

Species	<i>Arabidopsis thaliana</i>	<i>Oryza sativa</i>	<i>Oryza coarctata</i>	<i>Zea mays</i>	<i>Picea sitchensis</i>	<i>Pteris vittata</i>	<i>Selaginella moellendorffii</i>	<i>Marchantia polymorpha</i>	<i>Klebsormidium nitens</i>	<i>Volvox carteri</i>	<i>Dictyostelium discoideum</i>
<i>Arabidopsis thaliana</i>		80.6	83.1	78.4	82.3	84.1	80.7	84.1	78.4	67.4	59.4
<i>Oryza sativa</i>	80.6		88.6	84.8	88	83.8	81.9	83.6	80	68.1	59.7
<i>Oryza coarctata</i>	83.1	88.6		86.2	90.2	86.9	83	85.1	79.5	66.6	62
<i>Zea mays</i>	78.4	84.8	86.2		84.4	83.3	76.3	78.9	76.9	65.6	60.3
<i>Picea sitchensis</i>	82.3	88	90.2	84.4		87.2	85	86.2	79.6	66.6	60.9
<i>Pteris vittata</i>	84.1	83.8	86.9	83.3	87.2		85.8	90	82.5	64.8	64.6
<i>Selaginella moellendorffii</i>	80.7	81.9	83	76.3	85	85.8		84.4	78.4	66.4	61
<i>Marchantia polymorpha</i>	84.1	83.6	85.1	78.9	86.2	90	84.4		82.1	63.9	61.3
<i>Klebsormidium nitens</i>	78.4	80	79.5	76.9	79.6	82.5	78.4	82.1		66.5	56.3
<i>Volvox carteri</i>	67.4	68.1	66.6	65.6	66.6	64.8	66.4	63.9	66.5		57.2
<i>Dictyostelium discoideum</i>	59.4	59.7	62	60.3	60.9	64.6	61	61.3	56.3	57.2	

Figure S1. Sequence identity of *NHX1* in representative plant species.

	<i>OsNHX1</i>	<i>OsNHX2</i>	<i>OsNHX3</i>	<i>OsNHX4</i>	<i>OsNHX5</i>	<i>OsNHX6</i>	<i>OsNHX7</i>
<i>OsNHX1</i>		81.7	80.3	63	28.4	35	20.3
<i>OsNHX2</i>	81.7		79.9	62.6	30.1	34.5	20.6
<i>OsNHX3</i>	80.3	79.9		61.9	29.1	36.1	19.4
<i>OsNHX4</i>	63	62.6	61.9		26.8	29.8	18.1
<i>OsNHX5</i>	28.4	30.1	29.1	26.8		49.4	15.5
<i>OsNHX6</i>	35	34.5	36.1	29.8	49.4		14.9
<i>OsNHX7</i>	20.3	20.6	19.4	18.1	15.5	14.9	

Figure S2. Sequence identity of putative NHXs mapped in *Oryza sativa*.

	<i>O. sativa</i> subsp. <i>japonica</i> <i>O. sativa</i> subsp. <i>indica</i> <i>O. glaberrima</i> <i>O. nivara</i> <i>O. rufipogon</i> <i>O. meridionalis</i> <i>O. glumaepatula</i> <i>O. punctata</i> <i>O. barthii</i> <i>O. brachyantha</i> <i>O. coarctata</i>										
<i>O. sativa</i> subsp. <i>japonica</i>		91.8	91.8	91.6	91.4	91.4	87.9	90.3	90.6	89	89.3
<i>O. sativa</i> subsp. <i>indica</i>	91.8		100	99.8	99.6	99.6	95.7	98.5	99.6	97.9	97.2
<i>O. glaberrima</i>	91.8	100		99.8	99.6	99.6	95.7	98.5	99.6	97.9	97.2
<i>O. nivara</i>	91.6	99.8	99.8		99.8	99.4	95.9	98.3	99.4	98.1	96.8
<i>O. rufipogon</i>	91.4	99.6	99.6	99.8		99.3	95.7	98.1	99.2	97.9	96.8
<i>O. meridionalis</i>	91.4	99.6	99.6	99.4	99.3		95.3	98.7	99.2	98.1	97.2
<i>O. glumaepatula</i>	87.9	95.7	95.7	95.9	95.7	95.3		94.2	95.2	93.5	92.9
<i>O. punctata</i>	90.3	98.5	98.5	98.3	98.1	98.7	94.2		98.3	98.3	97.8
<i>O. barthii</i>	90.6	99.6	99.6	99.4	99.2	99.2	95.2	98.3		97.5	97.5
<i>O. brachyantha</i>	89	97.9	97.9	98.1	97.9	98.1	93.5	98.3	97.5		97.9
<i>O. coarctata</i>	89.3	97.2	97.2	96.8	96.8	97.2	92.9	97.8	97.5	97.9	

