Short Tutorial

Step 0: Start AntMap

Fig. 1

When you use AntMap on Windows, start AntMap with double-clicking the "AntMap" icon (Fig. 1). For other operating systems (i.e., platforms), See below.

Box 1.

Linux and Solaris

Before executing "AntMap-linux" or "AntMap-solaris", you should change mode of these files to be executable. To do that, type

```
chmod 755 AntMap-xxxx
```

on your command line system ("`xxxx`" should be "`linux`" or "`solaris`"). After changing the mode of files, you can execute AntMap by clicking the "AntMap-linux" or "AntMap-solaris" file icon.

Mac OS X

Note that you can execute "AntMap-macx" from the command line, but cannot execute by clicking the "AntMap-macx" file icon.

AntMap can also be executed by using the executable jar file "AntMap.jar" on any platforms (Linux, Solaris and Mac OS as well as Windows). To execute the jar file, run:

```
java –jar AntMap.jar
```

on your command line system. Some platforms may have bindings already set up such that you can execute the jar file just by clicking on the "AntMap.jar" file icon, which will run the command line equivalent. Note that you should change mode of the jar file to be executable when you are on "Linux" or "Solaris" platforms as described in Box 1.

Step 1: Open an input file.

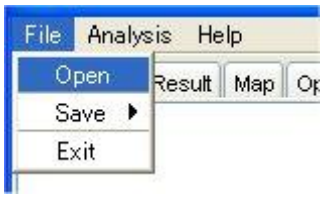

Fig. 2

Open an input file in MapMaker format (*.raw) through “File-Open” menu (Fig. 2). Here, open “sample.raw” contained in the “antmap” folder.

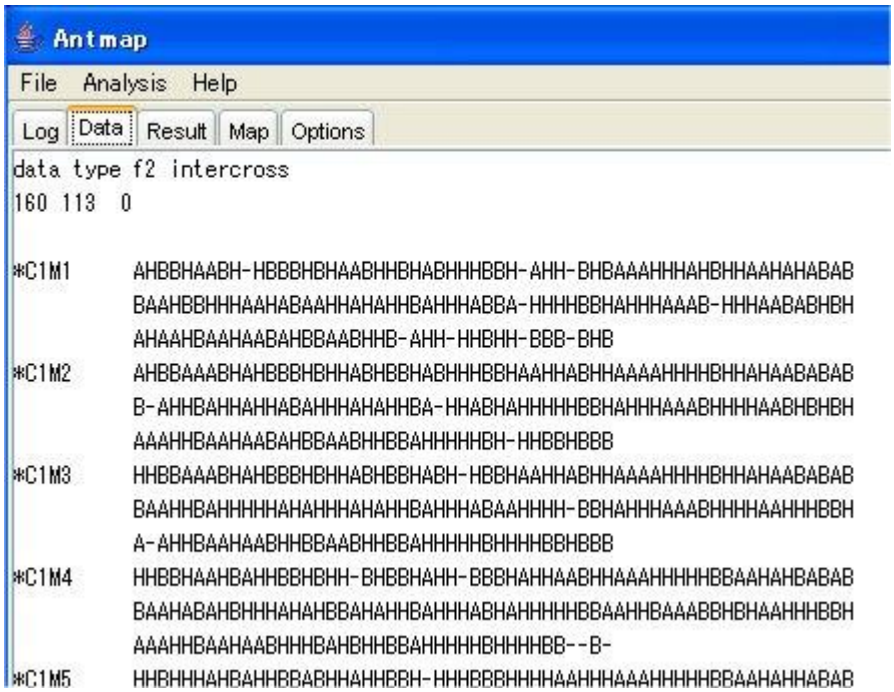

Fig. 3

After opening the file, contents of the file will appear in the “Data” panel (Fig. 3).

