

Supplementary Materials

Identification of reductive dehalogenases that mediate complete debromination of penta- and tetra-brominated diphenyl ethers in

Dehalococcoides

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Supplementary Figures

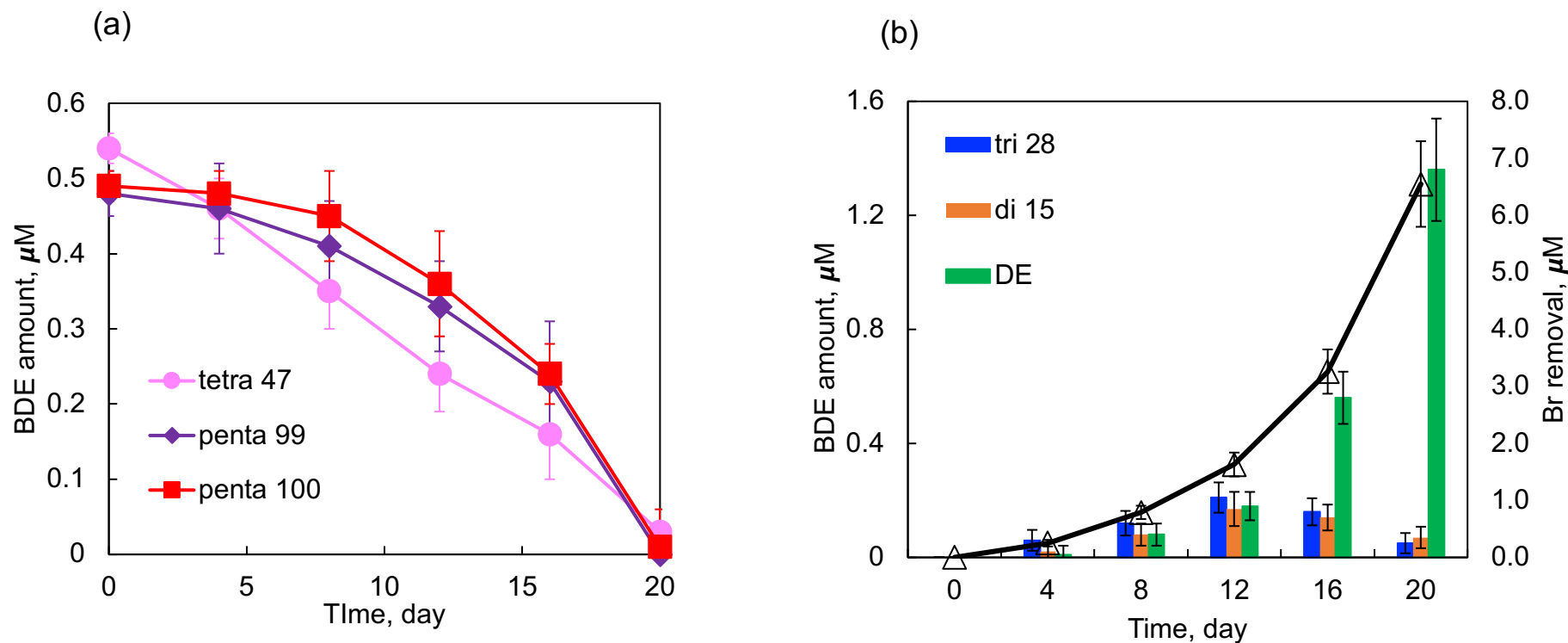


Figure S1. Sediment-free culture TZ50 debrominated a) parent and b) daughter compounds from the penta- and tetra-BDE mixture to produce diphenyl ether. DE, diphenyl ether. Bromine removal was estimated from measured amounts of PBDE congeners.

