An effective approach to retrieve culturable *Acinetobacter* spp. from the soil environment

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AIM

To design and test an effective approach to recover taxonomically diverse *Acinetobacter* strains from environmental soil samples.

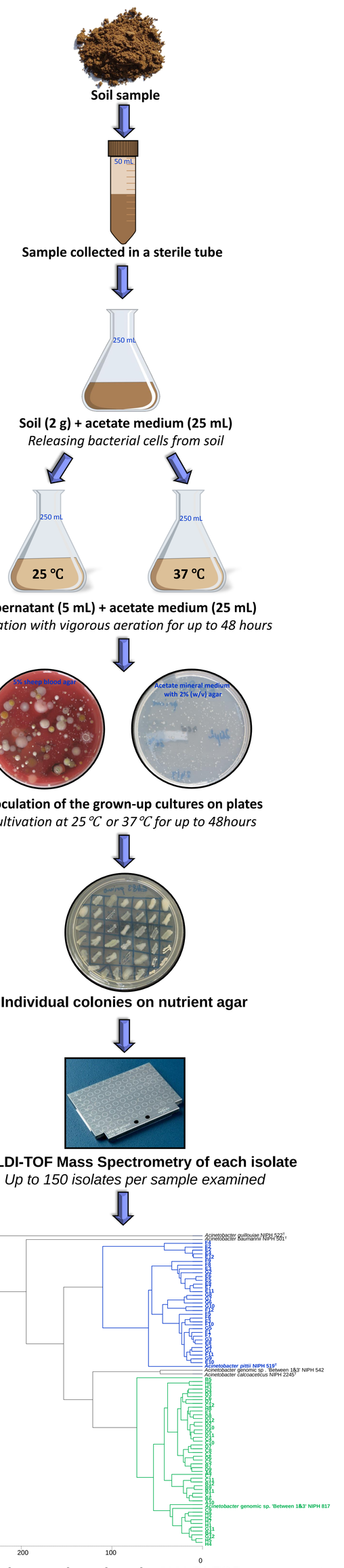

Fig. 1. A stepwise approach for a soil sample treatment and for the identification of *Acinetobacter* isolates.

SAMPLES

Samples **A**, **B** and **C**, collected during 2014, originated from a tropical rainforest in Sumatra (Indonesia) and two protected wetland areas in the Czech Republic, respectively.

METHODS

Samples were processed as shown in **Fig.1**.

MALDI-TOF MS identification was performed by matching the obtained spectra to those of the current Bruker Daltonics database supplemented with homemade entries representing all known and provisional *Acinetobacter* spp.[1]

STRAINS

The final classification of strains at the species level was based on the comparative analysis of the *rpoB* gene, MALDI-TOF MS and comprehensive physiological and metabolic testing (**Fig. 2**).

INTRODUCTION

A basic prerequisite for understanding the role of a given group of bacteria in ecological processes is a comprehensive knowledge of their physiological and metabolic features at the species level. To gather such information, obtaining pure bacterial cultures and their laboratory characterization are inevitable.

Bacteria of the ubiquitous genus *Acinetobacter* play an important role in biological processes in soil ecosystems. However, the taxonomic diversity of these bacteria in natural environments is largely unknown.

RESULTS

Sample **A** yielded 11 unique *Acinetobacter* strains. Of these, four were allocated to three known species while seven represented novel species (**Fig. 2**).

Sample **B** showed an extremely high species diversity: 16 strains were classified as seven known and five new species (**Fig. 2**).

Sample **C** produced 10 strains belonging to three known and three novel species (**Fig. 2**).