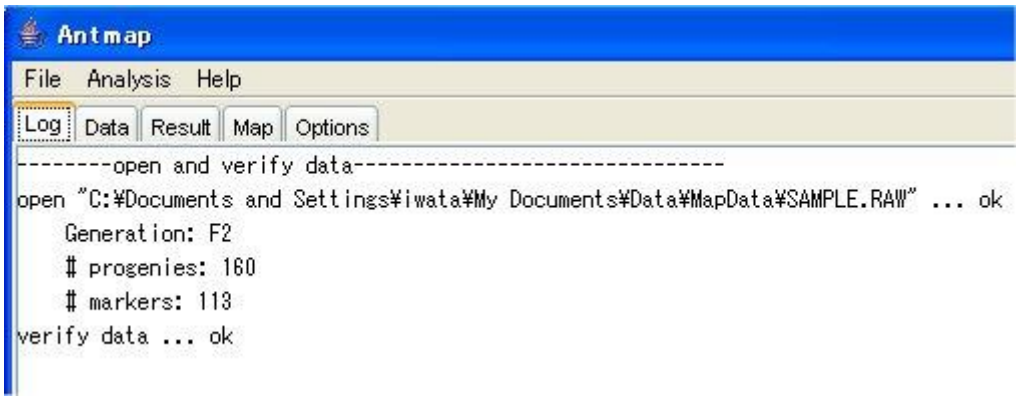

Fig. 4

Click the “Log” tab, and you can see a summary of the input data (Fig. 4).

Step 2: Segregation ratio test.

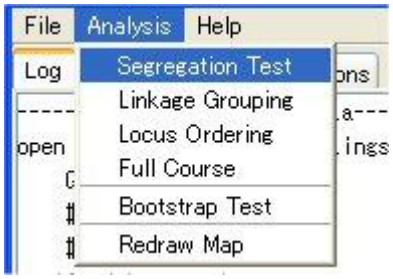

Fig. 5

Select “Segregation Test” from the “Analysis” menu (Fig. 5). Then you can see the results of segregation ratio tests in the “Result” panel (Fig. 6).

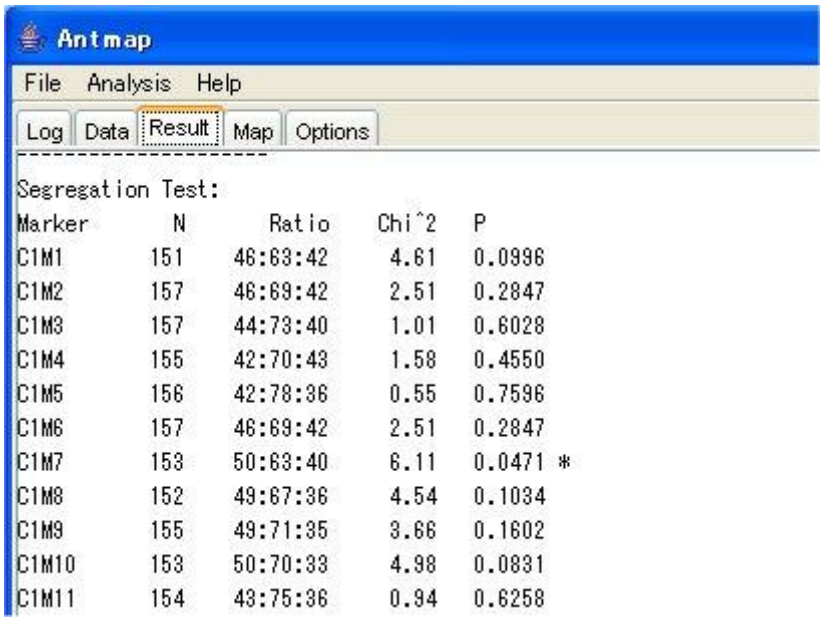

Fig. 6

Step 3: Linkage grouping

Fig. 7

Click the “Options” tab. Then you can see the “Grouping” option panel (Fig. 7).

You can choose one of the two grouping methods: “nearest neighboring locus” and “all combinations”. The former makes a group by sequentially combining a locus which shows the smallest recombination value against it. This algorithm has been implemented by MAPL (Ukai et al. 1991). The latter will produce similar results with “group” command of MapMaker.

You can also choose the grouping criterion, threshold value and the minimum number of markers for a single group.

Here, we will keep these options unchanged except for the threshold value.

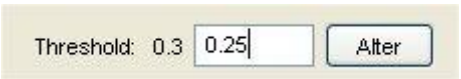

Fig. 8

Type “0.25” into an input area, and push the “Alter” button. Then you can change the threshold value from 0.3 to 0.25.

