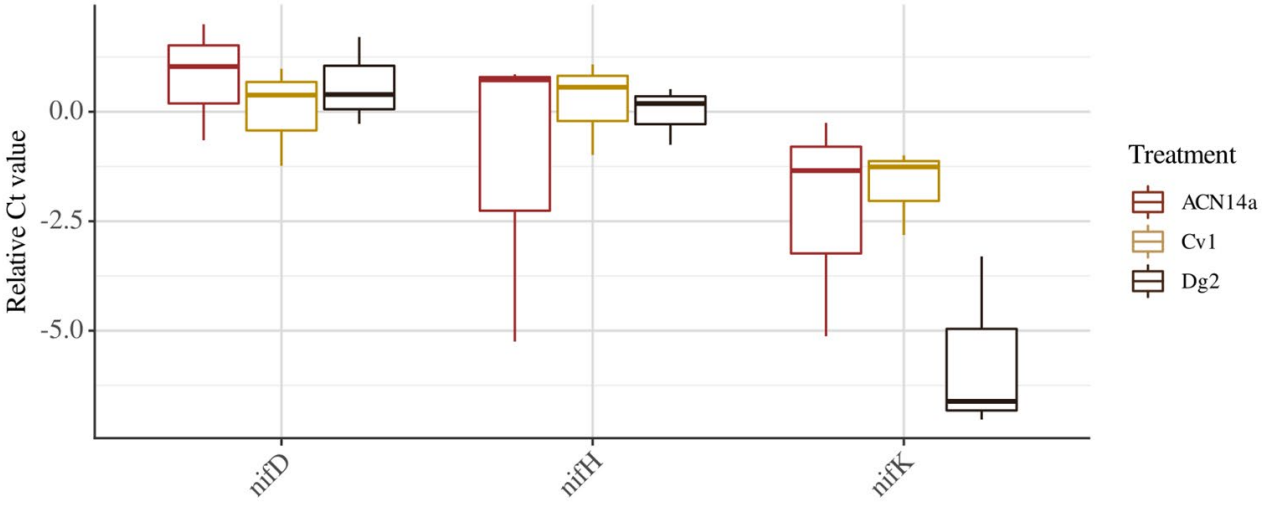
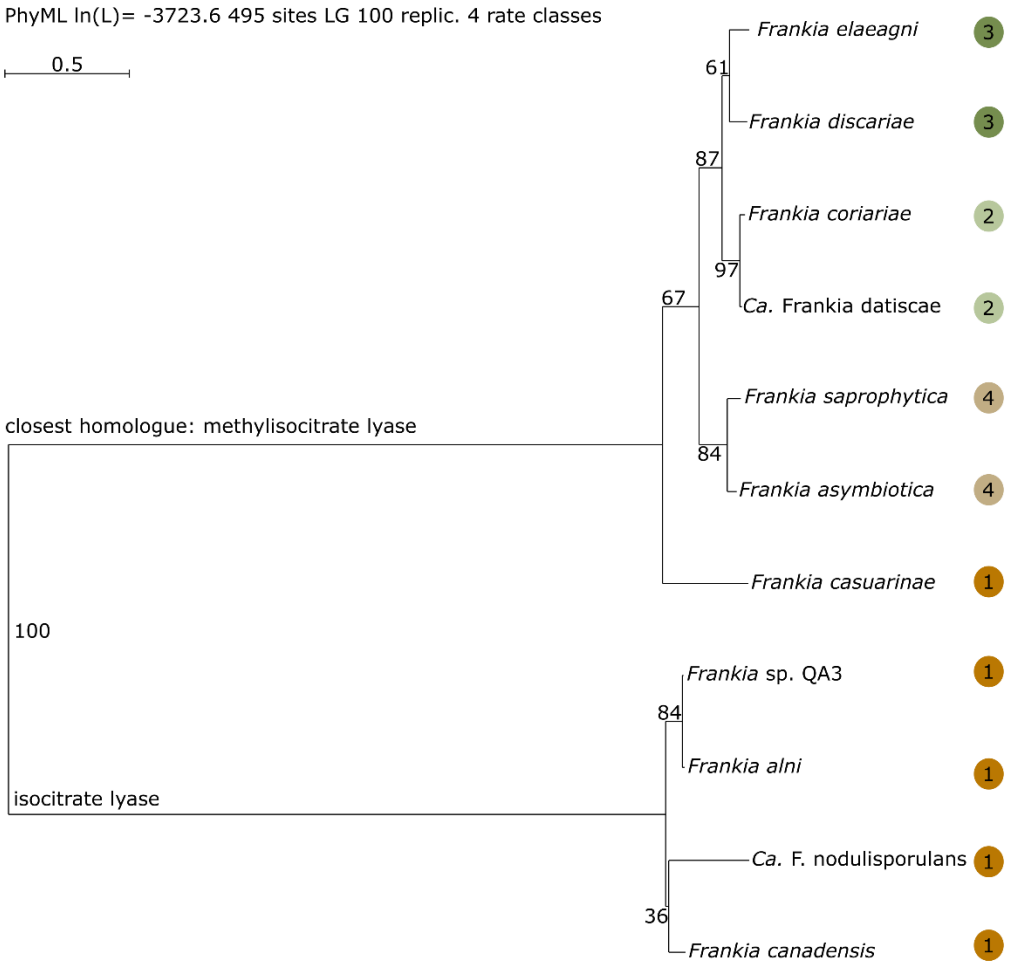
