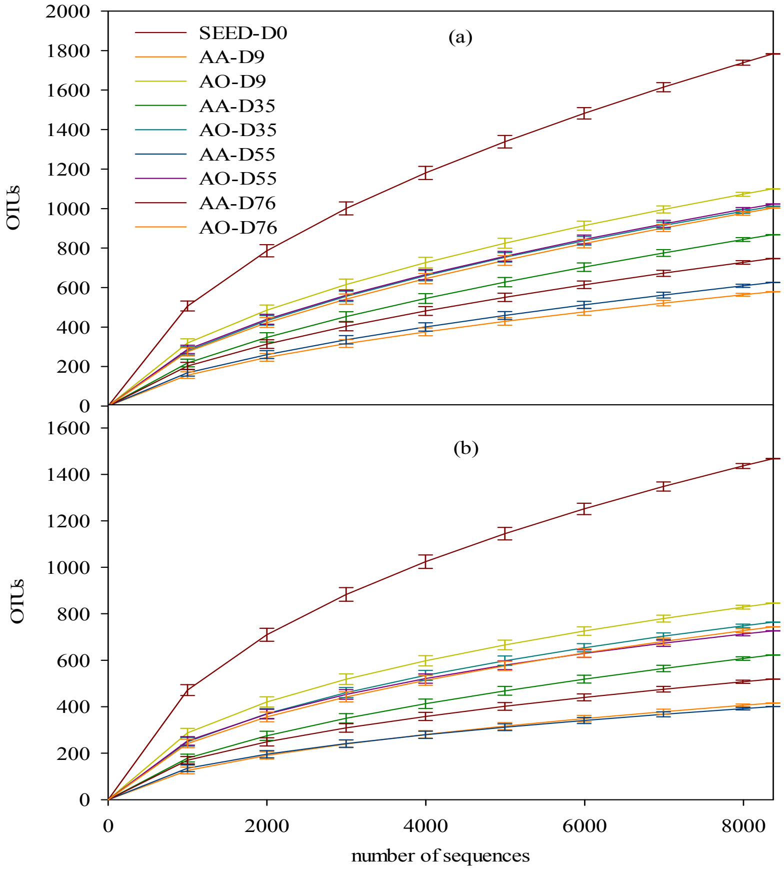


Fig. S1. Rarefaction curves of the 9 samples at cutoff levels of 3% (a) and 6% (b). The rarefaction curve, plotting the number of observed OTUs as a function of the number of sequences, was computed using RDP Pyrosequencing Pipeline Rarefaction tool.

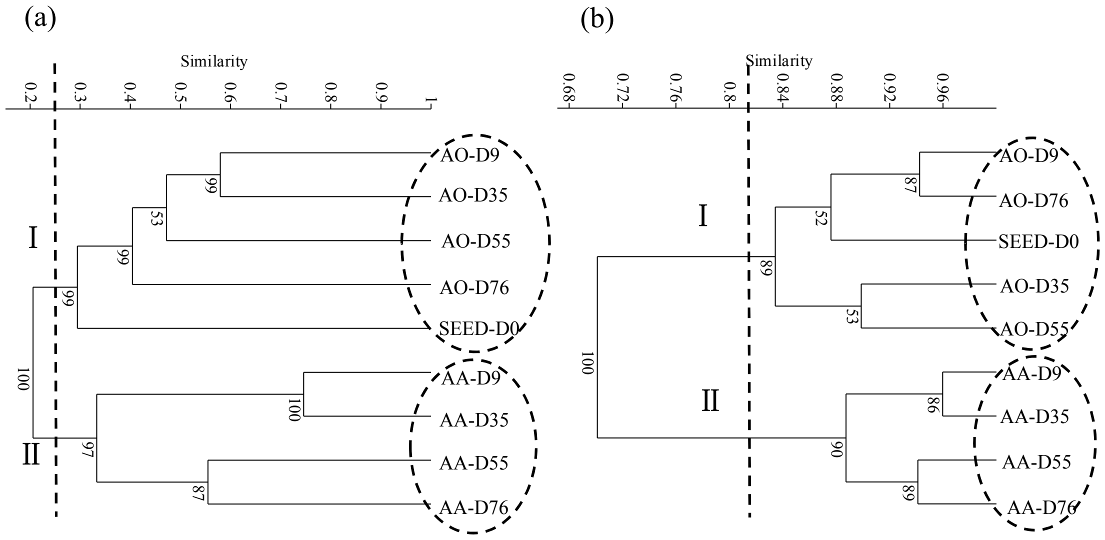


Fig. S2. Cluster analysis based on Bray-Curtis distances of 9 samples. (a) at 3% cutoff OTU level; (b) at Phylum level. The dot lines show the similarity cutoff levels to cluster the 9 sludge samples into distinct A-A and A-O groups.