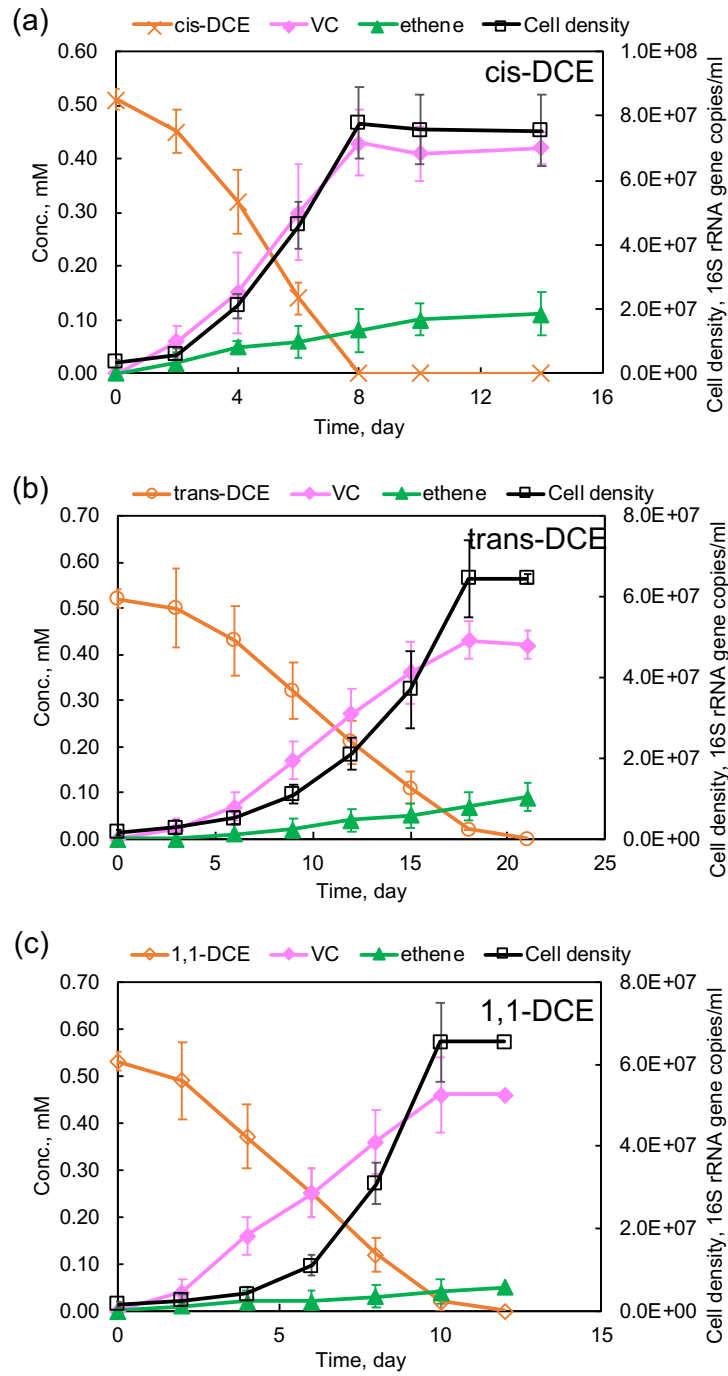


Figure S2. Dechlorination of a) *cis*-DCE, b) *trans*-DCE, and c) 1,1-DCE by the TZ50 enrichment culture. VC, vinyl chloride; DCE, dichloroethene.

