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BACTERIAL EVOLUTION FOR BIODEGRADATION: UNDERSTANDING AND USING BIODEGRADATION METABOLISM

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Bacteria in soil and water have evolved naturally to biodegrade *s*-triazine ring compounds like cyanuric acid and melamine. Melamine was recently linked to the hospitalization of 150,000 children in China from their drinking contaminated milk. Cyanuric acid is a natural product and is formed during the decomposition of dichloroisocyanuric acid, a chemical widely used to chlorinate swimming pools. In my research, I have studied how bacteria biodegrade cyanuric acid. Cyanuric acid is biodegraded to carbon dioxide and ammonia by a series of three enzymes: cyanuric acid hydrolase, biuret hydrolase and allophanate hydrolase. My research has shown that cyanuric acid hydrolase cleaves a single bond in the cyclic amide cyanuric acid to yield carboxybiuret. Carboxybiuret undergoes decarboxylation to yield biuret. Prior to the present work, no one had been able to stabilize a biuret hydrolase sufficiently to study the enzyme in detail. We searched through DNA databases to find bacterial genomes that might encode biuret hydrolases. Several candidate biuret hydrolase genes were cloned and expressed and one was stable and further studied. The biuret hydrolase was found to be very selective for biuret and produce allophanic acid as a product. It was shown not to be reactive with carboxybiuret, the direct product of the cyanuric acid hydrolase reaction. Potential uses for these enzymes will be discussed.

BACTERIAL EVOLUTION FOR BIODEGRADATION: NEW ENZYMES TO HANDLE INDUSTRIAL *S*-TRIAZINES

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Evolutionary biologists typically speak of evolution as it occurs over the course of millions of years. But microorganisms evolve much more rapidly than this to develop metabolism to "eat" new industrial chemicals. When the chemicals consumed are environmental pollutants, the "eating" by bacteria is known as biodegradation. We have extensively studied the biodegradation *s*-triazine compounds, a major class of industrial chemicals. Common industrial *s*-trazine compounds are melamine and atrazine. Atrazine is a herbicide that is used extensively in corn fields. Atrazine has been used by farmers for fifty years and it has become more biodegradable over time. Our studies have revealed how bacteria biodegrade atrazine and how that biodegradation has evolved. The first step in the biodegradation is the enzyme-catalyzed removal of the chlorine substituent from atrazine. Two other enzymes work on the product of the first enzyme to produce cyanuric acid. Cyanuric acid is further biodegraded to carbon dioxide and ammonia. Ammonia is adsorbed by the bacteria and used to make their proteins and DNA. The capability to biodegrade atrazine has spread amongst bacteria by horizontal gene transfer (HGT). HGT is a process in which plasmids carrying genes for a particular function move through populations. In this way, the huge populations on earth continually evolve to biodegrade new chemicals that humans synthesize and put into the environment. This knowledge has been used to engineer biotreatment systems for degrading environmental pollutants.