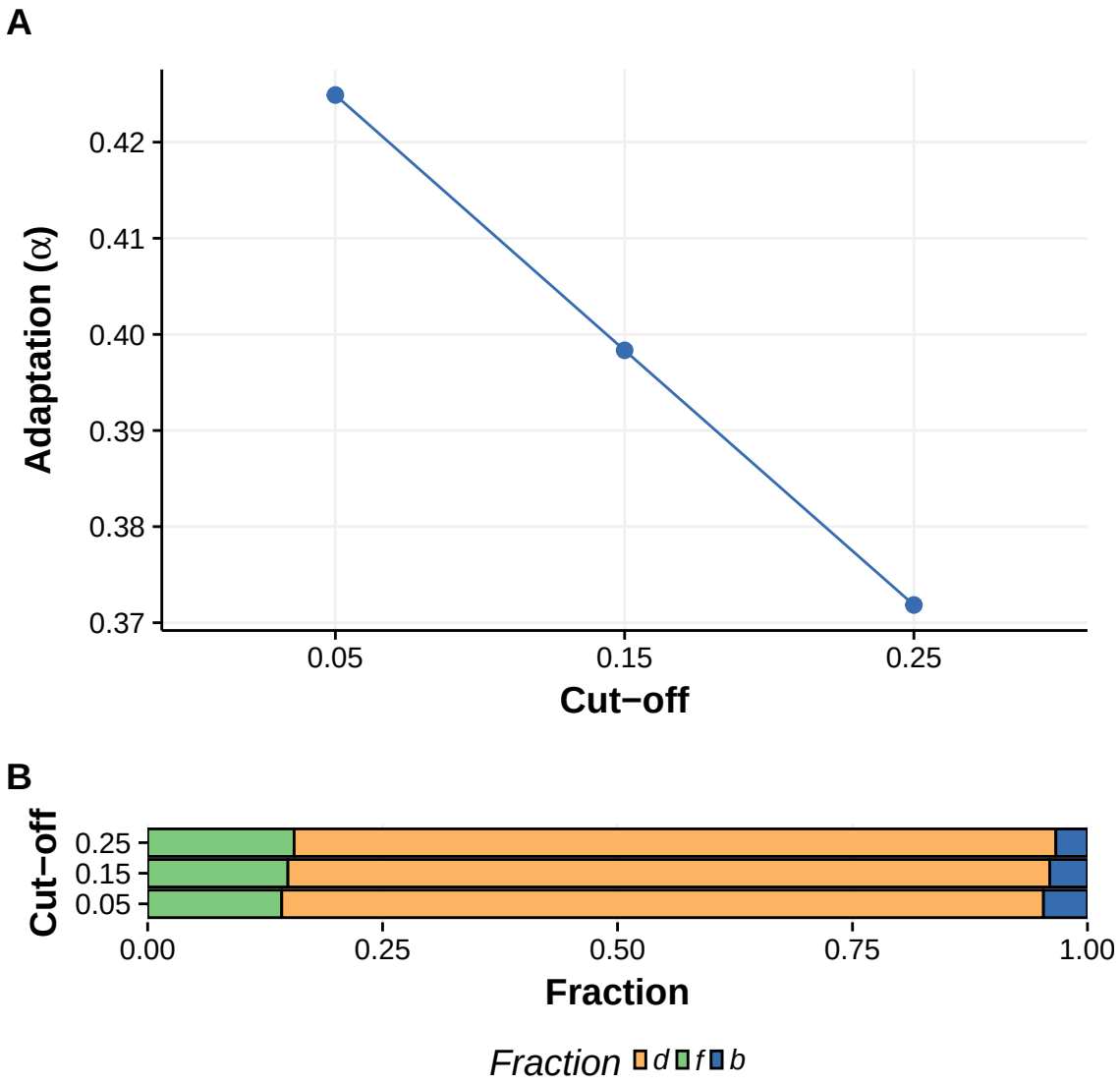
```
DGRP(daf=mydafdata, divergence=mydivergencedata)
#> $Results
#>                alpha.symbol Fishers exact test P-value
#> Cutoff =  0       0.2364499              1.480943e-183
#> Cutoff =  0.05    0.4249071               0.000000e+00
#> Cutoff =  0.2     0.3842950               0.000000e+00
#>
#> $`Divergence metrics`
#> $`Divergence metrics`$`Global metrics`
#>           Ka        Ks     omega
#> 1 0.02102543 0.0847345 0.2481331
#>
#> $`Divergence metrics`$`Estimates by cutoff`
#>                omegaA.symbol omegaD.symbol
#> Cutoff =  0       0.05867104     0.1894620
#> Cutoff =  0.05    0.10543351     0.1426996
#> Cutoff =  0.2     0.09535630     0.1527768
#>
```

```
#>
#> $`MKT tables`
#> $`MKT tables`$`Number of segregating sites by DAF category - Cutoff =  0`
#>
#>
#> Table: cutoff
#>
#>                  DAF.below.cutoff   DAF.above.cutoff
#> ---------------  -----------------  -----------------
#> Neutral class                    0              45101
#> Selected class                   0              35816
#>
#> $`MKT tables`$`Number of segregating sites by DAF category - Cutoff =  0.05`
#>
#>
#> Table: cutoff
#>
#>                  DAF.below.cutoff   DAF.above.cutoff
#> ---------------  -----------------  -----------------
#> Neutral class                17189              27912
#> Selected class               22490              13326
#>
#> $`MKT tables`$`Number of segregating sites by DAF category - Cutoff =  0.2`
#>
#>
#> Table: cutoff
#>
#>                  DAF.below.cutoff   DAF.above.cutoff
#> ---------------  -----------------  -----------------
#> Neutral class                26931              18170
#> Selected class               28322               7494
#>
#> $`MKT tables`$`MKT standard table`
#>
#>
#>                   Polymorphism   Divergence
#> ---------------  -------------  -----------
#> Neutral class            45101        52537
#> Selected class           35816        54641
#>
#>
#> $Fractions
#>          0       0.05        0.2
#> d 0.810538 0.81053943 0.81053627
#> f 0.189462 0.14269958 0.15277678
#> b 0.000000 0.04676099 0.03668695
```