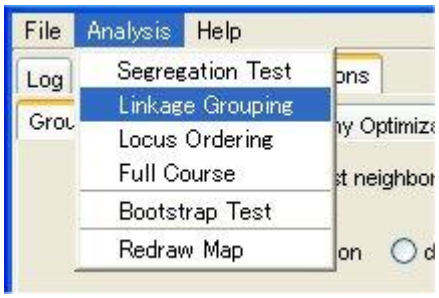

Fig. 9

Select the “Linkage Grouping” from the “Analysis” Menu. Then you can see the results of linkage grouping in the “Result” panel (Fig. 10).

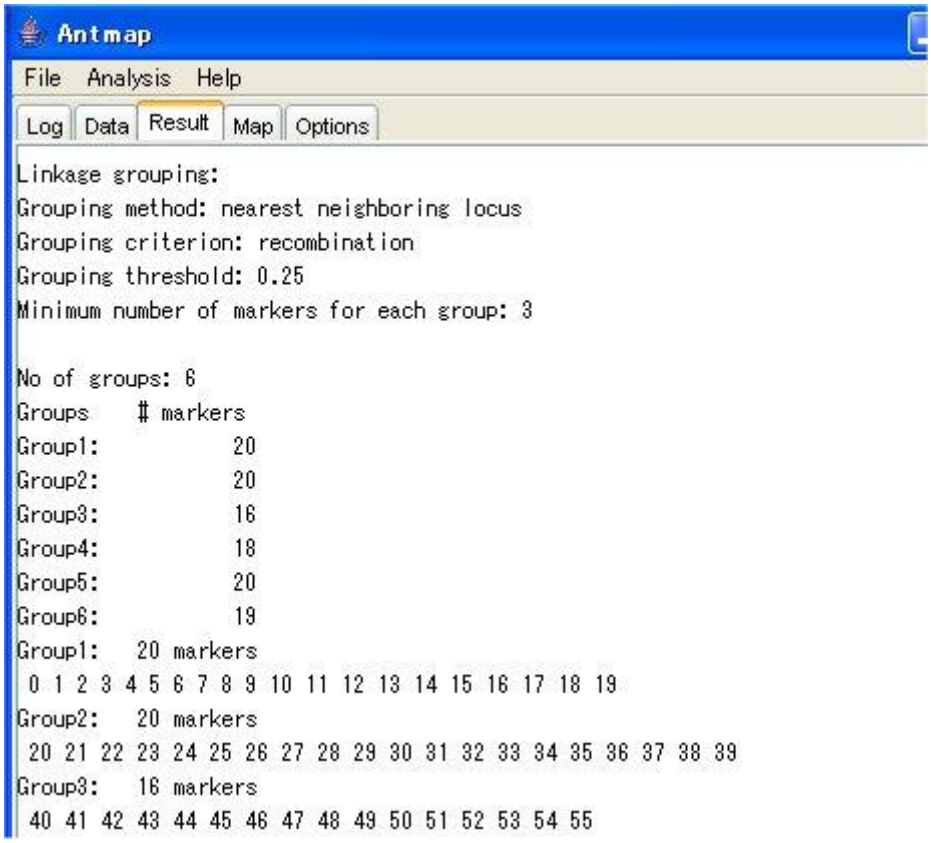

Fig. 10

When you analyze your data, you may not be able to achieve a good separation of markers to linkage groups from the start. In such a case, please find a good set of the threshold value, criterion and method through try-and-errors.

