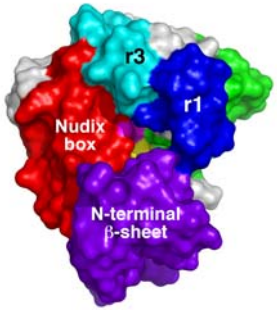
Common Enzyme has Unusual Appetite

Novel substrate identified for protein from radiation resistant bacterium

Deinococcus radiodurans can survive thousands of times more radiation exposure than a human. The biological mechanism for its radiation resistance may be related to an uncommonly large suite of housekeeping proteins called Nudix hydrolyases. A research team from Brookhaven National Laboratory, the University of Toronto, and Pacific Northwest National Laboratory have used EMSL's state-of-the-art nuclear magnetic resonance spectroscopy capabilities to help determine the crystal structure for one of these *D. radiodurans* Nudix hydrolases, DR_0079, and uncovered its preference for a novel substrate.



DR_0079 (205F) surface structure highlighting the Nudix box (red) and regions of the protein associated with substrate binding.

Nudix hydrolyases are ubiquitous, existing in the genomes of organisms as diverse as viruses and humans. They are identified by a highly conserved, 23-residue consensus sequence, called the Nudix box, which forms part of the substrate-binding and catalytic site. The rest of a Nudix hydrolase's sequence determines the overall protein structure, which in turn influences substrate specificity. Substrates for Nudix hydrolases are typically nucleotide-based (related to the building blocks of DNA and RNA). The classic Nudix protein, MutT, protects cells by converting dangerous promutagenic nucleoside triphosphates into safe nucleoside monophosphates – the former can be incorporated into DNA and lead to cancer, and the latter cannot. DR_0079 is unusual because it converts nucleoside *diphosphates* instead of nucleoside triphosphates into nucleoside monophosphates.

The team used X-ray diffraction data collected at the National Synchrotron Light Source to study the structure of DR_0079. To relate structure to function, the preferred substrate of the enzyme was verified and the molecular mechanism probed using ^{31}P NMR spectroscopy at EMSL. This technique made it possible to study the protein's activity in real time and with molecular detail.

Scientific impact: The new function documented for DR_0079 expands the range of possible Nudix substrates. Novel findings from real-time, molecular-level structure-function studies further EMSL's goals to predict biological functions from molecular and chemical data and to advance from static to dynamic studies in native environments.

Societal impact: Understanding the molecular basis for the radiation resistant properties of *D. radiodurans* may lead to novel bioremediation methods and to strategies that protect humans from the deleterious effects of ionizing radiation.

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