

# Resident Microbial Dynamics during Natural Digestion of Olive Oil Mill Waste Waters in an Integrated Valorization System

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## Abstract

An integrated olive mill waste water (OMWW) processing plant established in Vissas region in Syros island, Greece, is described. It consists of a biological treatment unit for processing OMWWs separated after waste sedimentation and of a solid drying unit. The final products are liquid effluents that are used for irrigation and dried digested biomass, which is used for fertilization.

According to the process, the wastes from the oil mill, apart from the seeds, are initially transferred to a conical tank and 0.5% CaCO<sub>3</sub> is added to result in a pH rise up to 8.0, in order to facilitate sedimentation. At this stage, sedimentation up to 80-90% is achieved. The liquid effluents (OMWWs) from the tank are transferred to the biological treatment unit, while the remaining sludge is removed to the drying unit, where natural digestion is carried out.

The biological treatment unit is composed by 3 parts: (i) an aerobic tank, (ii) a sedimentation tank and (iii) a final tank for collection of “clean” wastes. The sludge from the sedimentation tank is carried to the drying tanks, while the liquid effluents (clean water) from tank can be used for irrigation.

The drying tanks are made of cement and at the bottom there is a layer of thick nylon, then a layer of thin gravel (10-15 cm thickness) and finally a layer of sand (10-15 cm thickness). Inside the gravel layer, perforated tubes are located for the collection of liquid effluents that are transferred to the biological treatment unit. The drying tanks are covered with nylon to cause significant increase of temperature, which can rise up to 70°C during the summer. The final product of olive mill solid waste (OMSW) is dried biomass, which after mixing with soil can be used for fertilization.

Microbial dynamics during natural digestion of the oil mill waste solids carried out in the drying tanks was investigated based on DGGE profiling of PCR-rDNA fragments. The PCR-DGGE protocol revealed that *Klebsiella oxytoca*, *Citrobacter koseri* and members of *Lactobacillus*, *Lactococcus* and *Enterococcus* or *Clostridium* genera and of *Enterobacteriaceae*, *Spirochaetales*, *Ruminococcaceae* or *Lachnospiraceae* family were the main bacterial populations detected, while the major yeast identified belonged to *Pichia* and *Candida* spp.

In conclusion, valorization of OMWWs is a challenging opportunity for sustainable and competitive development for the olive oil industry. In the present study, a processing plant for exploitation of OMWWs was described, resulting in zero residues. In addition, the DGGE profiling of resident microbiota during natural digestion of solid OMWWs was determined, showing a high species diversity. However, more research is still required in both fields in order (i) to apply composting or vermicomposting processes in OMSW as promising alternatives and (ii) to better understand the microbial associations and biological mechanisms during digestion of OMWWs. Application of sophisticated molecular technologies, such as next generation sequencing (NGS), is expected to provide additional information on the microbial interactions in such complicated ecosystems.