Step 4: Locus ordering

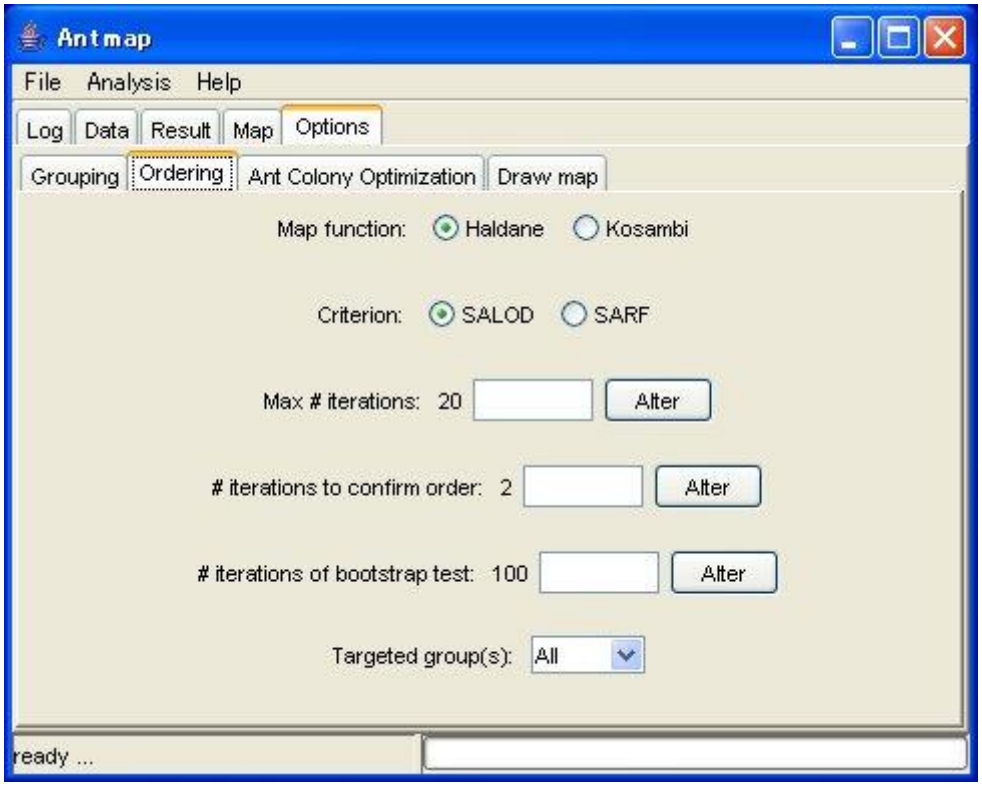

Fig. 11

Click the “Options” tab, and click the “Ordering” tab. Then you can see the “Ordering” option panel (Fig. 11).

In the locus ordering, you can choose one of the two criteria: log-likelihood and “SARF”. “SARF” is an abbreviation for “Sum of Adjacent Recombination Fractions” (Liu 1998). AntMap will search a locus order which maximizes log-likelihood or minimizes “SARF”.

You can also choose the maximum number of iterations and the number of iterations to confirm order. The details of these options are given in the “AntMap Options” section.

A map function for calculating a map distance between adjacent markers can be selected from “Haldane” (Haldane 1919) or “Kosambi” (Kosambi 1944) functions.

Here, we will keep these options unchanged.

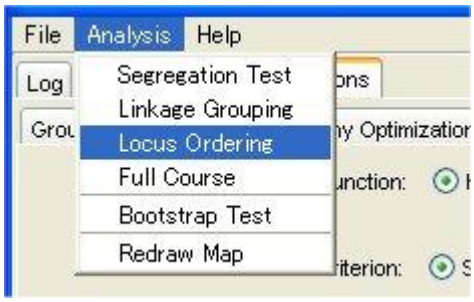

Fig. 12

Select the “Locus Ordering” from the “Analysis” Menu. Then you can see the results of locus ordering in the “Result” panel (Fig. 13).

