SUPPORTING INFORMATION

The lid domain of the MCP hydrolase DxnB2 contributes to an increased specificity for recalcitrant PCB metabolites.

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HPLC analysis of DxnB2-mediated hydrolysis of 3-Cl HOPDAs. Reactions were analyzed using a Waters 2695 HPLC system (Waters Corp., Milford, MA) equipped with a Hewlett-Packard ODS Hypersil C_{18} column (5 μm, 125 mm × 4 mm) operating at a flow rate of 1 mL/min and equilibrated at 90% solvent A (0.5% H_3PO_4 in H_2O) and 10 % solvent B (methanol). Reactions of 2 mM DxnB2 with 100 μM 3-Cl or 50 μM 3,9,11-triCl HOPDA were carried out in potassium phosphate (I = 0.1 M), pH7.5, quenched with H_3PO_4 (0.5% v/v) and passed through a 0.45 μm filter prior to injection. Samples were eluted using two sequential gradients: (i) 10 to 30% B from 0 to 20 min and (ii) 30 to 60% B from 20 to 80 minutes. Retention times were 4.8 min for 2-hydroxy-3-chloro-2,4-pentadienoic acid (λ_{max} 244 nm), 14.8 min for benzoic acid (λ_{max} 229 nm), and 51.3 min for 3,5-dichlorobenzoic acid (λ_{max} 234 nm).

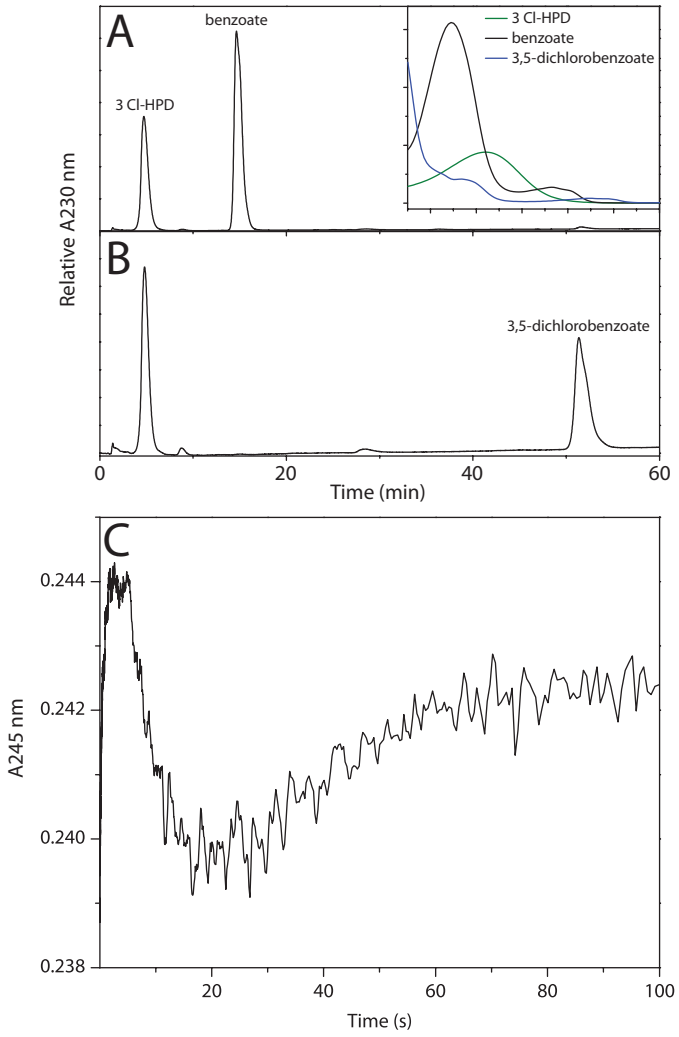

Figure S1. HPLC chromatograms of DxnB2-mediated hydrolysis of 3-Cl HOPDA (A) and 3,9,11-triCl HOPDA (B). The electronic absorbance spectrum of each distinct product is inset in (A). (C) A representative stopped-flow experiment monitoring turnover of 3-Cl HOPDA at 245 nm. The multiphasic nature and poor signal-to-noise were not interpreted since they may be partly attributed to several product species.

