## Journal of Pesticide Science

Supplementary materials DOI:10.1584/jpestics.D17-089

## Identification of the novel *hcbB* operon catalyzing the dechlorination of pentachlorophenol in the Gram-positive bacterium *Nocardioides* sp. strain PD653.

Koji ITO, Kazuhiro TAKAGI,<sup>\*</sup> Yoshitaka MATSUSHIMA, Akio IWASAKI, Naoto TANAKA, Yu KANESAKI, Fabrice MARTIN-LAURENT and Shizunobu IGIMI

> Pesticide Science Society of Japan http:pssj2.jp/eng/

## Supplemental materials

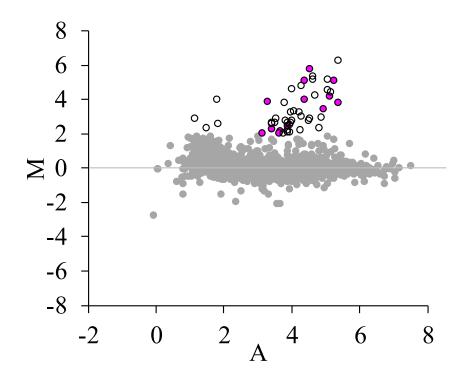


Fig. S1 Differential expression analysis of strain PD653 cells 8 h after exposed to HCB versus untreated. The  $log_2$  of the ratio of fold changes of each ORF (M) is plotted against the average abundance of that ORF in (A). For each plot, M and A values were obtained from triplicate samples. Open circles indicate the genes upregulated ( $log_2 > 2$ ) in the presence of HCB, and the gene cluster containing open reading frames (ORFs) responsible for the metabolism of PCP are plotted by magenta circles.

