

Evaluation of the ISO standard 11063 “Soil DNA Extraction Procedure” for Assessing Microbial Abundance and Community Structure

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For the last two decades, soil microbial diversity studies have been dependent upon the extraction and characterization of soil DNA. Therefore, the DNA extraction procedure has become a critical step in describing soil microbial biodiversity. Historically, ascertaining overarching microbial ecological theories has been hindered as independent studies have used numerous custom and commercial DNA extraction procedures which affect the assessments of diversity and community composition. For that reason, a standardized soil DNA extraction method (ISO 11063) was developed. However, this standard has only been optimized for examining soil bacteria, and not for other soil microbes (archaea and fungi). Similar to other extraction procedures, this standard protocol relies on different physical and chemical lysis steps, and is mainly adapted to molecular tools such as qPCR and molecular fingerprinting techniques. The recent development of massively parallel sequencing technologies which permit more representative and accurate taxonomical inventories, has instigated a reassessment of the biases associated with the DNA extraction procedure.

Therefore, the aim of this study was to assess the most appropriate soil DNA extraction procedure for studying soil diversity. In this context, three different procedures were tested (the ISO standard, an optimization of the ISO procedure (ISOm), and a custom procedure (GnS-GII)) on five soils of differing land use and physicochemical properties. The quality of the DNA extracted according to these different procedures was evaluated in terms of yield, microbial abundance and diversity. These analyses are being confirmed by a 454 pyrosequencing analysis.

Results clearly showed that DNA yield was higher using the ISOm and GnS-GII procedures compared to the ISO. Moreover, the abundance of bacteria, archaea and fungi were found to be lower with the ISO extraction method. Bacterial diversity patterns exhibited similar results, with a strong impact of soil pH on diversity profiles. Fungal community structure was strongly affected by the DNA extraction protocol, with soil type having only a secondary effect for the ISOm and GnS-GII methods. Archaeal diversity patterns were influenced both by soil physicochemical properties and the DNA extraction procedure.

These results indicate that the ISO standard is a suitable extraction procedure for studies focusing on soil bacteria. However, for larger scale studies which aim to examine bacteria, archaea and fungi the standardized procedure would not be appropriate. Indeed, the ISOm procedure, which is an improvement of the standard, produces results which are more reproducible and are more representative of soil microbial communities.

Key words: soil DNA, ISO, bacteria, fungi, archaea, microbial biomass, genetic structure