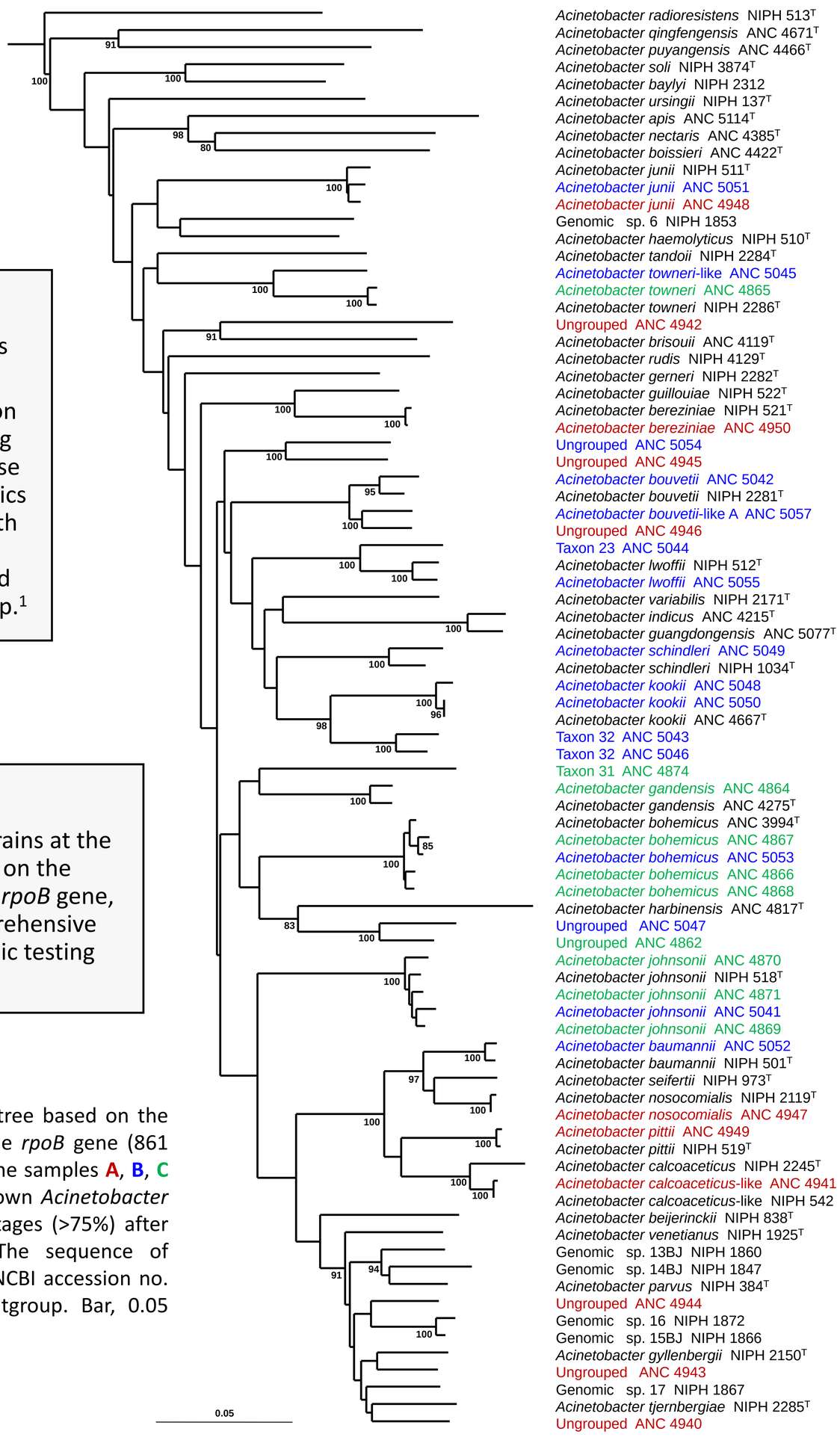

Fig. 2. Rooted neighbour-joining tree based on the partial nucleotide sequence of the *rpoB* gene (861 nt) of environmental strains of the samples **A**, **B**, **C** and the representatives of all known *Acinetobacter* spp. (in black). Bootstrap percentages (>75%) after 1000 simulations are shown. The sequence of *Pseudomonas aeruginosa* PAO1 (NCBI accession no. NC002516) was used as the outgroup. Bar, 0.05 substitutions per nucleotide site.

CONCLUSION

We have developed an effective approach which allows for quick taxonomic screening of hundreds of colonies and enables effective recovery of taxonomically diverse *Acinetobacter* strains from environmental samples.

References: [1]Touchon et al. 2014; *Genome Biol Evol* 6: 2866-2882.

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