Locus tag	Fold	Predicted function	Closest related protein	Identity (%)	Accession no.	Organisms
-	induction		*			
D653_1060	8	Hypothetical protein	Hypothetical protein CF8_0913	52	EON25013.1	Nocardioides sp. CF8
PD653_1061	5	Hypothetical protein	NAD-dependent malic enzyme	77	OJH68642.1	Streptomyces viridifaciens
PD653_1062	10	SAF domain-containing protein	Pyridine nucleotide-disulfide oxidoreductase	77	WP_018155833.1	Demetria terragena
PD653_1063	7	Hypothetical protein	Hypothetical protein	66	WP_036490206.1	Nocardioides sp. CF8
PD653_1064	7	Type II secretion system protein E	Type II secretion system protein E	69	EON25017.1	Nocardioides sp. CF8
PD653_1065	6	Integral membrane protein	Integral membrane protein	68	EON25018.1	Nocardioides sp. CF8
PD653_1066	6	Type II secretion system protein	Type II secretion system protein	73	EON25019.1	Nocardioides sp. CF8
PD653_1067	1	Hypothetical protein	Hypothetical protein ASE01_16845	50	KRB75034.1	Nocardioides sp. Root190
PD653_1068	7	TadE family protein	TadE family protein	63	EON25020.1	Nocardioides sp. CF8
PD653_1069	3	Predicted transcriptional regulator	Hypothetical protein GA0115240_11986	54	SCD69461.1	Streptomyces sp. DvalAA-14
PD653_1070	1	Flp pilus assembly protein TadG	TadE family protein	65	EON25021.1	Nocardioides sp. CF8
PD653_1071	6	Superoxide dismutase	Hypothetical protein CF8_0923	69	EON25022.1	Nocardioides sp. CF8
PD653_1072	6	Peptidoglycan-binding LysM	Peptidoglycan-binding LysM	68	EON25023.1	Nocardioides sp. CF8
PD653_1108	14	GntR family transcriptional regulator	Transcriptional regulator, GntR family	78	ABL83953.1	Nocardioides sp. JS614
PD653_1109	16	Flavin reductase domain protein FMN-binding protein	Putative oxidase	61	CND52477.1	Mycobacterium tuberculosis
PD653 1110	4	HTH-type transcriptional regulator MalT	HTH-type transcriptional regulator MalT	74	KMO66989.1	Mycobacterium chlorophenolicum
PD653 1111	1	ATPase AAA				
PD653 1112	55	Pyridoxine 5'-phosphate oxidase	Putative pyridoxine/pyridoxamine 5'-phosphate oxidase	65	KM083753.1	Mycobacterium chlorophenolicum
PD653 1113	34	Hypothetical protein	Xylose isomerase-like TIM barrel	50	KM083752.1	Mycobacterium chlorophenolicum
PD653 1114	11	4-hydroxyphenylacetate 3-hydroxylase	FADH(2)-dependent monooxygenase TftD	84	KMO66988.1	Mycobacterium chlorophenolicum
PD653 1115	19	Hypothetical protein	Cupin domain protein	74	KMO66987.1	Mycobacterium chlorophenolicum
PD653 1116	2	Putative uncharacterized protein npdX	SnoaL-like domain protein	58	KMO66986.1	Mycobacterium chlorophenolicum
PD653_1117	4	XRE family transcriptional regulator with cupin sensor	GTP-binding protein LepA	81	KQW47527.1	Nocardioides sp. Root1257
PD653 1118	1	Transcriptional regulator	XRE family transcriptional regulator with cupin sensor	71	CND52544.1	Mycobacterium tuberculosis
PD653 1119	5	Transcriptional regulator, TetR family	TetR family transcriptional regulator	70	KQW53742.1	Nocardioides sp. Root1257
PD653 1120	3	NAD(P) transhydrogenase subunit alpha	NAD(P) transhydrogenase subunit alpha	70	KQW 55742.1 KQV74634.1	Aeromicrobium sp. Root344
PD653_1120 PD653_1121	6	NAD(P) transhydrogenase beta subunit	NAD(P) transhydrogenase subunit beta	73	SFD02090.1	Nocardioides terrae
	4			34		
PD653_1122	4	Serine/threonine protein kinase	hypothetical protein ASD11_15130	54	KQY55832.1	Aeromicrobium sp. Root495
PD653_1123	35	Dehydrogenases with different specificities (related to short-chain alcohol dehydrogenases)	Gluconate 5-dehydrogenase	78	KMO83627.1	Mycobacterium chlorophenolicum
PD653 1124	15	Mannose-6-phosphate isomerase				
PD653 1125	5	Hypothetical protein	MULTISPECIES: LysR family transcriptional regulator	58	WP 085870507.1	Nocardioides
PD653 1131	4	Peptidase M24	, , , , , , , , , , , , , , , , , , ,			
PD653 1132	2	Uncharacterized protein	Glyoxalase-like domain protein	84	KMO83754.1	Mycobacterium chlorophenolicum
PD653 1133	4	Enoyl-CoA hydratase (Fragment)	Putative enoyl-CoA hydratase echA8	69	KMO83648.1	Mycobacterium chlorophenolicum
PD653 1134	5	Mitomycin radical oxidase	Mitomycin radical oxidase	62	KMO83755.1	Mycobacterium chlorophenolicum
PD653 1135	36	FAD synthetase	Hypothetical protein	55	WP 082168625.1	Mycobacterium chlorophenolicum
PD653 1136	77	Uncharaterized protein	3-oxoacyl-[acyl-carrier-protein] reductase FabG	77	KMO66993.1	Mycobacterium chlorophenolicum
PD653 1137	36	Hypothetical protein		.,		, cooucier ium enter opnenoneum
PD653_1137	23	Phosphogluconate dehydrogenase (Decarboxylating), NAD binding domain protein	2-hydroxy-3-oxopropionate reductase	73	KMO66992.1	Mycobacterium chlorophenolicum
PD653_1139	22	Alkylhydroperoxidase AhpD family core domain	Carboxymuconolactone decarboxylase family protein	71	KMO66991.1	Mycobacterium chlorophenolicum
PD653 1140	8	Putative regulatory protein	HTH-type transcriptional regulator MalT	46	KMO66963.1	Mycobacterium chlorophenolicum
2D653_1140	1	Esterase/lipase	Carboxylesterase NlhH	74	KMO67023.1	Mycobacterium chlorophenolicum
PD653_1141 PD653_1142	1	ClcR	MULTISPECIES: LysR family transcriptional regulator	99	WP 085870507.1	Nocardioides
PD653_1142 PD653_1143	1	UspA domain-containing protein	Nucleotide-binding universal stress protein, UspA family	56	SHG98930.1	Jatrophihabitans endophyticus

