GraphSNP

GraphSNP USER MANUAL

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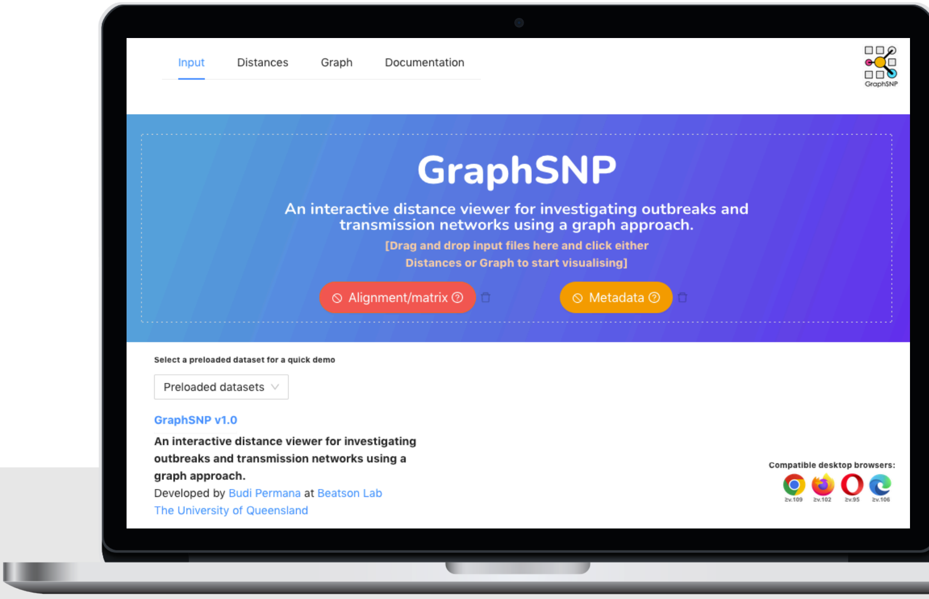
v.2023.02.07

Using GraphSNP

GraphSNP is a single page application (SPA) visualisation tool that runs on the browser. Users can visualise and explore data by loading their input files or setting up multiple projects (available on offline use only) for multiple input datasets.



GraphSNP

An interactive distance viewer for investigating outbreaks and transmission networks using a graph approach.

[Drag and drop input files here and click either Distances or Graph to start visualising]

Select a preloaded dataset for a quick demo

GraphSNP v1.0

An interactive distance viewer for investigating outbreaks and transmission networks using a graph approach.

Developed by Budi Permana at Beatson Lab

The University of Queensland

Compatible desktop browsers:

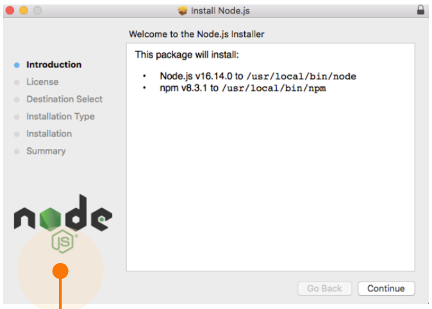
Use it online

GraphSNP is deployed in https://graphsnp.beatsonlab.com for online use. Users can visit the web page using modern browsers (e.g., Google Chrome, Firefox, Microsoft Edge, and Safari), drag and drop the input files, and instantly perform interactive data visualization and analysis.

Use it offline

Users also can use GrapSNP offline by serving it through a local HTTP server. GraphSNP SPA can be downloaded from https://github.com/nalarbp/graphsnp/build/.

Example of serving GraphSNP using HTTP-server "*serve*" tool

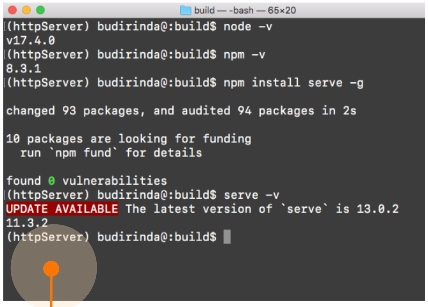


Welcome to the Node.js Installer

This package will install:

- Node.js v16.14.0 to /usr/local/bin/node
- npm v8.3.1 to /usr/local/bin/npm

Install Node.js
(available at https://nodejs.org/en/)



