Chemical Characterization and Physical and Biological Activities of Rhamnolipids Produced by *Pseudomonas aeruginosa* BN10

Nelly Christova\(^a\), Boryana Tuleva\(^a\), Rashel Cohen\(^b\), Galya Ivanova\(^c\), Georgy Stoev\(^d\), Margarita Stoilova-Disheva\(^a\), and Ivanka Stoineva\(^e\)*

\(^a\) Institute of Microbiology, Bulgarian Academy of Sciences, 1113 Sofia, Bulgaria

\(^b\) Institute of Physical Chemistry, Bulgarian Academy of Sciences, 1113 Sofia, Bulgaria

\(^c\) Institute of Organic Chemistry with Centre of Phytochemistry, Bulgarian Academy of Sciences, 1113 Sofia, Bulgaria. Fax: +359 2 8700 225.

E-mail: ane@orgchm.bas.bg

\(^d\) Central Laboratory for Veterinary Control and Ecology, 1528 Sofia, Bulgaria

* Author for correspondence and reprint requests

**Z. Naturforsch.** 66c, 394–402 (2011); received December 14, 2010/March 23, 2011

*Pseudomonas aeruginosa* BN10 isolated from hydrocarbon-polluted soil was found to produce rhamnolipids when cultivated on 2% glycerol, glucose, \(n\)-hexadecane, and \(n\)-alkanes. The rhamnolipids were partially purified on silica gel columns and their chemical structures elucidated by combination of one- and two-dimensional \(^1\)H and \(^13\)C NMR techniques and ESI-MS analysis. Eight structural rhamnolipid homologues were identified: \(\text{Rha-C}_{10}\text{-C}_{8}\), \(\text{Rha-C}_{10}\text{-C}_{10}\), \(\text{Rha-C}_{10}\text{-C}_{12:1}\), \(\text{Rha-C}_{10}\text{-C}_{12}\), \(\text{Rha}_{2}\text{-C}_{10}\text{-C}_{8}\), \(\text{Rha}_{2}\text{-C}_{10}\text{-C}_{10}\), \(\text{Rha}_{2}\text{-C}_{10}\text{-C}_{12:1}\), and \(\text{Rha}_{2}\text{-C}_{10}\text{-C}_{12}\). The chemical composition of the rhamnolipid mixtures produced on different carbon sources did not vary with the type of carbon source used. The rhamnolipid mixture produced by *Pseudomonas aeruginosa* BN10 on glycerol reduced the surface tension of pure water from 72 to 29 mN m\(^{-1}\) at a critical micellar concentration of 40 mg l\(^{-1}\), and the interfacial tension was 0.9 mN m\(^{-1}\). The new surfactant product formed stable emulsions with hydrocarbons and showed high antimicrobial activity against Gram-positive bacteria. The present study shows that the new strain *Pseudomonas aeruginosa* BN10 demonstrates enhanced production of the di-rhamnolipid \(\text{Rha}_{2}\text{-C}_{10}\text{-C}_{10}\) on all carbon sources used. Due to its excellent surface and good antimicrobial activities the rhamnolipid homologue mixture from *Pseudomonas aeruginosa* BN10 can be exploited for use in bioremediation, petroleum and pharmaceutical industries.

**Key words:** *Pseudomonas*, Rhamnolipids, Emulsification, Antimicrobial Activity