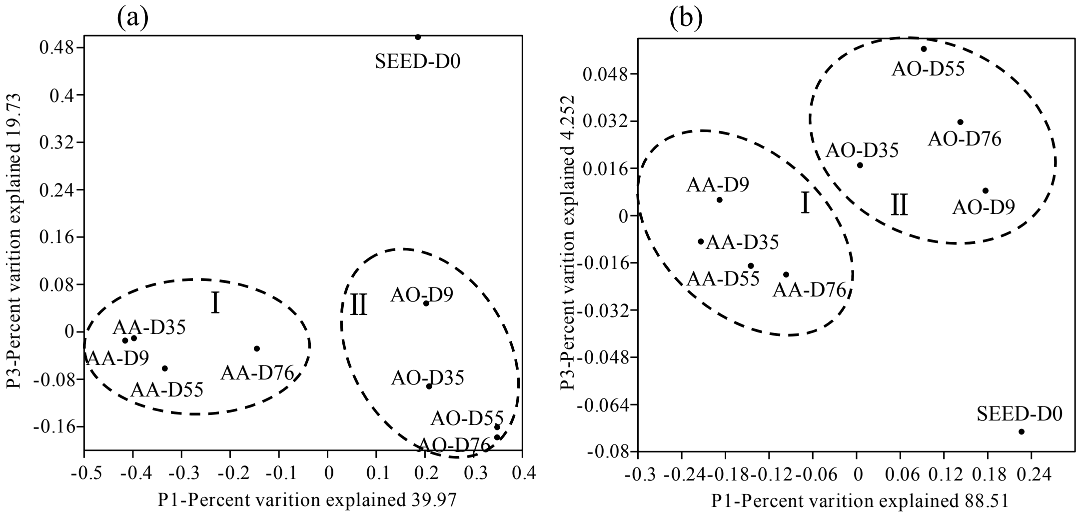
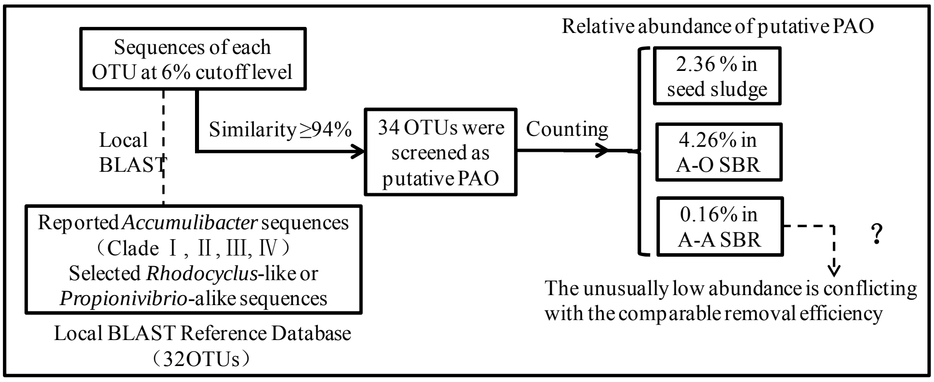


Fig. S3. PCoA based on Bray-Curtis distances of 9 sludge samples. (a) at 3% cutoff-OTU level; (b) at Phylum level.

Traditional strategy

(a)



Proposed strategy

(b)