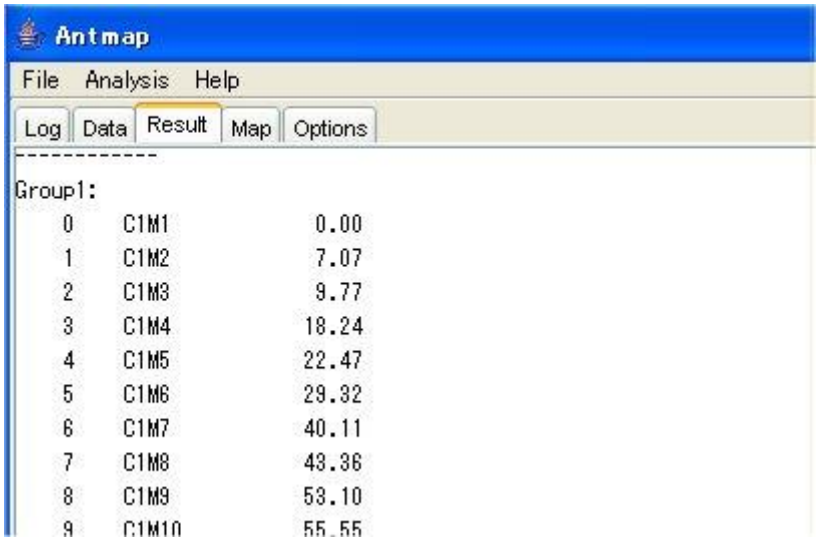

Fig. 13

You can also obtain a graphic of linkage map in the “Map” panel (Fig. 14).

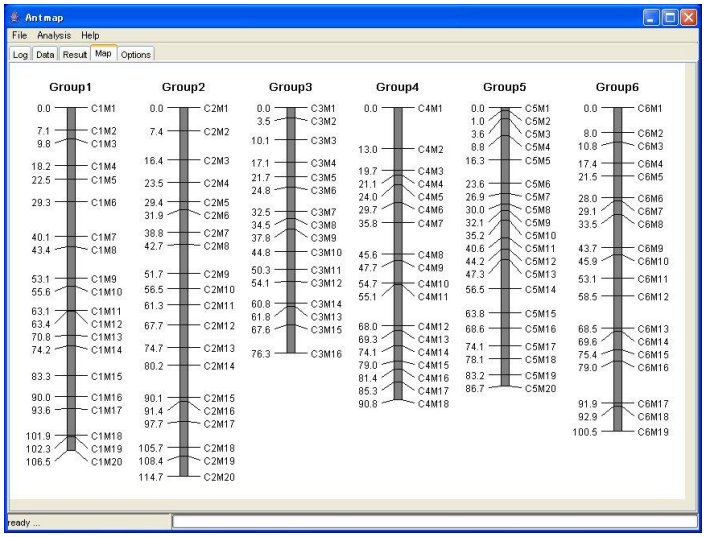

Fig. 14

Step 5: One-step mapping

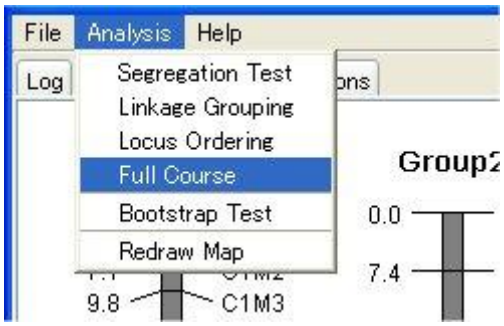

Fig. 15

Select “Full Course” from the “Analysis” Menu. Then, you can overall process from segregation ratio test (Step 2) to locus ordering (Step 4) at once.