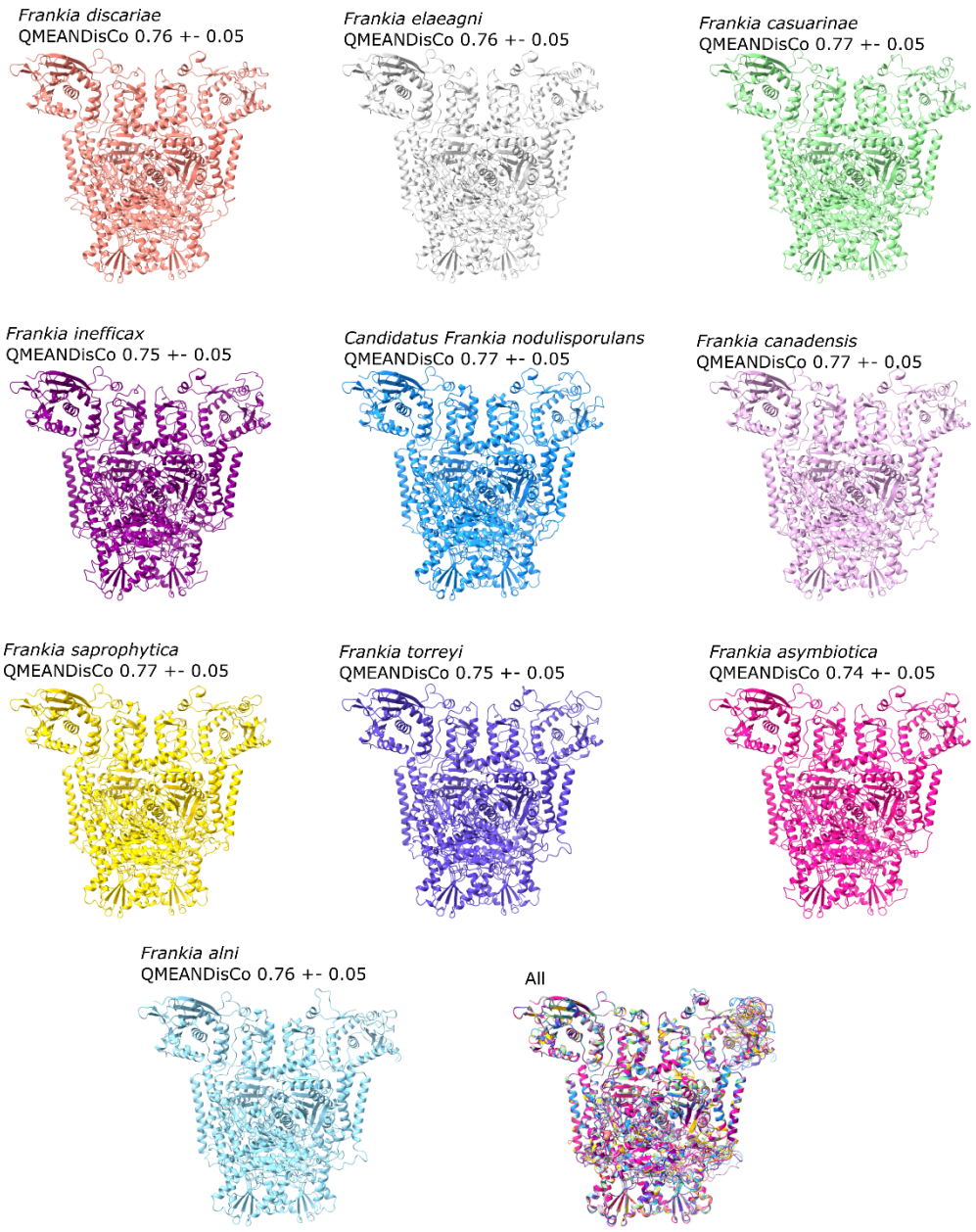


