PAGnet: an integrated genomic regulatory network in Pseudomonas aeruginosa.

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Abstract

PAGnet is an R package to analyze genomic regulatory network in Pseudomonas aerugionsa. This package provides Master Regulator Analysis (MRA) for identification of key transcription factors mediating a biological process or pathway in Pseudomonas aeruginosa Genomic netork (PAGnet).

Package

PAGnet 0.1.0

Contents

1 Overview

Regulatory networks including virulence-related transcriptional factors (TFs) determine bacterial pathogenicity in response to different environmental cues. Pseudomonas aeruginosa, a Gram-negative opportunistic pathogen of humans, recruits numerous TFs in quorum sensing (QS) system, type III secretion system (T3SS) and Type VI secretion system (T6SS) to mediate the pathogenicity. Although many virulence-related TFs have been illustrated individually, very little is known about their crosstalks and regulatory network. Here, based on chromatin immunoprecipitation coupled with high-throughput sequencing (ChIP-seq) and transcriptome profiling (RNA-seq), we primarily focused on understanding the crosstalks of 19 virulence-related TFs, which led to construction of a virulence regulatory network named PAGnet (Pseudomonas aeruginosa Genomic network) including 48 crosstalk targets.

The PAGnet uncovered the intricate mechanism of virulence regulation and revealed master regulators in QS, T3SS and T6SS pathways. The package **PAGnet** is designed for Master Regulator Analysis (MRA) over a list of regulons from PAGnet. We also provide an online PAGnet platform was established to provide the analysis for these TFs and more virulence factors.

```
## loading packages
library(PAGnet)
```

2 Quick Start

2.1 Master Regulator Analysis

The user can choose to use the default PAGnet or to upload their own regulatory network in a predefined format.

```
  library(PAGnet)
  data(PAGnet)

  #Use PAGnet as regulatory network
  #Select T3SS related genes as signatures
  #run MRA
  head(PAGnet)
##   TranscriptionFactor Target
## 1              PA5261 PA2523
## 2              PA5261 PA1727
## 3              PA5261 PA4396
## 4              PA5261 PA2189
## 5              PA5261 PA1736
## 6              PA5261 PA3763
  head(tf)
## [1] "PA5261" "PA3385" "PA4101" "PA2588" "PA1713" "PA2586"
  head(qs)
## [1] "PA1130" "PA3387" "PA3476" "PA3477" "PA3479" "PA3478"
```

The function `pagnet.mra` is used to perform MRA in PAGnet or user uploaded network. The MRA computes the overlap between the transcriptional regulatory unities (regulons) and the input signature genes using the hypergeometric distribution.Having completed master regulator analysis, a table will be returned.

```
  mra_results <- pagnet.mra(rnet=PAGnet,tflist=tf,signature = qs,
                            pValueCutoff = 0.05,pAdjustMethod="BH")
  mra_results
##        TF network.size regulon.size signature.size observed.signature.size
## 7  PA1430          422           45             20                      10
## 9  PA1003          422           21             20                       7
## 12 PA3477          422            7             20                       5
## 14 PA1431          422            9             20                       5
## 10 PA1898          422            6             20                       3
## 11 PA2593          422            2             20                       2
## 18 PA2227          422            7             20                       3
##    Pvalue adjust.Pvalue
## 7  < 1e-4        < 1e-4
## 9  < 1e-4        < 1e-4
## 12 < 1e-4        < 1e-4
## 14 < 1e-4        < 1e-4
## 10 0.0017        0.0065
## 11 0.0021        0.0066
## 18 0.0028        0.0076
```

To output the MRA results, the user can use `write.csv` to output csv file.

```
  ## write.csv(mra_results,"MRA_results.csv")
```