Figure S3. Sequence identity of NHX homologues identified in the *Oryza* genus.

	Logo	E-value	Site	Width	Role
1.		1.4e-514	12	50	
2.		3.2e-495	12	50	
3.		1.0e-494	12	50	
4.		2.5e-478	12	50	
5.		2.4e-337	10	50	
6.		2.7e-339	10	50	
7.		3.0e-229	11	29	Aspartate-alanine exchange (Asp-Al_Ex)
8.		1.2e-217	12	29	
9.		1.9e-216	12	29	Monovalent Cation:Proton Antiporter-1 (CPA1)
10.		2.9e-215	12	29	

Figure S4. Various protein motifs found in representative *Oryza* NHX1s.

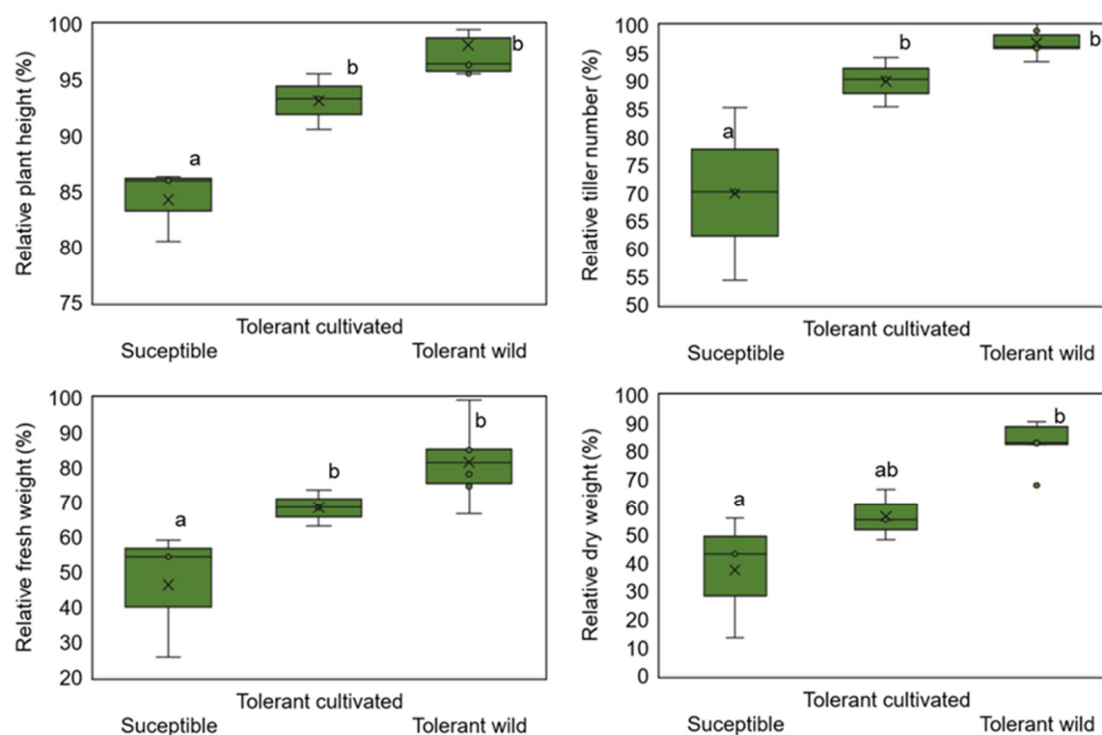


Figure S5. Variation of salinity tolerance in *Oryza* species. Phenotypic variation on the effect of salinity treatment on overall growth, Relative effects (% of control) of salinity on plant height, tiller number, fresh weight, and and dry weight. Different lowercase letters indicate significant differences at $P < 0.05$ ($n = 3-6$ *Oryza* lines) according to Duncan's Multiple Range Test.