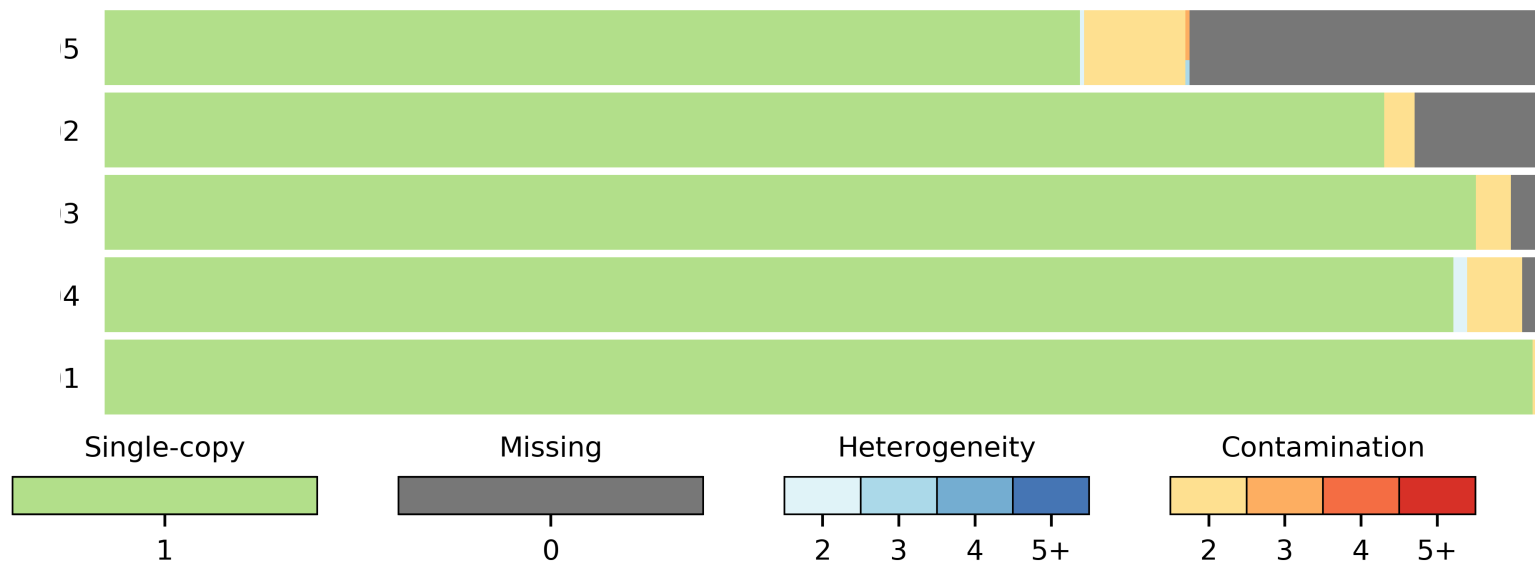
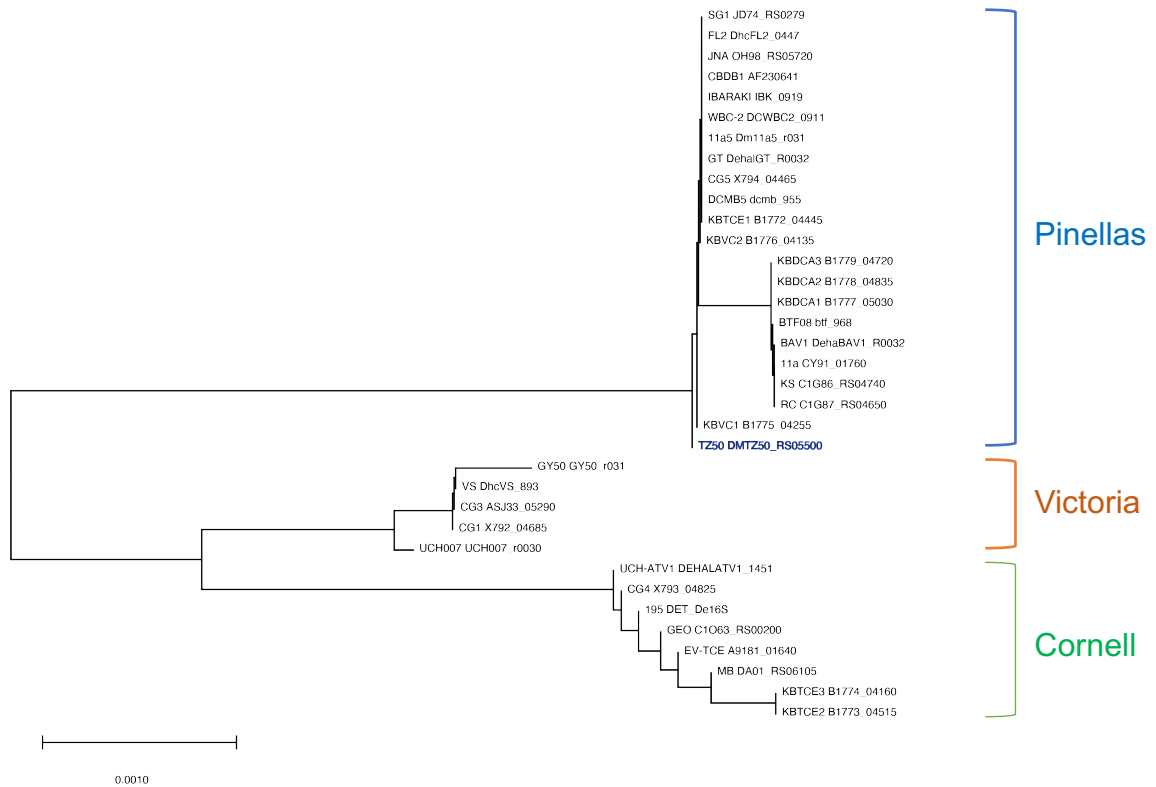


Figure S3. CheckM evaluation of binned metagenomic reads derived from the TZ50 enrichment culture. Kraken2 identified reads comprising bin 1 were as *Dehalococcoides*.