PD653_1144	3	Glutathion S-transferase	Glutathione S-transferase	85	KQW47837.1	Nocardioides sp. Root1257
PD653_1145	10	Oxidoreductase	Putative N5,N10-methylenetetrahydromethanopterin reductase	74	EGD43879.1	Nocardioidaceae bacterium Broad-1
PD653_1156	4	Dihydrolipoamide dehydrogenase	Dihydrolipoamide dehydrogenase	69	CND52706.1	Mycobacterium tuberculosis
PD653_1157	5	NAD-dependent aldehyde dehydrogenase	Succinate semialdehyde dehydrogenase	75	CND52739.1	Mycobacterium tuberculosis
PD653_1158	8	Pyruvate dehydrogenase	pyruvate/2-oxoglutarate dehydrogenase complex%2C dehydrogenase component beta subunit	82	CND52779.1	Mycobacterium tuberculosis
PD653_1159	8	Pyruvate dehydrogenase complex dihydrolipoamide acetyltransferase	Dihydrolipoamide acetyltransferase component of pyruvate dehydrogenase complex	54	GAY07691.1	Pseudonocardia sp. N23
PD653_1160	6	Uncharacterized protein				
PD653_1161	4	NAD-dependent malic enzyme	Malate dehydrogenase	77	OEV38320.1	Kitasatospora aureofaciens

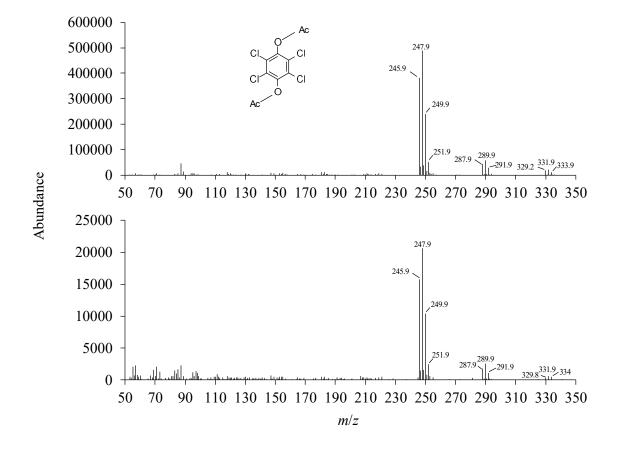


Fig. S2 GC-MS spectrum of a standard sample of acetylated TeCH (Top) and an acetylated metabolite (bottom) in the degradation of PCP.

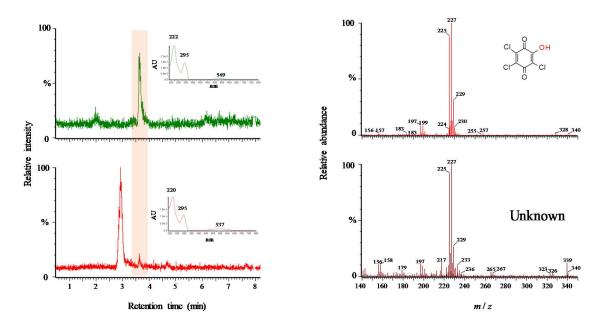


Fig. S3 UPLC-MS chromatogram (left) with UV-visible spectrum and the MS spectrum (right) for synthesized TCHQ (top), and those of the sample solution at 1 h (bottom) are shown.