



Isolation and morphological identification of microorganisms present in an activated sludge sample from a wastewater treatment plant

Maflah M.M.F.¹, Bandara S.M.D.C.², Nuwansi K.K.T.¹, Indika B.A.N.¹ and Ratnayake R.R.^{2*}

¹Department of Aquaculture and Seafood Technology, Faculty of Fisheries and Ocean Sciences,
Ocean University of Sri Lanka

²National Institute of Fundamental Studies (NIFS), Kandy, Sri Lanka

*Corresponding author: renuka.ra@nifs.ac.lk

Abstract

Activated sludge, a semi-solid material by product of wastewater treatment, consists of organic matter, microorganisms, and inorganic compounds. The microbial community within the sludge plays a crucial role in the treatment process. The study aimed to isolate and morphologically identify the microorganisms present in a sludge sample collected from the secondary clarifier tank of a Brewery wastewater treatment plant in Kaduwela, Sri Lanka. A sample of activated sludge was collected aseptically for microbial analysis, which included the isolation of bacteria, fungi, and cyanobacteria. Physiochemical parameters, such as pH, temperature, electrical conductivity, total dissolved solids, and salinity, were tested onsite. Microbial isolation was performed using the serial dilution method, followed by selective plating on Nutrient Agar (NA) for bacteria, Blue-Green (BG-11) agar for cyanobacteria, and Potato Dextrose Agar (PDA) for fungi. The spread plate technique was used to evenly distribute 100 µL of sludge samples. The plates were incubated at 37°C, with BG-11 plates exposed to a light intensity of 2000 lux. After colony formation, pure cultures were obtained using the streak plate method. Bacterial isolates were classified based on Gram staining, while fungal isolates were identified using Lactophenol Cotton Blue staining. Cyanobacteria were observed directly under a compound light microscope due to their inherent blue-green pigmentation. The results showed a diverse microbial community, including both gram-positive and gram-negative bacteria, which is typical in wastewater systems where microorganisms degrade organic and inorganic compounds. The fungal population mainly consisted of *Saccharomyces spp.*, a yeast species capable of fermenting sugars and contributing to the breakdown of organic matter in the sludge. However, no cyanobacteria were observed, likely due to the anaerobic and/or low-light conditions in the secondary clarifier tank, which are unfavorable for their survival. The findings highlight the importance of microbial diversity in wastewater treatment systems, where bacteria and fungi play a significant role in pollutant degradation. The absence of cyanobacteria aligns with the environmental conditions in the sludge sample, suggesting that their growth is limited by light availability and competitive interactions within the microbial community. This study provides valuable insights into the microbial community of sludge in a brewery wastewater treatment plant. Further studies are needed to molecularly identify these microorganisms and analyze their biochemical mechanisms in wastewater treatment.

Keywords: Microbial isolation, Sludge, *Saccharomyces spp.*, Brewery wastewater treatment, Gram staining