

Supplementary material of: **Assessing the Long-Term Efficiency of the MULESL System: A Sustainable Solution for Wastewater Treatment and Agricultural Water Reuse**

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The MULESL system was established by transforming an existing activated sludge basin at the Putignano WWTP in southern Italy (**Figure 1-A**), which has a daily treatment capacity of 3,600 m³. The WWTP's water line includes preliminary treatment (screening, equalisation, desanding), primary sedimentation (472 m³), and secondary treatment via a conventional activated sludge (CAS) process comprising denitrification (1270 m³), nitrification/oxidation (4570 m³), and secondary settling (924 m³). Tertiary treatment involves phosphorus removal using iron- and aluminium-based coagulants, followed by disinfection with sodium hypochlorite. The sludge line consists of anaerobic digestion and centrifugation for dewatering.

In the present study, the MULESL plant received influent sewage wastewater that had undergone only preliminary treatment (i.e., screening and desanding).

The MULESL system is designed to separate the biomass and liquid phases in a single treatment basin. The biomass compartment (180 m³), which was filled with highly porous plastic media (12 mm in diameter and 8 mm in height with 85% porosity), was physically divided from the liquid compartment by a vertical partition (**Figure 1-C**). This allowed overflow from the biomass zone to the liquid side. A recirculation pump circulated wastewater at a rate of 250 m³/h from the bottom of the liquid compartment to the plenum chamber of the biomass compartment. To maintain high levels of dissolved oxygen, air was continuously introduced into the liquid compartment, and pure oxygen was injected into the recirculation piping at a rate of 15 Nm³/h.

The MULESL system operates in sequential batch mode, consisting of four automated phases: feeding, recirculation, discharge, and idle. During the feeding phase, sewage was pumped at a rate of 120 m³/h into the plenum chamber of the biomass compartment. As it passed through the porous media, particulate matter was filtered out. The recirculation system is activated at the beginning of the cycle together with the feeding pump. When the liquid in the liquid phase compartment rose to a second threshold, the feeding pump stopped while recirculation continued.

During the discharge phase, the same pump used for the recirculation was used to extract treated effluent from the liquid compartment through a dedicated pipe. The subsequent idle phase allowed for maintenance operations, particularly cleaning the biomass compartment to restore lost porosity due to biomass accumulation. This was triggered when head losses at the bottom of the biomass zone reached 1.1 bar, with the sludge pump being used to extract excess sludge. This material was then removed, measured to calculate sludge production and retention time, and directed to the conventional WWTP sludge line.