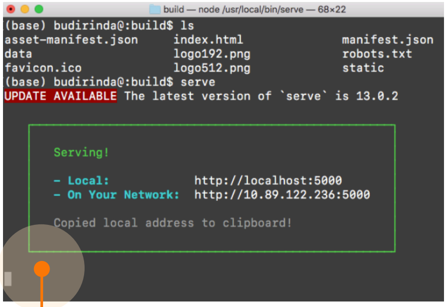
```
(httpServer) budirinda@:build$ node -v
v17.4.0
(httpServer) budirinda@:build$ npm -v
8.3.1
(httpServer) budirinda@:build$ npm install serve -g

changed 93 packages, and audited 94 packages in 2s

10 packages are looking for funding
  run `npm fund` for details

found 0 vulnerabilities
(httpServer) budirinda@:build$ serve -v
UPDATE AVAILABLE The latest version of `serve` is 13.0.2
11.3.2
(httpServer) budirinda@:build$
```

Install serve via npm
(npm install serve -g)



```
(base) budirinda@:build$ ls
asset-manifest.json     index.html              manifest.json
data                    logo192.png             robots.txt
favicon.ico             logo512.png             static
(base) budirinda@:build$ serve
UPDATE AVAILABLE The latest version of `serve` is 13.0.2


   Serving!

   - Local:            http://localhost:5000
   - On Your Network:  http://10.89.122.236:5000

   Copied local address to clipboard!
```

Run the HTTP server
(serve .)

Input Files

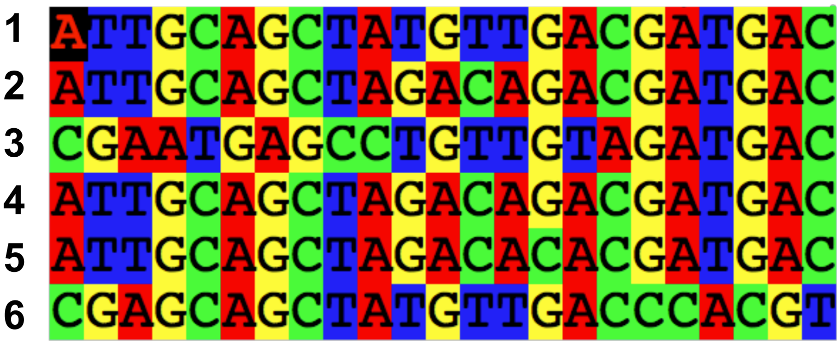
SNPs alignment

A text file containing a minimum of two equal lengths of fasta-formatted non-gap ATGC-exclusive nucleotide sequences (when other caharacters (e.g., N, '-') and or specific models need to be taken into account, users can use distance matrix generated by other tools, instead of alignment).

Example SNPs alignment input *(sample.fasta)*

```
>1
ATTGCAGCTATGTTGACGATGAC
>2
ATTGCAGCTAGACAGACGATGAC
>3
CGAATGAGCCTGTTGTAGATGAC
>4
ATTGCAGCTAGACAGACGATGAC
>5
ATTGCAGCTAGACACACGATGAC
>6
CGAGCAGCTATGTTGACCCACGT
```

Sample ID in fasta header


1 ATTGCAGCTATGTTGACGATGAC
2 ATTGCAGCTAGACAGACGATGAC
3 CGAATGAGCCTGTTGTAGATGAC
4 ATTGCAGCTAGACAGACGATGAC
5 ATTGCAGCTAGACACACGATGAC
6 CGAGCAGCTATGTTGACCCACGT

Example of pairwise SNP distances matrix *(sample_matrix.csv)*

dist	1	2	3	4	5	6
1	0	4	12	4	5	9
2	4	0	16	0	1	13
3	12	16	0	16	17	15
4	4	0	16	0	1	13
5	5	1	17	1	0	14
6	9	13	15	13	14	0

Pairwise distances matrix

Matrix in CSV format

```
dist,1,2,3,4,5,6
1,0,4,12,4,5,9
2,4,0,16,0,1,13
3,12,16,0,16,17,15
4,4,0,16,0,1,13
5,5,1,17,1,0,14
6,9,13,15,13,14,0
```

User can also input the pairwise distances matrix instead of SNP alignment. The symmetric matrix should be written in comma-separated value (CSV) format.

Metadata

A table contains information about the isolates or sample, written in CSV format. Critical requirements including: mandatory headers, no duplicated records in column **sample_id**. Column **collection_day** is required for transmission analysis.