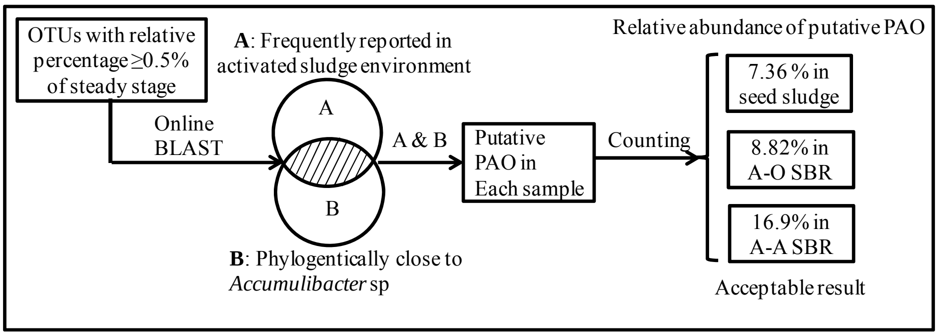


Fig. S4. Analysis scheme for identification of putative PAO. For identification of above-mentioned novel DPAO, another strategy emphasizing isolation source of related NCBI Genbank sequences was attempted, as illustrated by Fig.S4. Two assumptions were hypothesized for this strategy: (1) the novel DPAO should be an abundant population (with a least percentage above 0.5% or so) since too tenuous population was still incapable to explain the significant phosphorus removal efficiency. Theoretically, the situation of “vast but diverse” DPAO population should be considered, but this was conflicting with the ecological rule that population in the overlapped ecological niche could not co-exist after long time evolution. This assumption was also partially testified by the fact that more than 94% of the identified PAO in A-O SBR through traditional strategy had a relative abundance higher than 0.5%. (2) The DPAO sequence should be frequently recorded in NCBI Genbank, although they might not be correctly annotated as PAO or DPAO. Based on above two assumptions, all clustered OTUs in seed sludge, A-A and A-O sludge were separately ranked by their relative abundance. Only the abundant OTUs (relative abundance >0.5%) were further proceeded to avoid huge unnecessary work. Then the abundant OTUs were subjected to online BLAST for a comprehensive survey of the isolation source of their close relatives.

Table S1 Ranked OTUs in A-A SBR by their relative abundances and isolation environments description of their related sequence.$^{\delta}$

OTUs	Proportion (%)		Similar sequence			Major environments
	A/A	A/O	Sequence NO.	Similarity (%)	Origin	
1*	16.8	4.56	JQ072864	100	Full-scale EBPR (12)	Widely reported in biological P-removal and wastewater treatment systems
			HQ158656	100	SBR (25)	
21*	14.5	1.36	HQ917038	100	SBR (2)	Widely reported in water treatment systems, phylogenetically similar to *Thauera*
			HQ403231	100	SBR (10)	
25	9.01	7.51	JF817650	100	Microbial fuel cells (24)	Commonly reported in anaerobic conditions, soil and sediment
			HQ049856	93	Oil sands tailings pond (17)	
462	8.38	0.02	FM242265	100	Coastal sediment (15)	Commonly reported in anaerobic conditions, soil and sediment
			AM422228	98	Sediment (7)	
176	2.49	0.08	JF817705	100	Microbial fuel cells (24)	Commonly reported in microbial fuel cells
			FJ375431	100	Microbial fuel cells (13)	
6	2.35	0.12	CU918778	100	Anaerobic digestion of sludge (18)	Commonly reported in anaerobic conditions
			FJ66054	100	Pilot-scale WWTP (19)	
312*	1.92	0.03	JN090838	97	EBPR SBR (9)	Widely reported in biological P-removal wastewater treatment systems
			HQ010731	97	SBR (11)	
806	1.92	0.02	GU300327	90	Coastal sediment (1)	Commonly reported in soil and sediment.
			AF507646	89	Soil (6)	
129*	1.80	0.62	FJ434606	100	Bioreactor (4)	Widely reported in wastewater treatment systems and also the biological P-removal systems
			AF502223	100	SBR (14)	
34	1.62	0.65	JF345459	100	Citrus rhizosphere (22)	Commonly reported in soil and plant rhyzosphere
			JQ965569	100	Cropped rice (5)	
293	1.49	1.00	HQ119190	100	Soil (23)	Commonly reported in soil and plant rhyzosphere
			GU375391	100	Soil (3)	
87	1.44	2.56	HE602881	100	Caves (16)	Commonly reported in soil, sediment and occasional

			JN107387	100	Plant (8)	in wastewater treatment systems
100	1.28	0.25	JN563818	100	Sea (20)	Commonly reported in soil, sediment and occasional in wastewater treatment systems
			JQ965582	100	Cropped rice (5)	
801	1.03	0	HQ010842	93	SBR (11)	Commonly reported in soil and wastewater treatment systems
			AY157111	93	WWTP (21)	

* The related sequences were widely reported in biological phosphorus removal wastewater treatment systems

δ OTUs with significant higher hit ratio originated from nutrient removal activated sludge environment may indicate their universal distribution and functioning in such environments, therefore they should be especially concerned in DPAO identification.

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Table S2. Sequences and references for GAO BLAST

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