Step 6: Redraw a linkage map

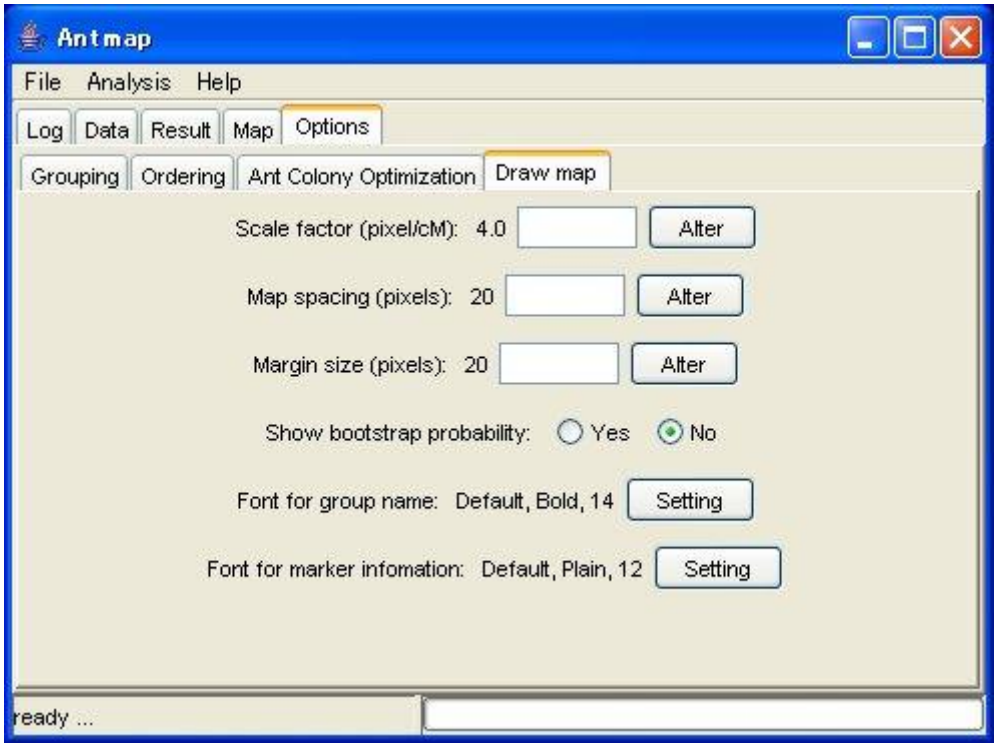

Fig. 16

Click the “Options” tab, and click the “Draw map” tab. Then you can see the “Draw map” option panel (Fig. 16).

Here, we will change the “Scale factor” option. Drawing size of linkage map can be changed through this option. Here, type “2” into an input area, and click the “Alter” button (Fig. 17).

Fig. 17

After changing the option value from 4 to 2, select “Redraw Map” from the “Analysis” menu. Then, you can obtain a smaller linkage map than one obtained previously (Fig. 18).