Mandatory column

Mandatory column for transmission analysis

Any additional column

Columns to set the color

sample_id	collection_day	Location	Source	Clade	Gene-A	Source:color	Gene-A:color
1	1	room A	clinical	A	present	#FF8076	Black
2	2	room B	clinical	A	present	#FF8076	Black
3	3	room C	clinical	A	present	#FF8076	Black
4	3	room A	environmental	A	absent	#53DE22	White
5	4	room B	environmental	A	absent	#53DE22	White
6	5	room C	environmental	A	absent	#53DE22	White

Main Interface

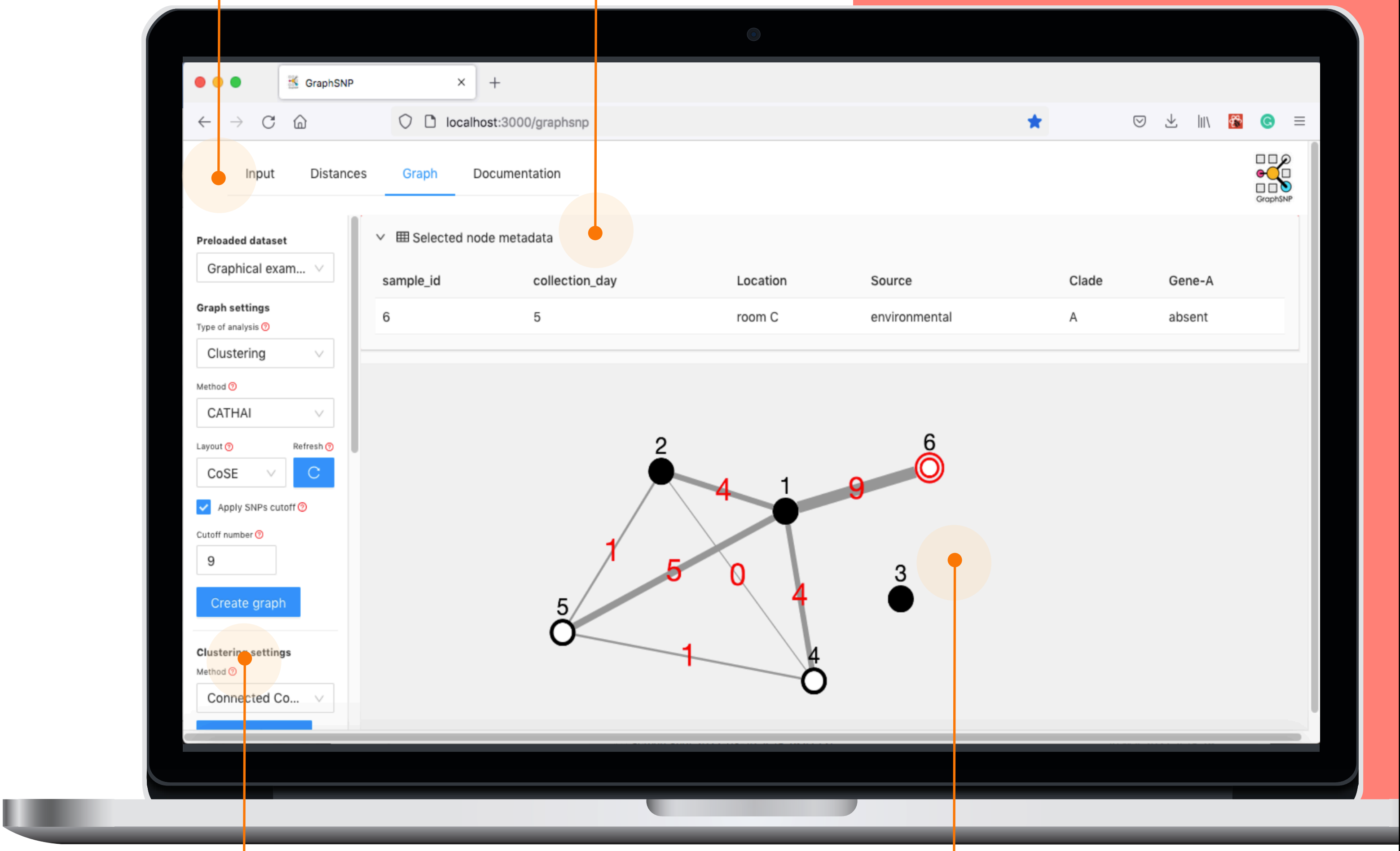
Page navigation

Navigation menu to let you jump between pages: *Input, Distances, Graph, and Documentation.*

Metadata table

Let you display metadata associated with selected node(s).

page Graph



Preloaded dataset: Graphical exam...

