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**OLIVE-MILL WASTE AS POTTING SUBSTRATE FOR OLIVE TREE CULTIVATION: EFFECTS ON THE MICROBIOTA OF SOIL AND RHIZOSPHERE**

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**Abstract**

The growing attention to environmental sustainability, and particularly to the reduction of waste production and non-renewable resource consumption, has encouraged the recycling and valorization of agro-industrial by-products. The use of peat in potted plant cultivation has raised concerns because of the destruction of peat bogs. Replacing this substrate with other organic materials is thus environmentally and economically appealing. Mediterranean countries, where the production of olive oil is continuously rising, produce large amounts of olive-mill waste over a short time with important environmental consequences. The humid husk generated by two-phase oil extraction systems could be used, either directly or after composting, as peat surrogate. The aim of this work was to investigate the impact on the soil and rhizosphere microbiota of these alternative substrates.

Soil and rhizosphere samples were collected from potted olive trees where either peat or fresh and composted humid husk were used. Cultivation-independent methods, namely PCR-DGGE and qPCR analyses, were applied to study the microbial communities and the nitrifier bacteria populations. The composting process changed the structure of both bacterial and fungal communities present in the humid husk but didn't seem to affect the total number of bacteria. On the contrary, fungi were more abundant in the fresh humid husk. The bacterial and fungal DGGE fingerprints were very different both in the soil and the rhizosphere when the substrates based on olive-mill waste were used in place of peat. The fresh husk seemed to have a more profound effect than the composted one. Besides modifying the community structure, the use of this substrate also showed a positive effect on the total number of microorganisms. In particular, it strongly stimulated the growth of fungi both in soil and rhizosphere. Interestingly, qPCR analysis of *amoA* gene abundance, showed that the number of nitrifier bacteria was significantly reduced in the rhizosphere of the olive trees potted using the husk-based substrates, particularly the fresh one, suggesting a possible negative effect on nitrogen availability for plant growth.

Taken together, these data demonstrate that replacing peat with husk-based substrates modifies the soil and rhizosphere microbiota and suggest that fungi play a primary role in the degradation of the complex organic matter present in the fresh waste. The inhibition of nitrifier bacteria in the rhizosphere suggests that care should be taken when using the fresh humid husk for olive tree pot growth. In this respect, composting may represent a valuable strategy for producing peat surrogates.

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