Supporting Information

Knocking out *OsPT4* gene decreases arsenate uptake by rice plants and inorganic arsenic accumulation in rice grains

Yue Cao,[†] Dan Sun,[†] Hao Ai,[‡] Hanyi Mei,[†] Xue Liu,[†] Shubin Sun,[‡] Guohua Xu,[‡] Yungen Liu,[⊽] Yanshan Chen^{*,†} Lena Q. Ma^{†,§}

[†]State Key Lab of Pollution Control and Resource Reuse, School of the Environment, Nanjing University, Jiangsu 210023, China

State Key Laboratory of Crop Genetics and Germplasm Enhancement, Key Laboratory of Plant Nutrition and Fertilization in Low-Middle Reaches of the Yangtze River, Ministry of Agriculture, Nanjing Agricultural University, 210095, China

∇Research Institute of Rural Sewage Treatment, South West Forestry University, Yunnan 650224, China

§Soil and Water Science Department, University of Florida, Gainesville, FL 32611, United States

*Corresponding author at State Key Laboratory of Pollution Control and Resource Reuse, School of the Environment, Nanjing University, Jiangsu 210023, China; +86 025 8968 0631

E-mail addresses: chenyanshan@nju.edu.cn, lqma@ufl.edu

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Figure S1. Characterization of mutants of *OsPT4* gene. (A) Position of T-DNA / Tos17 insertion of *ospt4-1* (*M1*) / *ospt4-2* (*M2*) mutants. Schematic representation of genes showing UTR (empty boxes) and CDS (black boxes) with numbers indicating length of each of them. (B) Identification of homozygous *ospt4* mutants. Two-rounds PCR were used to identify homozygous mutants by combination of PCR products. (C) Evaluation of gene-silencing effects on homozygous *ospt4* mutants (*M 1/2*) compared with wild-types (WT 1/2). The expression level was determined by quantitative reverse transcription PCR (qRT-PCR). *HistoneH3* and *Actin* were used as internal standards. Values are means \pm SE (n = 3).



Figure S2. As distribution (total As in shoots / total As in roots) in *ospt4* mutants and wild-types. 10-d old rice seedlings were cultivated hydroponically under P sufficient (+P) or P deficient (-P) conditions for 14 d, after which seedlings were exposed to 20 μ M AsV for 3 d. As distribution of *ospt4* mutants (M1/2) and wild-types (WT1/2) were analyzed. Values are means \pm SE (n = 4) and asterisk indicates that the values for *ospt4* mutants differ significantly (*P < 0.05) compared with wild-types. DW, Dry weight.



Figure S3. Total P concentration in *ospt4* mutants and wild-types. Total P concentration in shoots and roots of *ospt4* mutants (M 1/2) and wild-types (WT 1/2) after 14 days under different P regimes. Values are means \pm SE (n = 4) and asterisk indicates that the values for *ospt4* mutants differ significantly (*P < 0.05) compared with wild-types. DW, Dry weight.

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		Sequences
Mutants 1-2round-PCR	(5'-3')	GAGAGCATCATCGGTTACATCTTCTC
Mutants 2- 2round-PCR	(5'-3')	TTCGGGGGGGATCATGGACGTACCAG
OsPT4-2round-PCR	F(5'-3')	ATCTTCTCGTAGCCACGTGC
	R(5'-3')	CCTTCTGGAACAGGTTCGAC
OsActin-qRT-PCR	F(5'-3')	GGGTTCACAAGTCTGCCTATTGT
	R(5'-3')	ACGGGACACGACCAAGGA
OsHistone3-qRT- PCR	F(5'-3')	AGTTTGGTCGCTCTCGATTTCG
	R(5'-3')	CGCTTCCGTACGAGTGGTAGT
OsPT4-qRT- PCR	F(5'-3')	TTCTGCTAGTGTACCAAACAAAATTACA
	R(5'-3')	GTAAGTGGCATTTATAATATCAACAGTAACC

Table S1. Primers used for *ospt4* mutants identification and qRT-PCR analysis.

Rice line and P treatment	Vmax (nmol g ⁻¹ root FW h ⁻¹)	Km (μm)	Linear Slope	r ² _{adj}
WT1 -P	216.6±7.8	6.4±0.805	/	0.991
<i>M1</i> -P	201.7±13.9	8.5±1.8	/	0.977
WT2 -P	360.6±38.7	11.9±3.6	/	0.962
<i>M2</i> -Р	215.1±29.4	8.4 ± 3.6	/	0.905
WT1 +P	/	/	9.2±1.9	0.973
MI +P	/	/	6.6±1.9	0.959
WT2 +P	/	/	5.6 ±2.2	0.951
<i>M2</i> +P	/	/	7.1 ±2.7	0.919

Table S2. Fitted parameters of arsenate uptake kinetics of *ospt4* mutants and wild types.