Again, by default the argument **list_cutoff** uses a list of cutoffs with the following values: 0, 0.05, 0.1, and the argument **plot** is set to **FALSE**. This parameters can be customized, like in the following example:

```
DGRP(daf=mydafdata, divergence=mydivergencedata, list_cutoff=c(0.05,0.15,0.25), plot=TRUE)
#> $Results
```

```
#>                 alpha.symbol Fishers exact test P-value
#> Cutoff =  0.05    0.4249071                           0
#> Cutoff =  0.15    0.3983440                           0
#> Cutoff =  0.25    0.3718449                           0
#>
#> $Graph
```




```
#>
#> $`Divergence metrics`
#> $`Divergence metrics`$`Global metrics`
#>           Ka        Ks     omega
#> 1 0.02102543 0.0847345 0.2481331
#>
#> $`Divergence metrics`$`Estimates by cutoff`
#>                 omegaA.symbol omegaD.symbol
#> Cutoff =  0.05    0.10543351     0.1426996
#> Cutoff =  0.15    0.09884233     0.1492908
#> Cutoff =  0.25    0.09226702     0.1558661
#>
#>
```

```
#> $`MKT tables`
#> $`MKT tables`$`Number of segregating sites by DAF category - Cutoff =  0.05`
#>
#>
#> Table: cutoff
#>
#>                   DAF.below.cutoff   DAF.above.cutoff
#> ---------------  -----------------  -----------------
#> Neutral class                17189              27912
#> Selected class               22490              13326
#>
#> $`MKT tables`$`Number of segregating sites by DAF category - Cutoff =  0.15`
#>
#>
#> Table: cutoff
#>
#>                   DAF.below.cutoff   DAF.above.cutoff
#> ---------------  -----------------  -----------------
#> Neutral class                24843              20258
#> Selected class               27323               8493
#>
#> $`MKT tables`$`Number of segregating sites by DAF category - Cutoff =  0.25`
#>
#>
#> Table: cutoff
#>
#>                   DAF.below.cutoff   DAF.above.cutoff
#> ---------------  -----------------  -----------------
#> Neutral class                28616              16485
#> Selected class               29076               6740
#>
#> $`MKT tables`$`MKT standard table`
#>
#>
#>                   Polymorphism   Divergence
#> ---------------  -------------  -----------
#> Neutral class            45101        52537
#> Selected class           35816        54641
#>
#>
#> $Fractions
#>         0.05       0.15      0.25
#> d 0.81053943 0.81053552 0.8105368
#> f 0.14269958 0.14929076 0.1558661
#> b 0.04676099 0.04017372 0.0335971
```