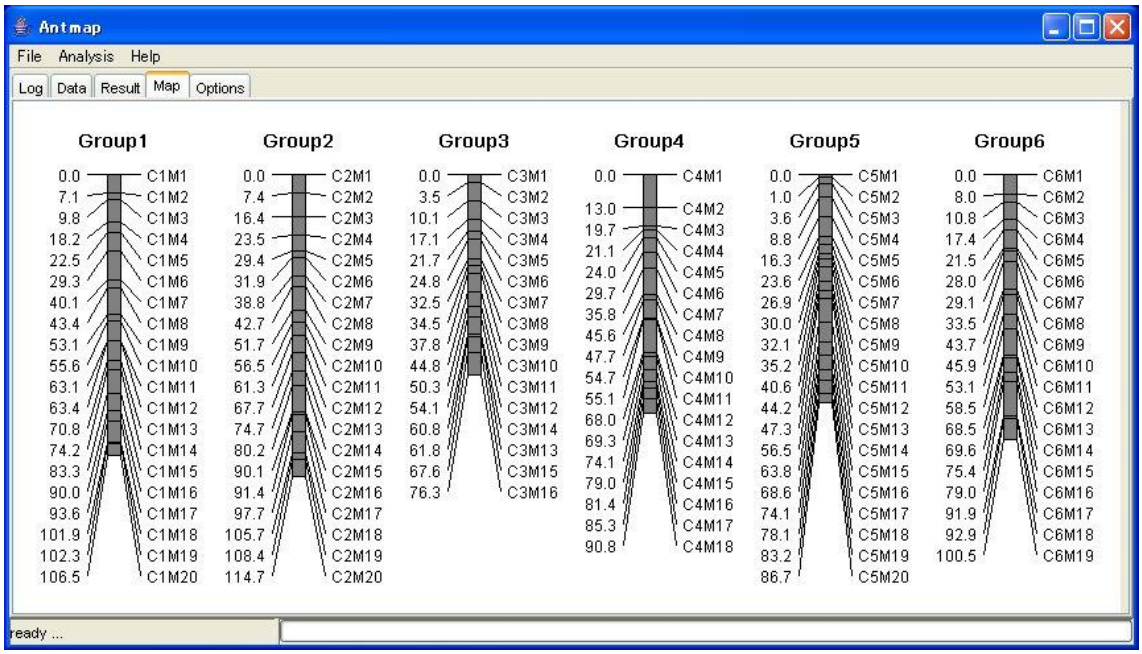

Fig. 18

Step 7: Bootstrap test for locus order

You can evaluate the reliability of estimated locus order by using bootstrap test. Bootstrap test (or bootstrapping) is a method for estimating the sampling distribution of an estimator by resampling with replacement from the original sample. In a bootstrap test, a random sample of size *n* is drawn from the original sample of size *n*, and estimates are obtained from the random sample. After repeating (iterating) this operation many times (e.g., 100-1000 times), the stability of estimates (e.g., standard error or confidence interval of estimators) is evaluated. For the details of bootstrap test, please see a good textbook such as Manly (1998). In the bootstrap test for locus order, we can obtain probability that a locus is located at its estimated order (Liu 1998).

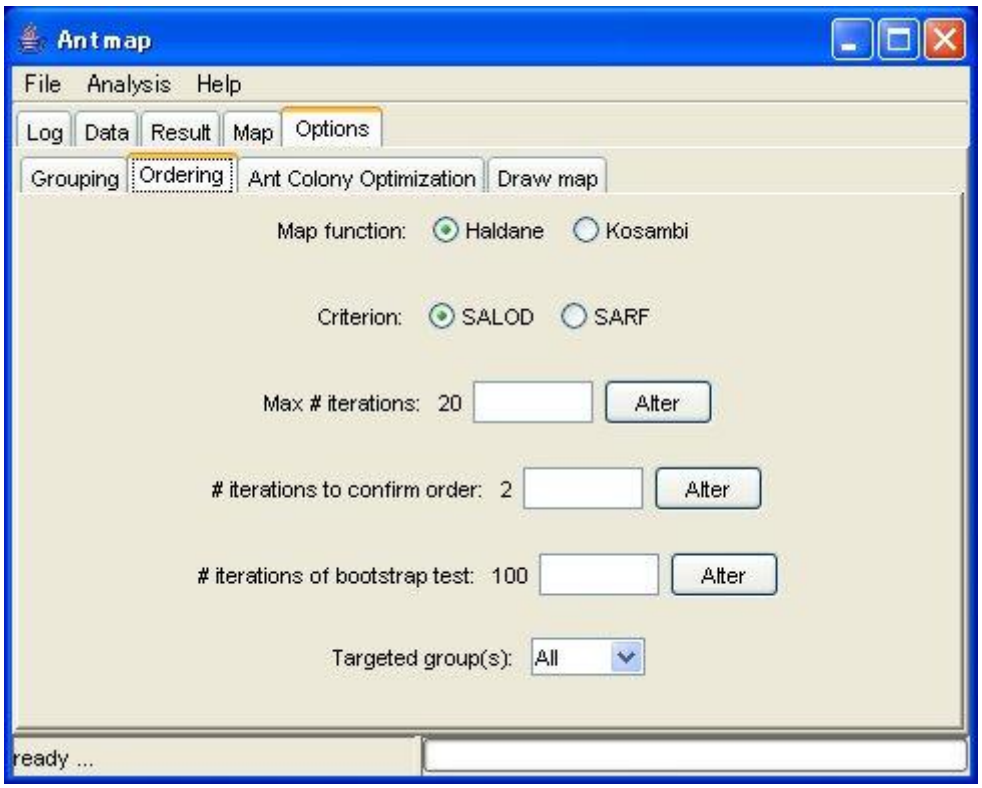

Fig. 19

Click the “Options” tab, and click the “Ordering” tab. Then you can see the “Ordering” option panel (Fig. 19).

You can change the number of iterations (repeats) of bootstrapping. To get a good estimate of percentage of correct locus order, 100 may be sufficient.

You can also choose a group which is targeted in the bootstrap test. Here, we will choose only Group3 to save our time (Fig. 20).

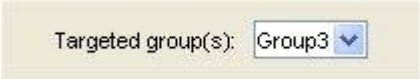

Fig. 20

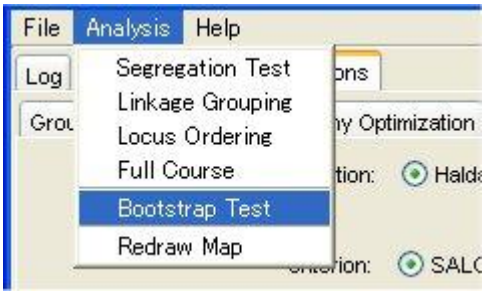

Fig. 21

Select the “Bootstrap Test” from the “Analysis” Menu (Fig. 21). Then you can see the results of bootstrap test for locus order in the “Result” panel (Fig. 22).