Graph settings

Type of analysis: Clustering

Method: CATHAI

Layout: CoSE

Refresh

☑ Apply SNPs cutoff

Cutoff number: 9

Create graph

Clustering settings

Method: Connected Co...

sample_id	collection_day	Location	Source	Clade	Gene-A
6	5	room C	environmental	A	absent

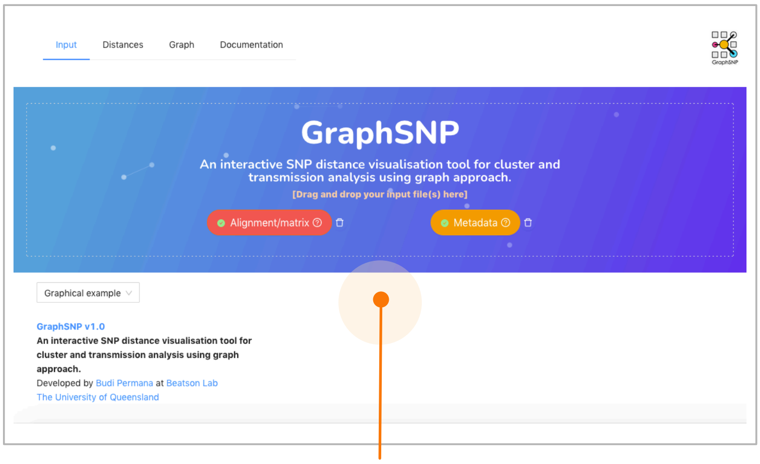
Sidebar settings

A sidebar menu provides you a control to adjust the visualisation.

Graph visualisation window

A window container where the interactive graph is being rendered.

page Input



GraphSNP

An interactive SNP distance visualisation tool for cluster and transmission analysis using graph approach.

[Drag and drop your input file(s) here]

Alignment/matrix

Metadata

Graphical example

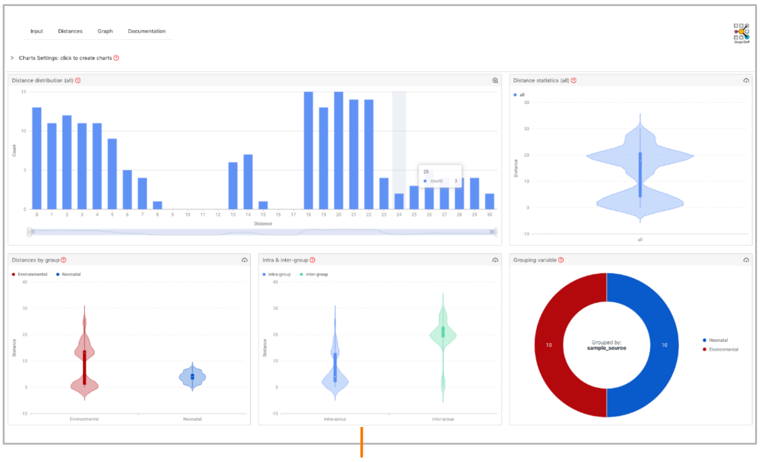
GraphSNP v1.0

An interactive SNP distance visualisation tool for cluster and transmission analysis using graph approach.

Developed by Budi Permana at Beatson Lab

The University of Queensland

page Distances



Input placeholder

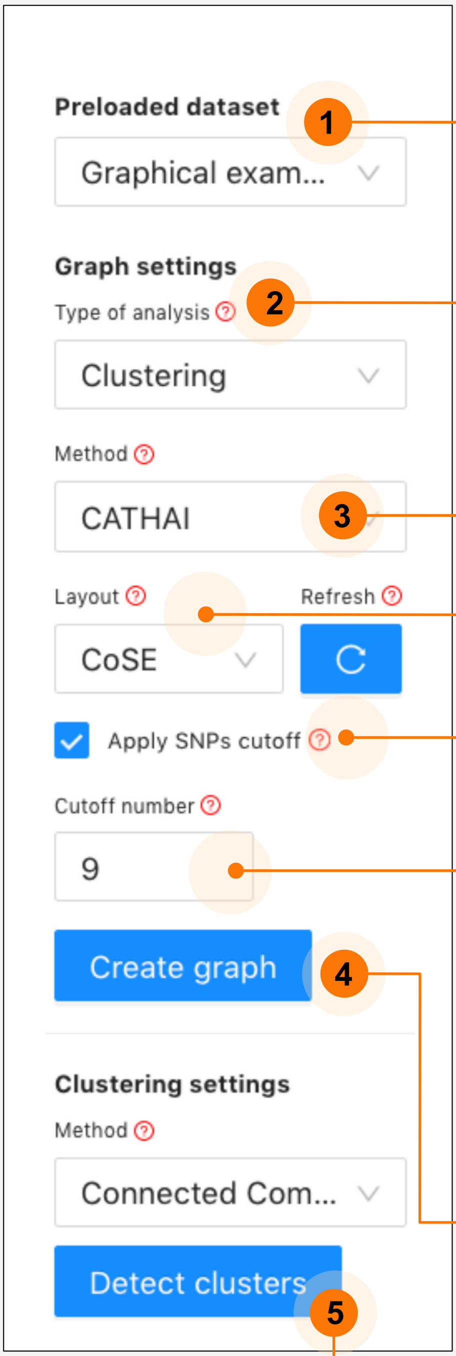
Drag and drop your input files here.

