

# *P. putida* secretes outer membrane vesicles for extracellular aromatic breakdown

## Background

- Natural microbial conversion processes funnel aromatic compound mixtures into single products and thus have emerged as a means to overcome heterogeneity towards valorization of the plant polymer lignin.
- However, the spatiotemporal mechanisms for lignin catabolism in bacteria – including the chassis microbe *Pseudomonas putida* KT2440 – remain unclear and thus hinder the industrialization of these processes.

## Conclusions

- P. putida* selectively and dynamically secretes proteins to the extracellular space simultaneous with a reduction in  $\beta$ -O-4 bonds in the lignin substrate.
- Many of these proteins are encapsulated into outer membrane vesicles (OMVs, **Fig. A**), including those involved in the  $\beta$ -ketoacid ( $\beta$ KA) pathway for aromatic catabolism, which are then secreted to the extracellular milieu.
- OMVs from lignin-rich cultivations convert  $\beta$ KA pathway intermediates *in vitro* and *in vivo*, demonstrating that enzymes within the OMVs are active.

## Significance

- We propose a mechanism for nutrient acquisition from aromatic compounds by soil bacteria in the extracellular environment (**Fig. B**) which holds promise for improving microbial lignin conversion.

**Figure (A)** A *P. putida* cell imaged by scanning electron microscopy after 72 h of cultivation in lignin-rich medium where the cell surface is covered with “blebbing” outer membrane vesicles (two blebbing events are highlighted with a white arrowhead). **(B)** Model for OMV-mediated nutrient acquisition and/or catabolism of toxic substrates in *P. putida*.

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