(a)



(b)

	TZ50	11a5	BTF08	CG5	WBC-2	DCMB5	BAV1	Ibaraki	KBDCA1	KBTCE1	CG1	GY50	VS	195	CG4
Pinellas	TZ50	*													
	11a5	99.59	*												
	BTF08	99.04	99.06	*											
	CG5	99.76	99.75	99.06	*										
	WBC-2	99.56	99.57	98.82	99.57	*									
	DCMB5	99.18	99.24	99.07	99.25	99.07	*								
	BAV1	98.96	98.98	99.12	99.07	98.83	99	*							
	Ibaraki	99.24	99.35	99.15	99.34	98.99	99.65	98.98	*						
	KBDCA1	99.25	99.22	99.23	99.3	98.83	99.34	99	99.26	*					
Victoria	KBTCE1	99.11	99.13	99.02	99.18	99.04	99.32	98.99	99.39	99.37	*				
	CG1	87.19	87.47	87.25	87.2	87.51	87.22	87.18	87.25	87.14	87.21	*			
	GY50	87.28	87.42	87.21	87.12	87.44	87.22	87.19	87.18	87.14	87.09	99.25	*		
	VS	87.26	87.37	87.45	87.08	87.5	87.12	87.16	87.24	87.11	87.27	97.66	97.87	*	
Cornell	195	86.98	87.13	87.24	87.11	87.11	86.98	86.98	86.9	87.14	86.95	90.44	90.44	90.37	*
	CG4	86.7	86.76	86.78	86.87	86.87	86.69	86.73	86.72	86.69	86.68	90.64	90.63	90.55	98.1

Figure S4. Phylogeny of the *Dehalococcoides* population in the TZ50 enrichment culture was inferred using a) a neighbor-joining phylogenetic tree of 16S rRNA gene sequences and b) ANIm genome comparison.

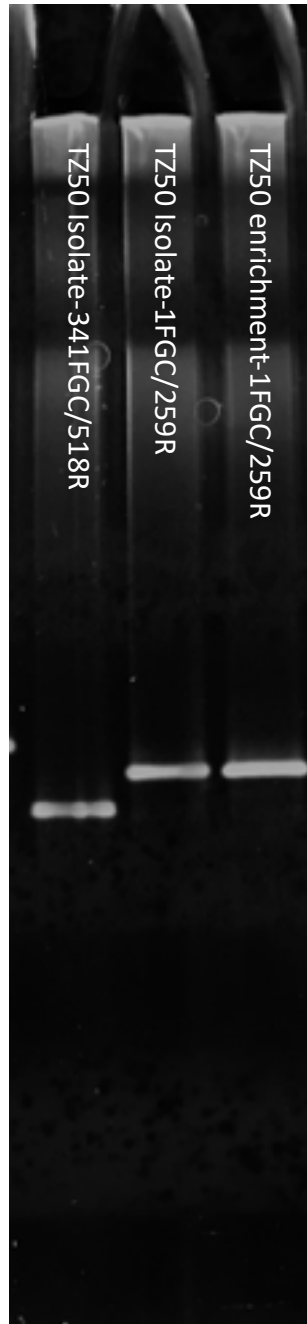


Figure S5. DGGE analysis of TZ50 enrichment culture and isolate. Lane 1: Strain TZ50 DNA amplified with universal bacterial primer pair 341FGC/518R; Lane 2: Strain TZ50 DNA amplified with *Dehalococcoides* specific primer pair 1FGC/259R; Lane 3: TZ50 enrichment culture DNA amplified with *Dehalococcoides* specific primer pair 1FGC/259R.