Chart visualisation window

A container where charts showing pairwise distances count and statistics is being rendered.

Cluster analysis

Cluster analysis and visualization can be performed in five simple steps: loading input files, select clustering as the type of analysis, select the clustering method, construct the graph, and detect/report clusters from the graph.



Preloaded dataset 1

Graphical exam...

Graph settings

Type of analysis 2

Clustering

Method

CATHAI 3

Layout — Refresh

CoSE

☑ Apply SNPs cutoff

Cutoff number

9

Create graph 4

Clustering settings

Method

Connected Com...

Detect clusters 5

Load input files

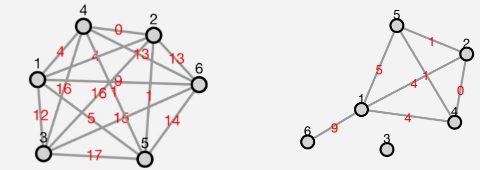
Select Clustering

Select reconstruction method

Select which layout to display the Graph.

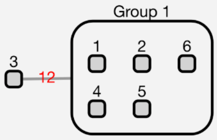
Layout options are based on Cytoscape.JS layouts (https://js.cytoscape.org/). *These algorithms (CoSE, fCoSE, CoSE Bilkent, and spread) determine the position of nodes and edges in a graph to provide a better graph visualisation, such as avoiding overlap and maintaining a clear structure of the graph.*

Network: *an undirected Graph based of pairwise distances. Node represents individual isolate. Edge represents distance.*



No cut-off

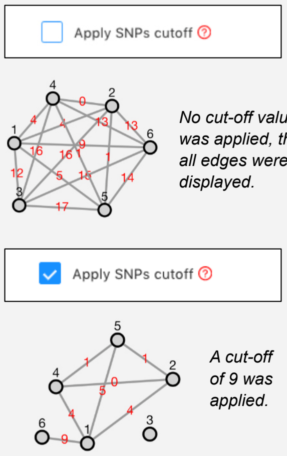
Cut-off = 9



Cut-off = 9

Threshold-based minimum spanning tree (MST): *Produce an MST of the identified cluster/s. Individual node represents isolate, while group node represents cluster given the threshold. Edges represents a minimum distance between cluster/s or between or to singleton/s.*

Apply a cutoff number to limit the maximum pairwise distance value to be displayed



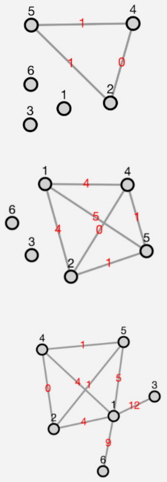
□ Apply SNPs cutoff

No cut-off value was applied, thus all edges were displayed.

☑ Apply SNPs cutoff

A cut-off of 9 was applied.

Set cut-off value



Cut-off = 1

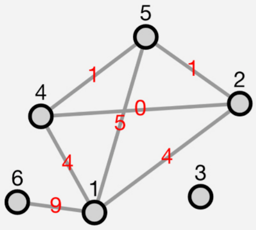
Cut-off = 5

Cut-off = 12

Create Graph

Identify cluster

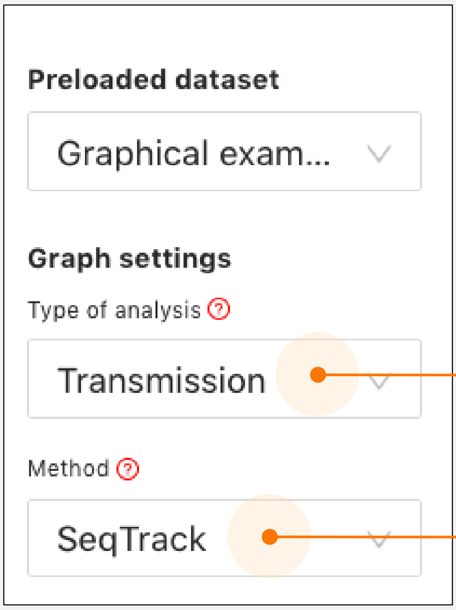
A cluster is defined by identifying node within the connected component/s, which created by removing edges greater than the threshold.



sample	clusterID
1	1
2	1
4	1
5	1
6	1
3	na

The clustering result can be downloaded as ***a CSV file.***

Transmission analysis



Preloaded dataset

Graphical exam...