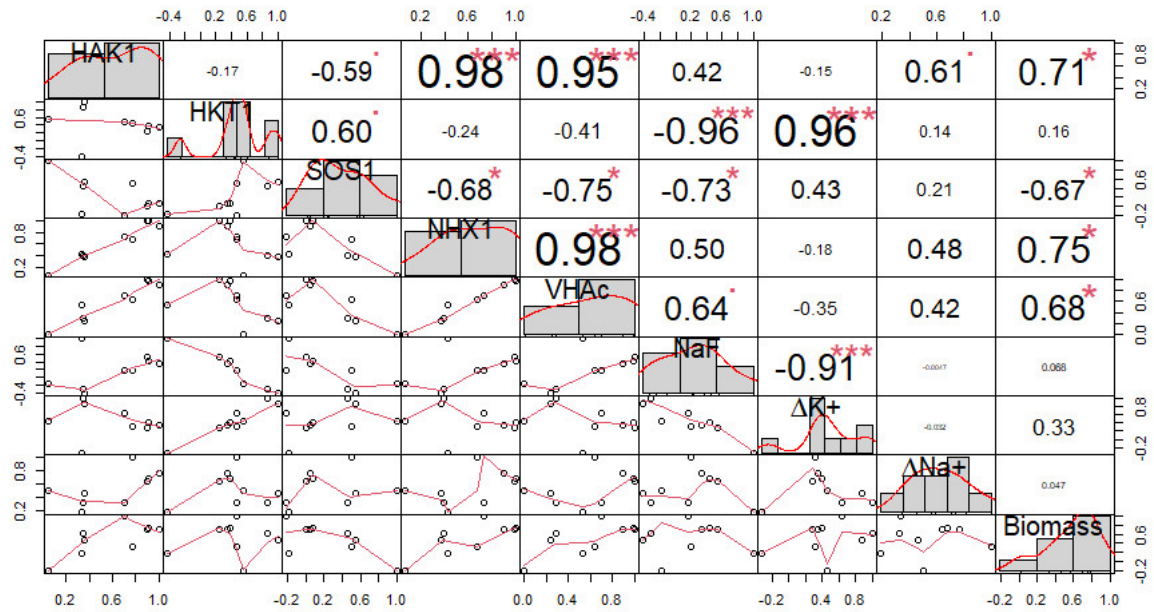


Figure S6. Relationship of gene expression of salinity transporters in ion transport and yield in response to salinity stress in cultivated and wild rice species. Spearman correlation matrix and histogram of phenotypic traits observed and gene expression of known salinity tolerant genes. The distribution of each variable is shown on the diagonal. ***, **, * indicate significant correlation at $P < 0.001$, $P < 0.01$ and $P < 0.05$. NaF- relative mesophyll Na^+ fluorescence, ΔK^+ - K^+ flux difference(stress-control), ΔNa^+ - Na^+ flux difference (stress-control), Biomass – relative above ground biomass harvested at full maturity, *HAK1*, *HKT1*, *SOS1*, *NHX1* and *VHAc* - relative gene expression under salinity stress.