2.2 Local shiny interface

The function `pagnet.mra.interface` is used to call a local interface of shiny to perform MRA like onlie platform.First, the user can choose to use the default PAVIRnet or to upload their own regulatory network in a predefined format. Second, the user needs to specify a gene signature associated with a biological function or pathway of interest, either by selecting a gene set from public databases or uploading a user-customized gene list. In the current version, the platform provides gene sets in Gene Ontology (GO) and KEGG databases obtained from Pseudomonas Genome DB. Having completed master regulator analysis, a table will be returned with information about each transcription factor's corresponding gene ID, gene name, number of target genes, total number of hits (all signature genes in the network), observed hits (signature genes in the TF's regulon), and a p-value calculated based on a hypergeometric test. The table is sorted according to the statistical significance indicated by the p-values, and the top significant TFs can be prioritized as master regulators.

```
  # pagnet.mra.interface()
```

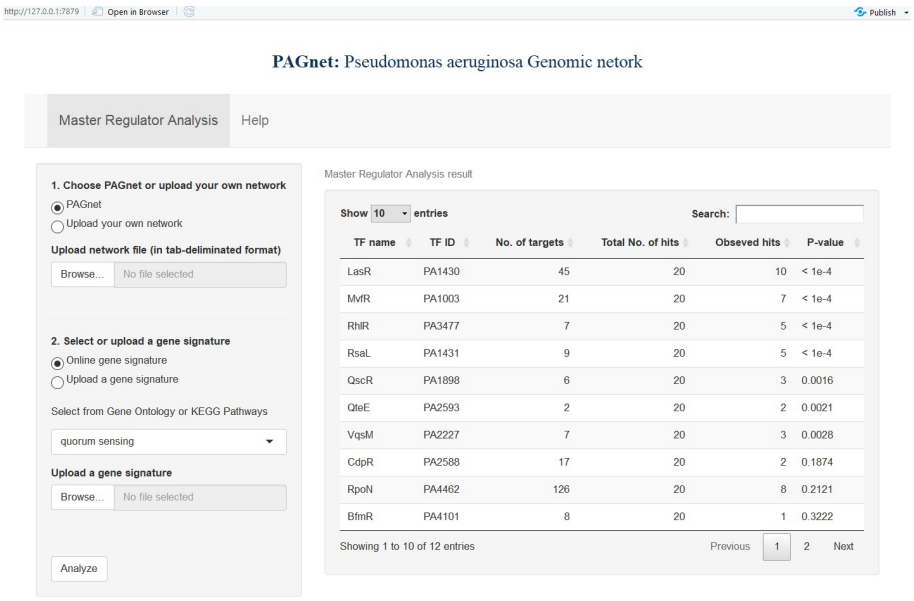

Figure 1: **"A local shiny interface of PAGnet"**

3 Need helps?

If you have any question/issue, please feel free to contact us.

4 Session Information

```
## R version 3.5.2 (2018-12-20)
## Platform: x86_64-w64-mingw32/x64 (64-bit)
## Running under: Windows 10 x64 (build 17763)
##
## Matrix products: default
##
## locale:
## [1] LC_COLLATE=Chinese (Simplified)_China.936
## [2] LC_CTYPE=Chinese (Simplified)_China.936
## [3] LC_MONETARY=Chinese (Simplified)_China.936
## [4] LC_NUMERIC=C
## [5] LC_TIME=Chinese (Simplified)_China.936
##
## attached base packages:
## [1] stats     graphics  grDevices utils     datasets  methods   base
##
## other attached packages:
## [1] PAGnet_0.1.0    BiocStyle_2.8.2
##
## loaded via a namespace (and not attached):
##  [1] Rcpp_0.12.18    bookdown_0.7    digest_0.6.17   rprojroot_1.3-2
##  [5] backports_1.1.2 magrittr_1.5    evaluate_0.11   stringi_1.2.4
```

```
##  [9] rmarkdown_1.10  tools_3.5.2     stringr_1.3.1   xfun_0.3
## [13] yaml_2.2.0      compiler_3.5.2  htmltools_0.3.6 knitr_1.20
```

5 References