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Modification of Nutrient Agar Medium to Culture Un-culturable Bacterial Strains Living in Unsanitary Landfills

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The decomposition of municipal solid waste is mediated by native microorganisms and it is essential to estimate the participating microorganisms to accelerate the decomposition of waste material. However, 99% of bacterial species remain unculturable in the standard Nutrient Agar (NA) medium. This research was focused on the modification of the NA medium to mimic the composition of the natural substrate where they live. Conventional NA medium was modified by adding separately 40% (M1), 32% (M2), 24% (M3), 16% (M4), 8% (M5) (v/v) of soil extracts obtained from the dumpsite. The Conventional NA medium was the control. Random soil samples were collected from the garbage dump and bacterial species were isolated in each modified medium. The isolated bacterial cultures were air-dried under aseptic conditions and powdered. To characterize them, Fourier Transform Infrared (FTIR) spectra were recorded for all bacterial samples in the 500 - 4000 cm⁻¹ region at 4 cm⁻¹ resolution by scanning 120 times. The average of the absorbance of each FTIR spectrum was used as variables of the distance matrix of the cluster analysis (Minitab 19). A total of 103 bacterial strains were isolated and 9 bacteria samples showed a similarity level of more than 98%. The 20, 14, 16, 16, 18, and 19 bacteria species were isolated from the modified media M1, M2, M3, M4, M5, and the control, respectively. According to the cluster analysis, six clusters were obtained. Cluster 1 was the largest and it consisted of 45.63% of the total bacterial isolates from M1, M2, and the control. Cluster 2 consisted of 30.10% of total isolates from M4-M5. All the other isolates were clustered in clusters 3, 4, and 5. The separate clustering of the isolated bacteria in the modified media showed dissimilarity among them, thus indicating the potential of the soil extracts to modify NA to culture un-culturable bacterial strains living in unsanitary landfills.

Keywords: Unculturable bacteria, Soil extracts, FTIR

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