

Microbial Valorization of Lignin: Using *Novosphingobium aromaticivorans* to Break the Bonds in Lignin and Convert Lignin Deconstruction Products into Value-added Chemicals

Wayne S. Kontur^{1,2*} (wskontur@wisc.edu), J. Miguel Perez^{1,2}, **Daniel R. Noguera**,^{1,2} and **Timothy J. Donohue**^{1,2}

¹University of Wisconsin-Madison; ²U.S. DOE Great Lakes Bioenergy Research Center, Madison, WI

Project Goal:

To develop the bacterium *Novosphingobium aromaticivorans* DSM 12444 into a model system for understanding how bacteria break the bonds between subunits in lignin and metabolize aromatic lignin deconstruction products, and into a platform for converting lignin deconstruction products into value-added chemicals

Lignin is a heterogeneous polymer of aromatic subunits that is a major component of lignocellulosic plant biomass (comprising up to ~25% of its dry weight). In biomass processing facilities that primarily focus on converting the sugar-rich biomass fractions (cellulose and hemicellulose) into value-added commodities, lignin is currently typically burned for heat and power generation. While this utilizes the chemical energetic value of lignin, it wastes lignin's potential as a renewable resource for value-added chemicals that are currently derived from petroleum (such as fine chemicals, food additives, plastic precursors, pharmaceuticals, etc.). To help improve the overall economic potential of generating fuels and other renewable commodities from lignocellulosic plant biomass, we are therefore investigating ways to generate value-added chemicals from lignin using microbes.

The bacterium *Novosphingobium aromaticivorans* DSM 12444 is a very promising model system for valorizing lignin, due to the following characteristics:

- It efficiently metabolizes all lignin-derived aromatic compounds that it has been tested for growth on
- It can naturally produce some potential value-added chemicals from lignin, such as the flavoring agent vanillin and the epoxy/plastic precursor 2-pyrone-4,6-dicarboxylic acid (PDC)
- It can metabolize glucose, which raises the possibility that a strain could be engineered to derive growth and energy from glucose, while converting all or most of the aromatic component of plant biomass into a desired commodity

We have been working on characterizing both the microbe's ability to break the bonds between aromatic subunits in the lignin polymer and its ability to metabolize the three major classes of mono-aromatic compounds that result from lignin's deconstruction (the syringyl (S), guaiacyl (G), and p-hydroxyphenyl (H) compounds). As part of this work, we have identified and characterized enzymes involved in both processes, such as those responsible for breaking the β -

aryl ether bond, the most common inter-subunit bond in natural lignin, and those responsible for opening the aromatic ring of the mono-aromatic constituents.

Our understanding of the metabolic pathways of *N. aromaticivorans* has enabled us to develop a strain which can convert S, G, and H compounds into PDC at nearly the maximum theoretical yields. In addition, we are using what we have learned from PDC production to investigate developing strains of the microbe that can produce additional value-added compounds from lignin.

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