Asymptotic MKT

Petrov reference + explanation

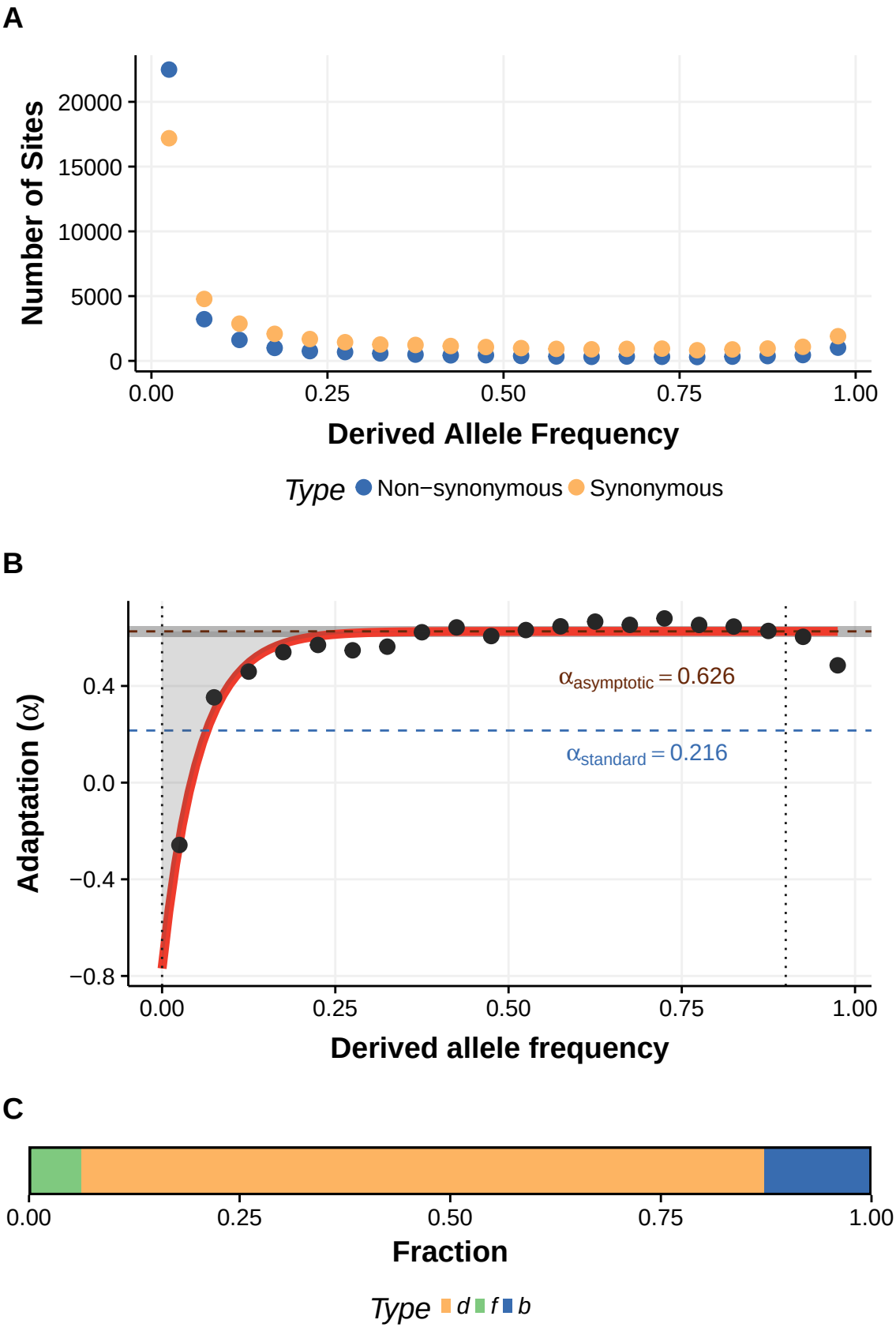
```
asymptoticMK(daf=mydafdata, divergence=mydivergencedata, xlow=0, xhigh=0.9)
```

```
#>         model         a         b        c alpha_asymptotic    CI_low
#> 1 exponential 0.6258904 -1.395108 18.96187        0.6258904 0.6043099
#>     CI_high alpha_original
#> 1 0.6474287      0.2157308
```

iMK

Asymptotic explanation + Sergi slightly deleterious approach

```
iMK(daf=mydafdata, divergence=mydivergencedata, xlow=0, xhigh=0.9, plot=TRUE)
#> $`Asymptotic MK table`
#>         model      a       b       c alpha_asymptotic CI_low CI_high
#> 1 exponential 0.6259 -1.3951 18.9619           0.6259 0.6045  0.6474
#>   alpha_original
#> 1         0.2157
#>
#> $`Fractions of sites`
#>   Type   Fraction
#> 1    d 0.81053796
#> 2    f 0.06232362
#> 3    b 0.12713842
#>
#> $Graphs
```


A
Number of Sites
20000
15000
10000
5000
0
0.00
0.25
0.50
0.75
1.00
Derived Allele Frequency
Type
Non−synonymous
Synonymous
B
Adaptation (α)
0.4
0.0
−0.4
−0.8
αasymptotic = 0.626
αstandard = 0.216
0.00
0.25
0.50
0.75
1.00
Derived allele frequency
C
0.00
0.25
0.50
0.75
1.00
Fraction
Type
d
f
b

Miscelanious

If you have a bunch of data like the following, or simply have several genes datasets: Maybe you want to perform some test or compare the test results between your datasets. You could execute the funtion multipleDatasets, putting your datasets in a directory a name them with the extensions **ID.daf.txt/ID.divergence.txt**. Then execute the following commands to perform the tests:

The idList argument allow to the user pass a plain text file with the IDs, in the case you want to subset the analysis to just a few datasets. It is used when fullAnalysis = FALSE, list of IDs to analyze