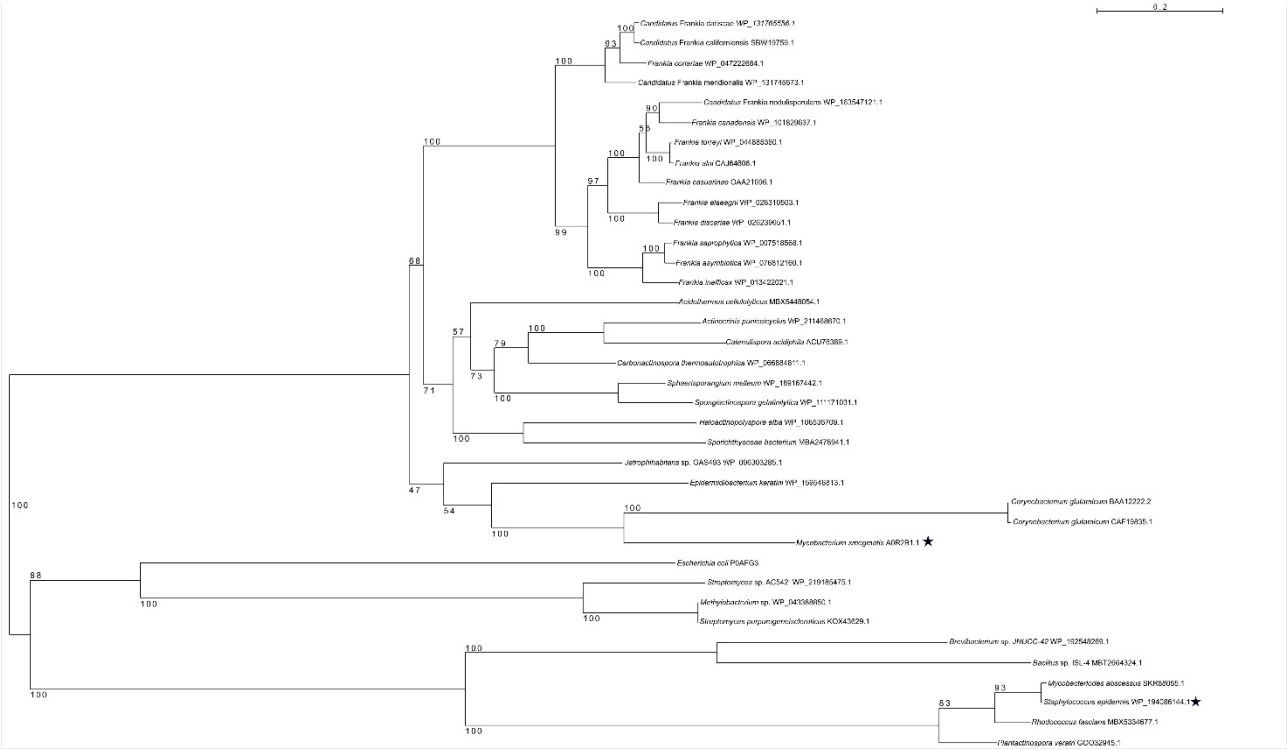
Supplementary Figure S1: Relative expression levels (delta Ct value) of the *nif* genes encoding the subunits of nitrogenase complex. The Ct value is normalised against the gene *infC*, encoding the translation initiation factor IF-3 (Alloisio *et al.,* 2010). An asterisk indicates a significant difference ($p < 0.5$), based one-way ANOVA of gene expression of four technical repeats of three biological repeats of nodules from *Alnus glutinosa* induced by *Frankia alni* ACN14a (red, left), *Ceanothus thyrsiflorus* induced by *Candidatus* Frankia californiensis Cv1 (yellow, centre), and *Datisca glomerata* induced by *Candidatus* Frankia californiensis Dg2 (brown, right).



