# **Supplementary information**



### Model A:

The haplotypes from the ice core were assigned to 12,500 years before present. The haplotypes from current glacier surface were assigned to 0 years before present.



## Model B:

The haplotypes from the ice core were assigned to 0 years before present. The haplotypes from current glacier surface samples were assigned to 0 years before present, except for randomly selected N haplotypes assigned 12,500 years before present, where N is the number of haplotypes from the ice core sample.

The haplotypes from the ice core



### Model C:

The haplotypes from the ice core were assigned to 0 years before present. The haplotypes from current glacier surface were assigned to 12,500 years before present.





## Model D:





The phylogenetic tree with the branch lengths estimated by the ML method without assuming any clock models.

Supplementary Figure S1: Comparisons and detailed explanations of the clock models used in this study. (A) Model A, in which the haplotypes from the ice core were assigned to 12,500 years before present, and the haplotypes from current glacier surface were assigned to 0 years before present. This is a modeling of the situation that all haplotypes from the ice core are real ancient samples, and all from current glacier surface are originated from modern populations. No contamination occurred from the modern populations. (B) Model B, in which the haplotypes from the ice core were assigned to 0 years before present. The haplotypes from current glacier surface samples were assigned to 0 years before present, except for randomly selected N haplotypes assigned 12,500 years before present, where N is the number of haplotypes from the ice core sample. This is a modeling of the situation that all haplotypes are originated from the modern populations, but some haplotypes were mistakenly assigned as 'ancient' by the contamination. (C) Model C, in which the haplotypes from the ice core were assigned to 0 before present, and the haplotypes from current glacier surface were assigned to 12,500 years before present. This model is the extreme case of the Model B. (D) Model D, in which all haplotypes were assigned to be modern (0 years before present). This is a modeling of the situation that all haplotypes are originated from the modern populations, and all samples were 'correctly' assigned as the modern populations. (E) The phylogenetic tree, in which the branch lengths were estimated by the ML method without assuming any clock models. The topologies and the branch lengths shown in panel (A) to (E) were estimated based on the OTU9. The haplotypes from the ice core were indicated by the blue rectangular symbols. If the time interval of 12,500 years is too short to accumulate the significant numbers of the mutations in ITS region, it can be expected that the fitness of Model A - Model D to the nucleotide alignment data are not significantly different. If the time interval of 12,500 years has evolutionary significant meaning in ITS regions, the comparisons of Model A - Model D can be used to detect the contaminations. In case of that all haplotypes are originated from the modern samples, Model D should be the best model and Model A - Model C have no significant differences.



**Supplementary Figure S2**: Cyanobacterial taxonomic classification of shotgun library reads composition in the Bot samples based on 16S rRNA gene sequences.





Supplementary Figure S3: Taxonomic classification of shotgun library reads composition in the ice core samples based on 16S and 18S rRNA gene analysis. Classification was performed by filtered high quality Illumina reads using Metaxa2 at phylum level. The read numbers of the 16S rRNA gene in Ice-dust and Bot were 90 and 4,094, respectively, which corresponded to  $3.7 \times 10^{-4}$  and  $1.5 \times 10^{-2}$  % of the total reads, respectively. The majority of the 16S rRNA genes in the Ice-dust sample were affiliated with Betaproteobacteria, Bacteroidetes, and Gammaproteobacteria, and those in the Bot sample were affiliated with Bacteroidetes, Betaproteobacteria, Gammaproteobacteria, and Verrucomicrobia.



**Supplementary Figure S4**: The maximum likelihood estimation of the demographic movements and the time of the most recent common ancestor (tMRCA) by the GENETREE program under the exponential growth model in OTU4 and OTU9 in cyanobacteria.