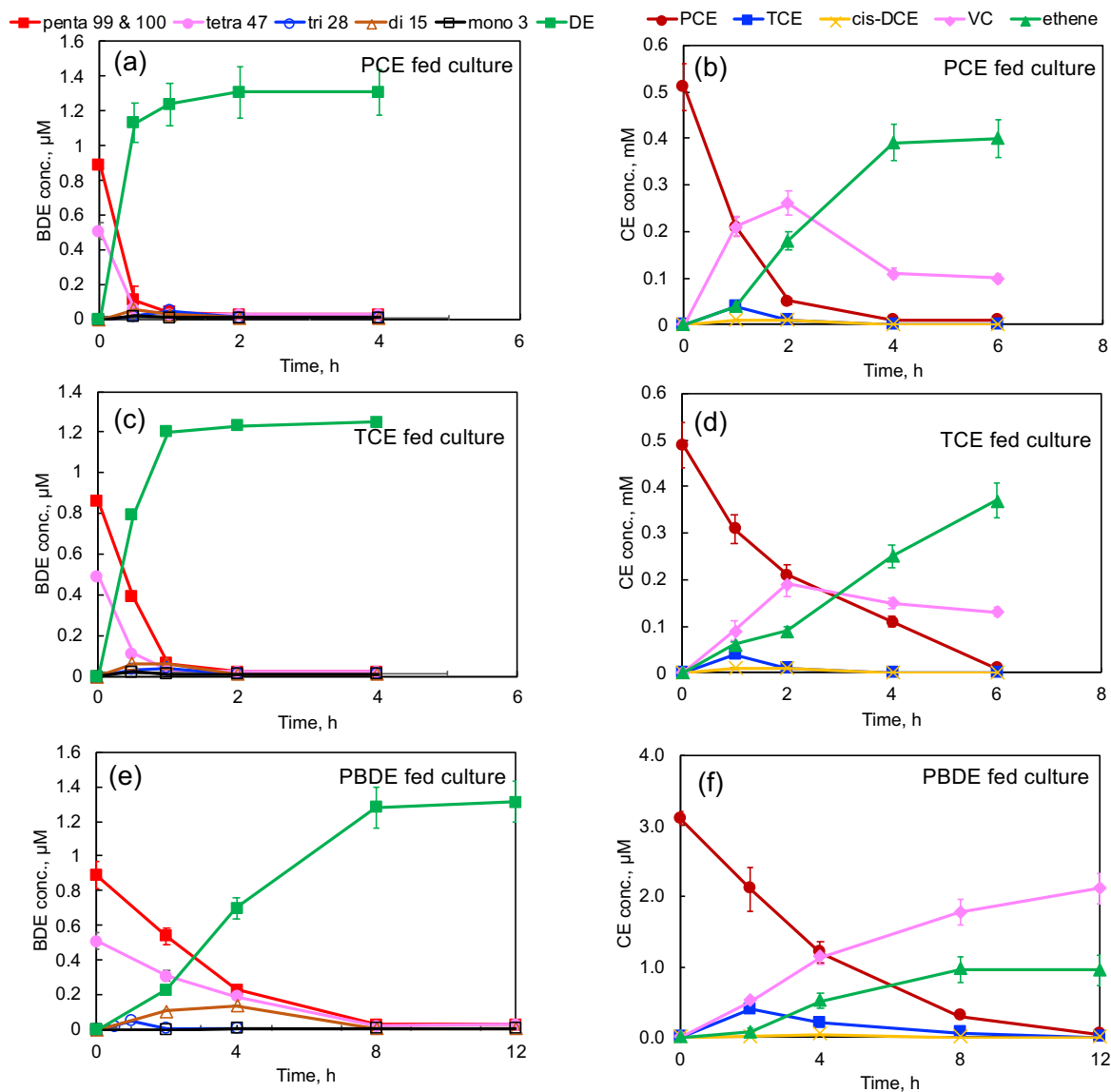


Figure S6. Enzymatic *in vitro* assays of crude cell extracts of *D. mccartyi* strain TZ50 cultivated with PCE (0.6 mM), TCE (0.6 mM), or the penta- and tetra-BDE mixture (1.5 µM). Panels a, b, and c show *in vitro* dehalogenation of the penta- and tetra-BDE mixture by cells extracted from cultures amended with PCE, TCE and the penta- and tetra-BDE mixture, respectively. Panels b, d, and f show *in vitro* dehalogenation of PCE by cells extracted from cultures amended with PCE, TCE and the penta- and tetra-BDE mixture, respectively.