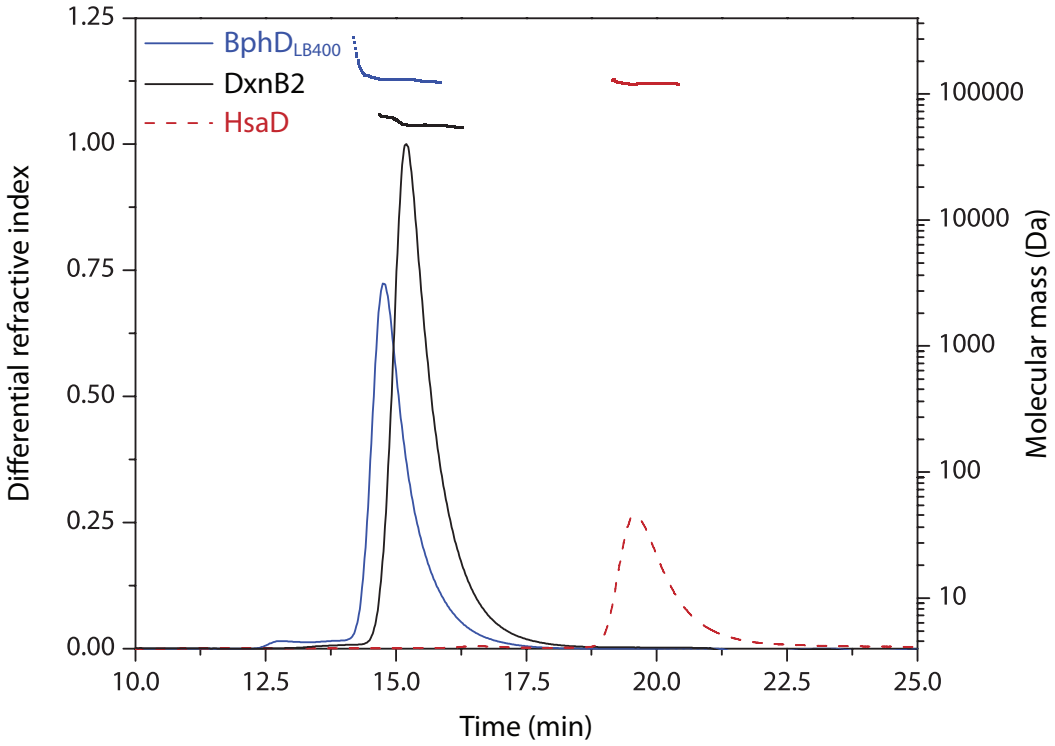

Figure S2. Superposition of chromatograms from SEC-MALS analysis of $BphD_{LB400}$ (blue), DxnB2 (black), and HsaD (red). The data used for molecular mass determination has been overlaid on the chromatogram.

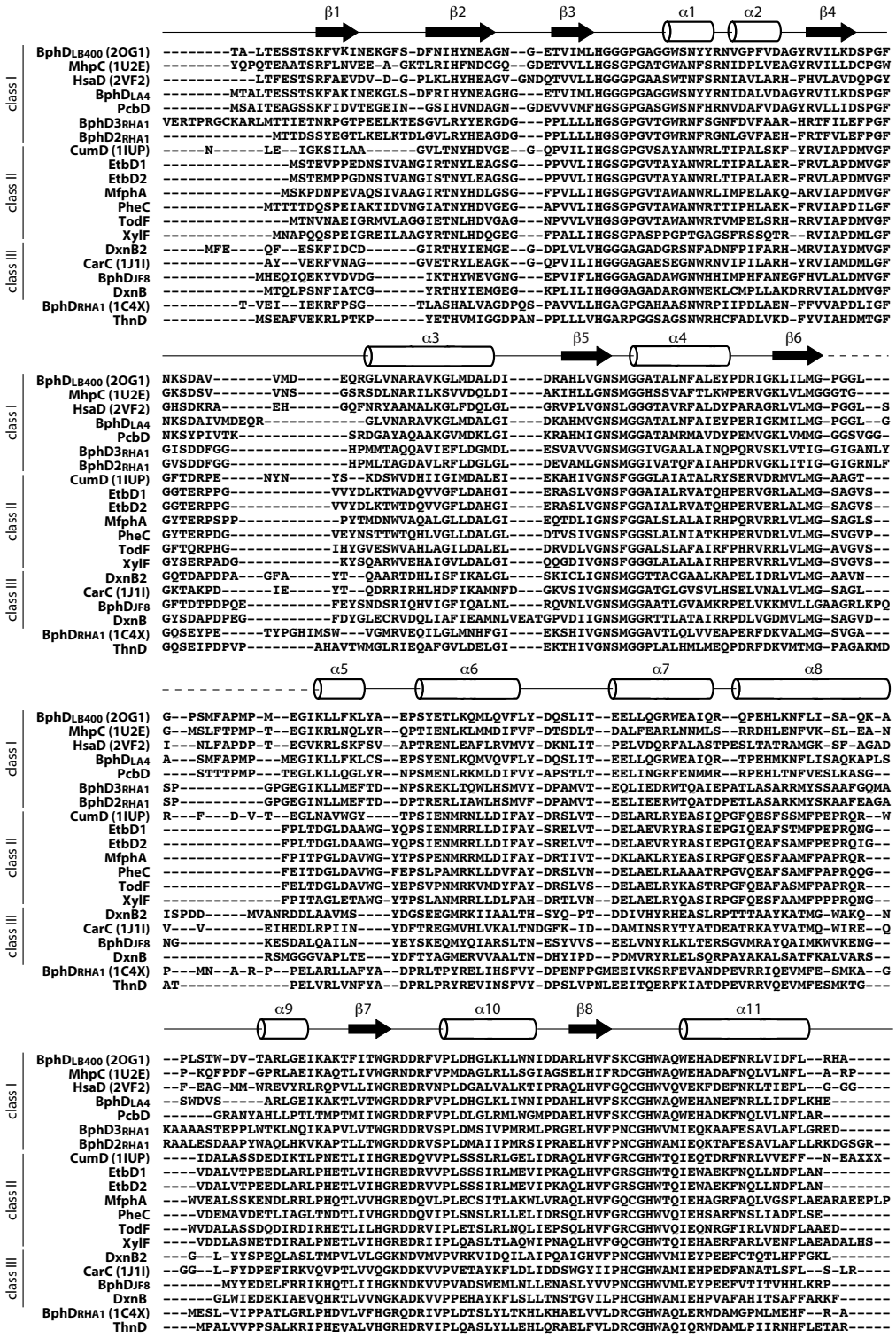

Figure S3. Sequence alignment resulting from the structural superposition of seven MCP hydrolases. The NC-loop is indicated by a dashed line and the secondary structural elements are numbered. The helix labeled 11 might also be considered a pair of shorter helices joined by a turn. For PDB IDs and biological source refer to Materials and Methods.