Theme: EPD: Challenges and best practices: Running a research program primarily with undergraduate students

Abstract 1857

"BioSolve: Exploring Biguanide Biodegradation Pathways through Collaborative Undergraduate Research Experiences"

Betsy M. Martinez-Vaz, *Hamline University*

Larry Wackett and Martin St Maurice

Keywords: Undergraduate research program, biodegradation CURE, microbial biochemistry, pharmaceuticals, wastewater

Biguanides are organonitrogen compounds prevalent in wastewater due to their widespread use in agricultural additives, pharmaceuticals, and personal care products. Metformin (1,2-N-dimethylbiguanide) is the most prescribed type II diabetes drug in the world and has been detected at significant levels in coastal waters and rivers worldwide. While recent studies reported the isolation of metformin-degrading bacteria and uncovered its catabolic pathway, the biodegradation of other biguanides and the connections between their metabolic pathways remain unexplored. This study describes establishing a collaborative undergraduate research program amongst three universities that focused on investigating the biodegradation of biguanides using microbes from soils and wastewater. As part of a course undergraduate research experience (CURE) and a summer research program, students prepared microbial enrichment cultures using soils from diverse locations and activated sludge samples from wastewater treatment facilities. The samples were grown on a minimal medium with biguanides including metformin, 1-N-methylbiguanide, and cyanoguanidine as the sole nitrogen sources for growth. Biodegradation and microbial growth were monitored by HPLC analyses, colorimetric assays, and optical density, respectively. Microbial consortia capable of degrading multiple biguanides were obtained and further characterized by genome sequencing and bioinformatics analyses. Bacterial strains that metabolized biguanides independently of microbial consortia were isolated and examined for the presence of candidate genes and enzymes potentially involved in biguanide metabolism. Guanylurea hydrolase, an enzyme commonly found in several biguanide catabolic pathways, was selected for further mutagenesis and biochemical analyses. The results of these studies suggest that the microbial catabolism of biguanide compounds shares common guanylurea and guanidine intermediates as well as the enzymes involved in their metabolism. The CURE and summer research activities were implemented over two years in introductory as well as intermediate courses at the participating universities. Preliminary assessment results show students' learning gains in collaboration skills, data interpretation, and the application of microbial enzymes to enhance wastewater treatment processes.

This work was funded by Chemistry of Life Processes (CLP) Program of the National Science Foundation, grant no. 2203751 to Betsy Martinez-Vaz, Larry Wackett and Martin St. Maurice.

105937, https://doi.org/10.1016/j.jbc.2024.105937