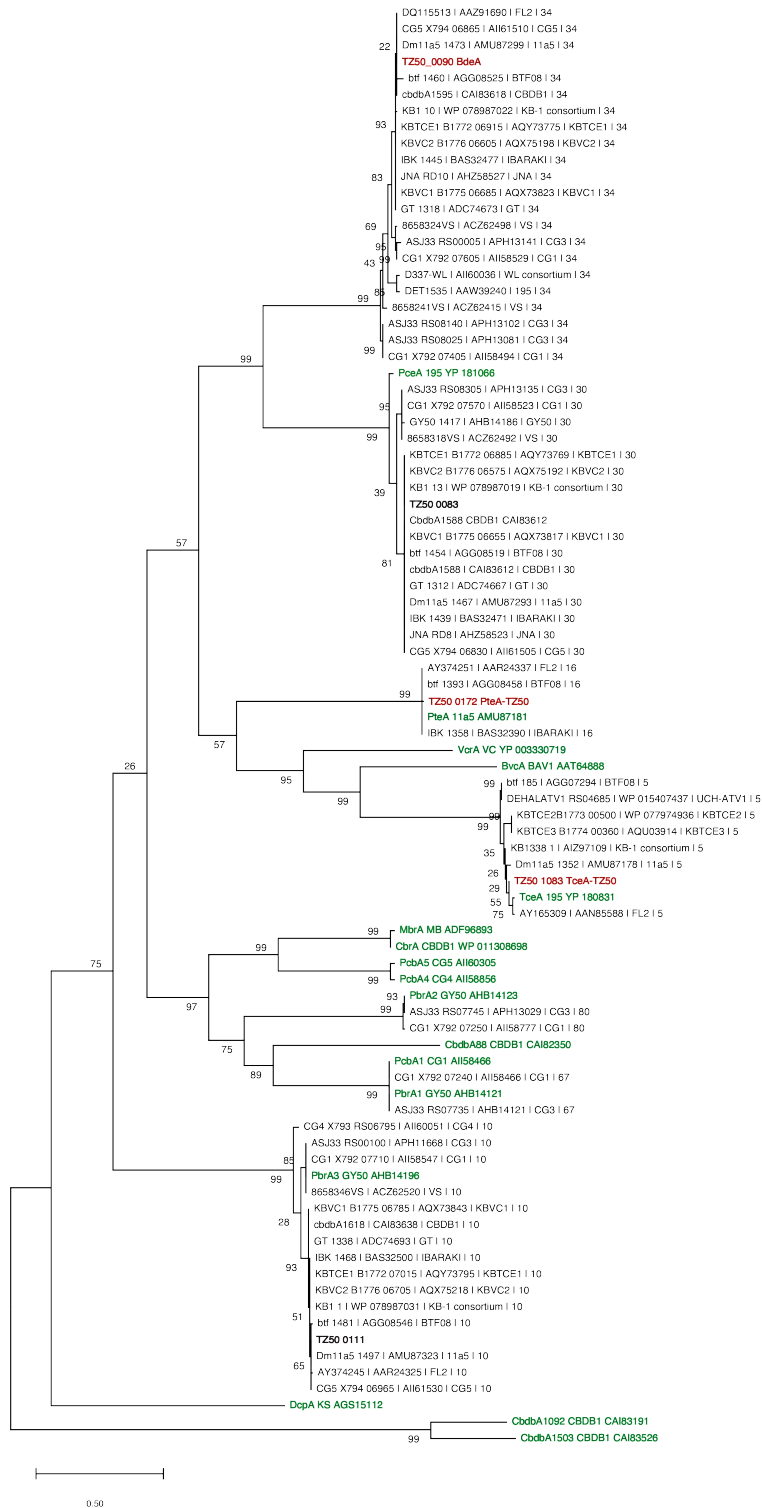


Figure S7. Maximum likelihood dendrogram of identified PBDE reductive dehalogenases (RDase) in *D. mccartyi* strains TZ50 and GY50, as well as associated orthologous and functionally characterized RDases in other *D. mccartyi*. The final tree is supported by 1,000 bootstrap replicates. Where the information is available, locus tag, NCBI accession number, and source of amino acid sequences included in the analysis are indicated next to the leaves on the dendrogram. Green font indicates functionally characterized RDases in other *D. mccartyi*; red font indicates PBDE RDases in strain TZ50, black font in bold indicates other RdhAs in strain TZ50.