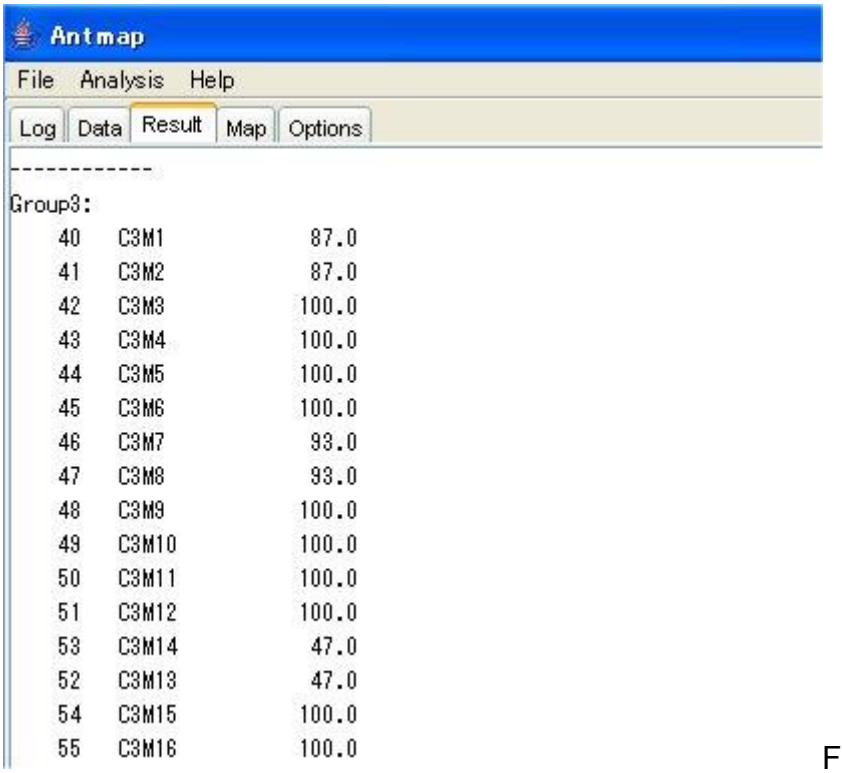

Fig. 22

You can also obtain a graphic of linkage map with bootstrap values in the “Map” panel (Fig. 23).

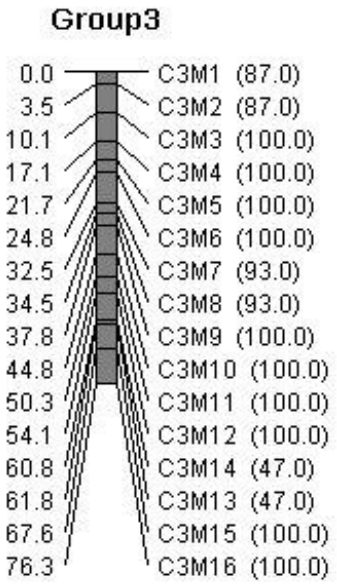

Fig. 23

FYI: The bootstrap test for all linkage groups may take long time even by high-end PC. Thus, you have better set your computer to perform this test at your lunch time or after going home.

Step 8: Save results of linkage mapping

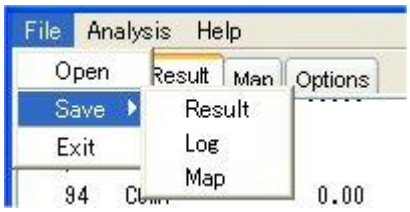

Fig. 24

You can save information in “Result”, “Log” and “Map” panels through the “Save” submenu in the “File” Menu. The information in “Result” and “Log” is saved as a text file. The information in “Map” (i.e., a graphic of linkage map) is saved as a JPEG (*.jpg) file.