Graph settings

Type of analysis: Transmission

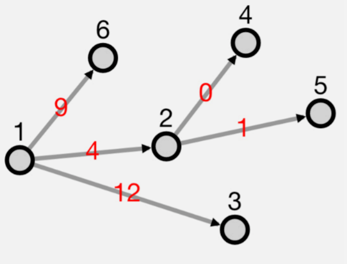
Method: SeqTrack

Select Clustering

Select reconstruction method

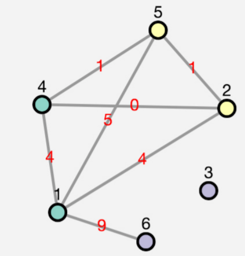
Performing transmission analysis is similar to cluster analysis. Users only need to select Transmission instead of Clustering. Currenltly, only one method is implemented: SeqTrack [1]

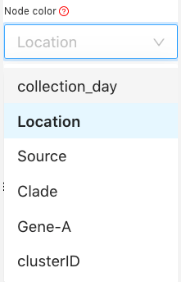
1. Jombart, T., et al., *Reconstructing disease outbreaks from genetic data: a graph approach.* Heredity (Edinb), 2011. **106**(2): p. 383-90.



Most parsimonious transmission tree created using SeqTrack algorithm.

Graph settings

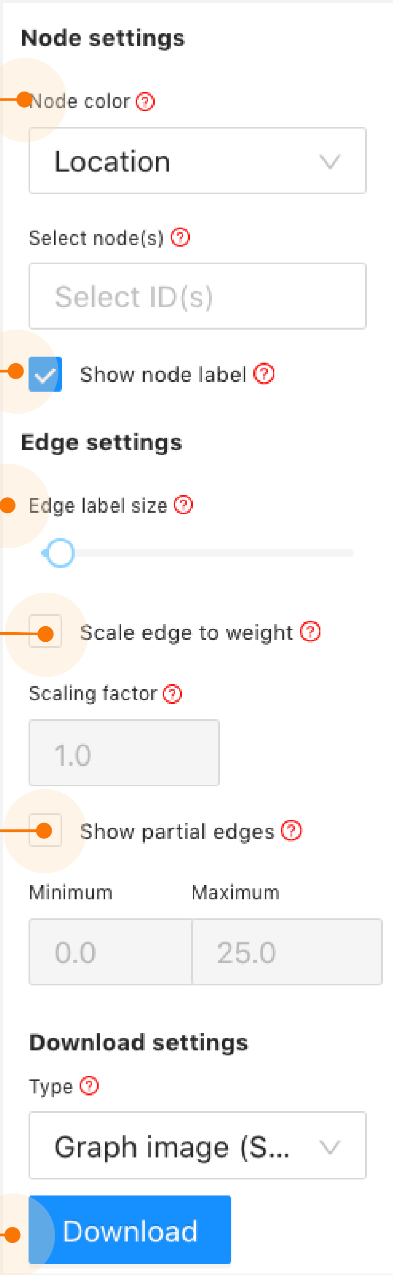




Node color: Location

Color nodes by the selected column in metadata or by the clustering result.

Here, we color the nodes based on Location column in metadata.



Node settings

Node color: Location

Select node(s): Select ID(s)

☑ Show node label

Edge settings

Edge label size

☐ Scale edge to weight

Scaling factor: 1.0

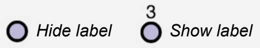
☐ Show partial edges

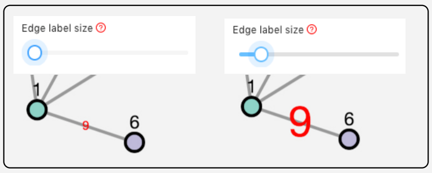
Minimum: 0.0 Maximum: 25.0

Download settings

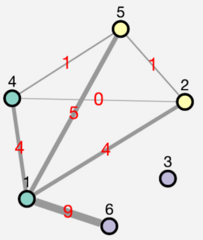
Type: Graph image (S...