Tables.**Table S1.** Genomic features of *D. mccartyi* strain TZ50

Genome features	
# of Contigs (>1 kb)	8
# of Scaffolds	6
Size	1.41 Mbp (N50 = 0.81 Mbp)
GC content	47.1%
Averaged coverage	1451.3
# of predicted open reading frames	1,526
# of 16S rRNA genes	1
# of tmRNA genes	1
# of tRNA genes	46

Table S2. Orthologous group (OG) classification of RdhA in *D. mccartyi* strain TZ50.

RdhAs ID	OG	Functional members of OG
TZ50_0034	73	
TZ50_0037	72	
TZ50_0040	NA	
TZ50_0043	51	
TZ50_0046	NA	
TZ50_0055	14	
TZ50_0066	33	
TZ50_0070	32	
TZ50_0073	19	
TZ50_0077	40	
TZ50_0083	30	PceA
TZ50_0090	34	BdeA
TZ50_0093	11	
TZ50_0111	10	PbrA3
TZ50_0117	17	
TZ50_0120	13	
TZ50_0127	15	
TZ50_0172	16	PteA
TZ50_0391	23	
TZ50_0430	22	
TZ50_0436	29	
TZ50_1083	5	TceA
TZ50_1230	341	
TZ50_1526	17	
TZ50_1528	12	

Table S3: Microcosms established in this study.

Location	Source	GPS	Collection date	Designation
Taizhou, Zhejiang, China	e-waste recycling site	121.38251, 28.54067	12-11-2016	TZ1
Taizhou, Zhejiang, China	e-waste recycling site	121.37524, 28.54084	12-11-2016	TZ2
Taizhou, Zhejiang, China	e-waste recycling site	121.36764, 28.53859	12-11-2016	TZ3
Taizhou, Zhejiang, China	e-waste recycling site	121.38800, 28.54472	12-11-2016	TZ4
Taizhou, Zhejiang, China	e-waste recycling site	121.39279, 28.55529	12-11-2016	TZ5
Taizhou, Zhejiang, China	e-waste recycling site	121.39590, 28.59311	12-11-2016	TZ6
Taizhou, Zhejiang, China	e-waste recycling site	121.39573, 28.60287	12-11-2016	TZ7
Taizhou, Zhejiang, China	e-waste recycling site	121.39583, 28.59301	12-11-2016	TZ8
Singapore	wastewater treatment plant	1.42852, 103.75814	05-03-2018	WWTP-N
Singapore	wastewater treatment plant	1.31197, 103.68323	20-06-2018	WWTP-W
Singapore	wastewater treatment plant	1.31647, 104.00443	13-04-2018	WWTP-E
Singapore	wastewater treatment plant	1.33268, 103.75542	06-03-2018	WWTP-U
Singapore	constructed wetlands	1.39572, 103.92418	28-02-2018	LH-1
Singapore	constructed wetlands	1.39492, 103.92310	28-02-2018	LH-2
Singapore	constructed wetlands	1.39532, 103.92333	28-02-2018	LH-3
Singapore	constructed wetlands	1.39584, 103.92305	28-02-2018	LH-4
Singapore	constructed wetlands	1.39441, 103.92344	28-02-2018	LH-5
Singapore	constructed wetlands	1.39433, 103.92397	28-02-2018	LH-6
Zhanjiang, Guangdong, China	industrial sediments	21.20910, 110.42186	24-10-2016	ZJ-1
Zhanjiang, Guangdong, China	industrial sediments	21.20894, 110.42177	24-10-2016	ZJ-2
Singapore	lake sediments	1.35161, 103.81882	01-03-2018	MR-1
Singapore	lake sediments	1.34835, 103.81427	01-03-2018	MR-2
Singapore	lake sediments	1.39140, 103.72086	11-06-2018	MR-3
Singapore	lake sediments	1.41693, 103.71973	11-06-2018	MR-4