Supplementary Figure S2: ML tree of isocitrate lyase gene, based on amino acid sequences. Isocitrate lyase sequences was taken from *Frankia alni* ACN14a genome and a BLAST search was performed on other *Frankia* genomes in NCBI. Amino acid sequences were aligned using clustal omega, and maximum likelihood tree with 100 bootstraps was built, using SeaView 5.0 (Gouy et al., 2021). The closest hit for several genomes of different clusters was found to be methylisocitrate lyase. Accession numbers are given in **Supplementary Table S2.** In color and with number, are the different clades to which each *Frankia* species belongs given. According to Dunn et al. (2009), methylisocitrate lyase cannot perform the same reaction as isocitrate lyase.



Supplementary Figure S3: models of 2-oxoglutarate decarboxylase in different *Frankia* species Models were built using SWISS-MODEL portal and visualised in ChimeraX. The global QMEANDisCo for each model is given.



Supplementary Figue S4: RAxML Maximum likelihood phylogeny for 2-oxoglutarate dehydrogenase and 2-oxoglutarate decarboxylase. Sequences were aligned with clustalO in Seaview 5.0 (Gouy et al., 2021). Rapid bootstrapping was performed using 100 bootstrap. LG substitution matrix was used. Black asterisks indicate the position of 2-OG decarboxylase from *Mycobacterium smegmatis* (reference A0R2B1) and 2-OG dehydrogenase of *Staphylococcus epidermis* (reference Q5HPC6), respectively. Sequence references are given after each name.

References used:

Alloisio, N., Queiroux, C., Fournier, P., Pujic, P., Normand, P., Vallenet, D., Médigue, C., Yamaura, M., Kakoi, K., and Kucho, K. (2010). The Frankia alni symbiotic transcriptome. Molecular Plant-Microbe Interactions, 23(5), 593–607.

Dunn, M. F., Ramírez-Trujillo, J. A., and Hernández-Lucas, I. (2009). Major roles of isocitrate lyase and malate synthase in bacterial and fungal pathogenesis. Microbiology (Reading, England), 155(Pt 10), 3166–3175.

Gouy, M., Tannier, E., Comte, N., and Parsons, D. P. (2021). Seaview version 5: A multiplatform software for multiple sequence alignment, molecular phylogenetic analyses, and tree reconciliation. Multiple Sequence Alignment: Methods and Protocols, 241–260