822/D

Isolation and identification of *Thiobacillus* species and cellulose degrading *Clostridium* species from Winogradsky column

P D S Weerasingha* and M M Gunawardane
Department of Microbiology, Faculty of Science, University of Kelaniya, Kelaniya

The Winogradsky column is a simple device for culturing large diversity of microorganisms and pond mud is a source to isolate a wide range of organisms. Stratification of microorganisms from mud in the Winogradsky column makes it easier for microbiologist to isolate functionally different bacteria. The aim of this study was to isolate and identify *Thiobacillus* and *Clostridium* species using the standard Winogradsky column. A stratified ecosystem was artificially constructed according to the basic structure of the Winogradsky column that was constructed by mixing $CaSO_4$, $CaCO_3$ and cellulose with mud and left under light for 4 weeks.

In the present study, *Thiobacillus versutus* was isolated from the mud in the upper part of the Winogradsky column using *Thiobacillu*s mineral salt selective medium and biochemically identified according to the methods given in Bergey's manual of systematic bacteriology; Volume 3. Since *Thiobacillus* species are involved in the biological sulfide oxidation, they can be used for oxidation of sulfur in soil and to increase the bioavailability of SO_4^{2-} in agricultural soil. They can also be applied to reduce the pH of alkali soils. One of the most significant applications of *Thiobacillus* species is for the bio-removal of sulfide (S^{2-}) ions which produce odorous gases such as H_2S in biogas systems and waste water treatment plants.

Clostridium acetobutylicum was isolated from the mud in the bottom of the Winogradsky column on *Clostridium* cellulose medium under anaerobic condition, and biochemically identified upto genus level with reference to the identification method described in the Cowen and Steel's manual of The identification of Medical Bacteria. Identification up to the species level was carried out using a key given in the Bergey's manual of systematic bacteriology; Volume 2. As *Clostridium acetobutylicum* produces cellulase enzyme it is used in the commercial production of cellulase. Another intense area of study of *Clostridium acetobutylicum* is H_2 production as an alternative energy source. They are commonly used in industrial production of solvents such as acetone, butanol and ethanol.

Keywords: *Clostridium acetobutylicum*, *Thiobacillusversutus*, Winogradsky column

sasankaweerasinghe89@gmail.com Tel: 0712303009