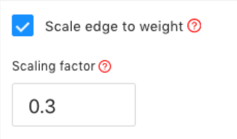
Show or hide node's label.





Change edge label size



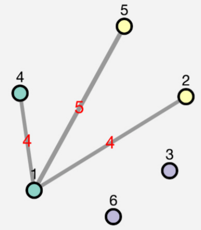


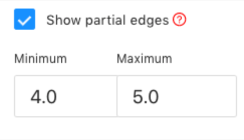
☑ Scale edge to weight

Scaling factor: 0.3

Here, we scale the edges with factor of 0.3

Change the thickness of the edge according to its weight. (e.g., the higher the SNP distance the thicker the line).





☑ Show partial edges

Minimum: 4.0 Maximum: 5.0

Here, only edges with distance 4 and 5 were displayed.

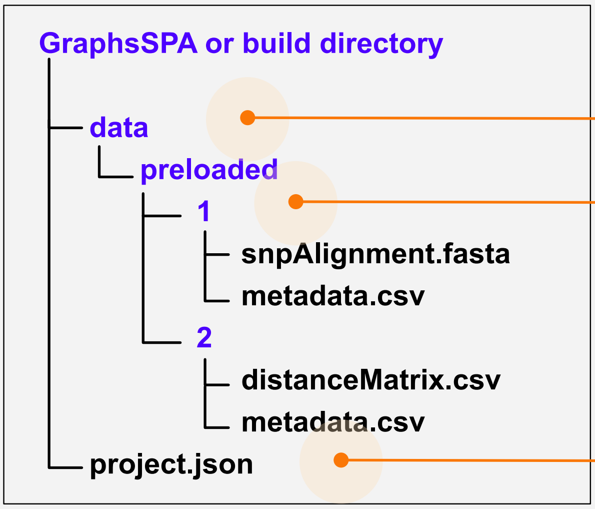
Only show edges which have weight within the specified range (min to max) (Note: It doesn't remove the edges but only hide it to the background)

Download Graph image (SVG or PNG) or Graph file (DOT) or clustering result (CSV)

Setting up preloaded dataset

When users use GraphSNP offline, they can set up multiple preloaded datasets. This feature allows users to ‘permanently’ link their input files to GraphSNP, avoiding the need to re-inputting their input files every time the browser refreshed.

Example of directory tree of GraphSNP preloaded datasets



```
GraphsSPA or build directory
│
├─ data
│  └─ preloaded
│     ├─ 1
│     │  ├─ snpAlignment.fasta
│     │  └─ metadata.csv
│     └─ 2
│        ├─ distanceMatrix.csv
│        └─ metadata.csv
└─ project.json
```

1 **Go to directory of *data* in GraphSNP SPA**

2 **Create a new directory in *preloaded* directory then add the input files.**

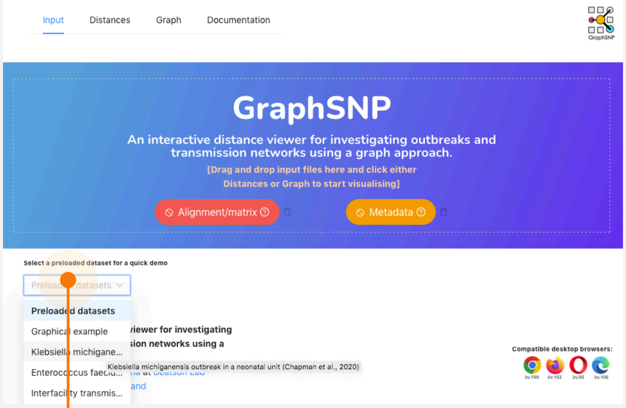
3 **Update project.json file**

Example project.json content

Add the dataset ID and input files path to project.json and save the file.

```
{
  "projects": [
   {
     "id": "1",
     "name": "Dataset 1: Graphical example",
     "matrixOrAlignment": "alignment",
     "snpDistance": "./data/preloaded/1/snpAlignment.fasta",
     "metadata": "./data/preloaded/1/metadata.csv"
   },
   {
     "id": "2",
     "name": "Dataset 2: NCBI Cluster of VREfm ST78",
     "matrixOrAlignment": "matrix",
     "snpDistance": "./data/preloaded/2/distanceMatrix.csv",
     "metadata": "./data/preloaded/2/metadata.csv"
   }
  ],
  "description": "This JSON file describes preloaded datasets to be rendered in the landing page. The path of these files must be written with directory 'public' as the root (e.g. ./data/ means 'data' is inside directory 'public'"
}
```

4 **Datasets is listed in GraphSNP *input* page**



GraphSNP

An interactive distance viewer for investigating outbreaks and transmission networks using a graph approach.

[Drag and drop input files here and click either Distances or Graph to start visualising]

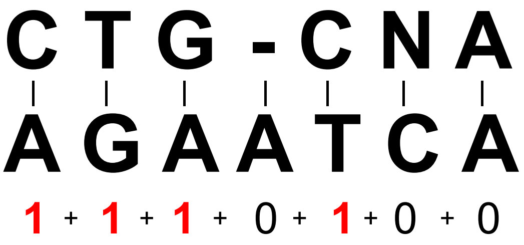
Select a preloaded dataset for a quick demo

Klebsiella michiganensis outbreak in a neonatal unit (Chapman et al., 2020)

Click the preloaded dataset dropdown button and select dataset of interest and GraphSNP will automatically load the input files.

Extra

1. How does GraphSNP determine the Hamming distance between two sequences?



Given two strings of equal length, GraphSNP counts the number of mismatches (differences) between the corresponding positions containing A, T, G, or C characters. Positions that containing any other characters are omitted from the counting.

Total distance = 4

2. What can I do if the GraphSNP distance calculation is not appropriate for my data?

GraphSNP also support a distance matrix input. It is recommended that users compute the distance using their preferred method and save the result as a distance matrix file. The following are several examples to consider:

#Using ape R pacakge

```
library(ape)
#read alignment file
seq <- read.dna('snps_alignment.fasta', format = "fasta")
#convert to distance matrix
dist <- as.data.frame(dist.dna(seq, model = 'N', as.matrix = T))
#adjust rownames
dist_GraphSNP <- cbind('rowCol' = rownames(dist), dist)
#write out distance matrix to a CSV file
write.csv(dist_GraphSNP, file = "GraphSNP_distanceMat.csv",
row.names = F)
```

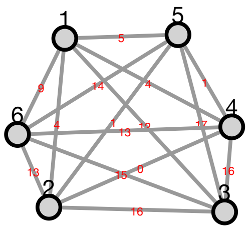
#Using snp-dists

```
#Create tool environment and
install snp-dists (https://
github.com/tseemann/snp-dists)
conda create -n snp_dists_env -c
conda-forge snp-dists
#Activate the environment
conda activate snp_dists_env
#Generate distance matrix and
output a CSV file
snp-dists -c
snps_alignment.fasta >
GraphSNP_distanceMat2.csv
```

3. How does GraphSNP generate a threshold-based cluster MST minimum spanning tree?

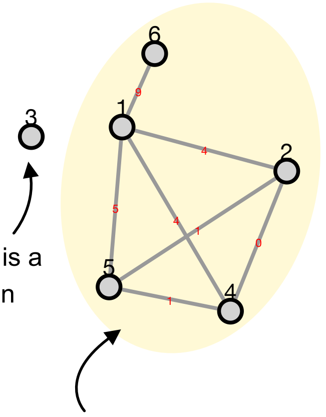
A threshold-based cluster MST refers to an MST of the identified cluster/s given a threshold, meaning this MST was constructed **AFTER** the cluster/s is defined. It involves 2 main steps: Identification of cluster/s (given a threshold) and constructed the MST between those clusters and or singletons.

A complete graph



A complete graph built from paiwise distance matrix.

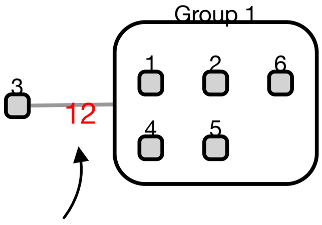
Step 1. Cluster definition (cut-off = 9)



Node 3 is a singleton

Node 1, 2, 4, 5, and 6 are assigned into one cluster (one connected component) using breadth-first search algorithm to identify connected components.

Step 2. MST construction



Kruskal's algorithm is applied to evaluate minimum distance edges between clusters, between singletons and between cluster to singletons to create an MST.

React Data-Driven Documents

Ant Design Cytoscape

THANK YOU

for reading this manual

Thanks to all awesome web frameworks and libraries run on the background, GraphSNP is now up and running and available worldwide. The following are some of the core libraries used by GraphSNP:

react
d3
antd
cytoscape
cytoscape-svg
redux
react-color
lodash
moment
moment-range
...

GraphSNP