Table S1. Identification and sequence analysis of NHX1 in representative species.

Classification	Species	Length (aa)	MW (kD)	PI
Dicot	<i>Arabidopsis thaliana</i>	529	58.87	7.84
	<i>Oryza sativa</i>	545	59.91	7.93
Monocot	<i>Oryza coarctata</i>	539	59.45	7.85
	<i>Zea mays</i>	515	57.55	8.21
Gymnosperm	<i>Picea sitchensis</i>	535	59.09	8.29
Fern	<i>Pteris vittata</i>	463	50.91	6.1
Lycophyte	<i>Selaginella moellendorffii</i>	532	58.93	8.58
Liverwort	<i>Marchantia polymorpha</i>	521	57.64	6.62
Steptophyte Alga	<i>Klebsormidium nitens</i>	547	59.69	6.65
Chlorophyte Alga	<i>Volvox carteri</i>	474	50.95	9.56
Dictyosteliida	<i>Dictyostelium discoideum</i>	674	75.52	4.31

Table S2. Number of NHX transporters mapped in representative species.

Classifications	Species	NHX subtypes			Total
		Vac	PM	TGN	
Algae	<i>Chlamydomonas reinhardtii</i>	1	3	1	5
Bryophytes	<i>Physcomitrella patens</i>	5	2	2	9
Lycophytes	<i>Selaginella moellendorffii</i>	2	2	2	6
Gymnosperm	<i>Picea sitchensis</i>	1	?	?	?
Monocots	<i>Zea Mays</i>	6	2	2	10
	<i>Sorghum bicolor</i>	4	2	1	7
Eudicots	<i>Oryza sativa</i>	4	2	1	7
	<i>Arabidopsis thaliana</i>	4	4	2	10
	<i>Gossypium hirsutum</i>	11	23	17	1
	<i>Solanum lycopersicum</i>	5	3	1	9
	<i>Glycine max</i>	11	7	3	21

Table S3. The identification and sequence analysis of the Na⁺/H⁺ antiporter (NHX) gene family in rice.

Gene	Rice MSU Locus ID	Length (aa)	MW (kD)	PI	Subcellular Localization
<i>OsNHX1</i>	LOC_Os07g47100.2	536	59.07	8.3	Vacuolar
<i>OsNHX2</i>	LOC_Os11g42790.1	546	59.92	7.9	Vacuolar
<i>OsNHX3</i>	LOC_Os05g05590.1	529	59.64	7.3	Vacuolar
<i>OsNHX4</i>	LOC_Os06g21360.1	560	58.22	6.3	Vacuolar
<i>OsNHX5</i>	LOC_Os09g11450.1	550	60.39	5.7	Plasma Membrane
<i>OsNHX6</i>	LOC_Os09g30446.1	462	31.07	5.5	Plasma Membrane
<i>OsNHX7</i>	LOC_Os12g44360.2	1149	127.91	6.8	Trans-Golgi Network

Table S4. Number of NHX transporters mapped in in representative *Oryza* species.

Species	Vac Class	Gene hit	PM Class	Gene hit	TGN Class	Gene hit
<i>Oryza sativa subsp. indica</i>	4	BGIOSGA027380 BGIOSGA026347 BGIOSGA018801 BGIOSGA029567	1	BGIOSGA035749	2	BGIOSGA029567 BGIOSGA030066
<i>Oryza sativa subsp. japonica</i>	4	OsNHX1 OsNHX2 OsNHX3 OsNHX4	1	SOS1	2	OsSTA230 OsNHX5
<i>Oryza rufipogon</i>	4	ORUF105G03390 ORUF106G13400 ORUF107G26490 ORUF111G23040	1	ORUF109G04390	2	ORUF109G15320 ORUF112G22630
<i>Oryza meridionalis</i>	3	OMERI05G03260 OMERI07G22210 OMERI11G17560	0	?	2	OMERI09G03010 OMERI09G11060
<i>Oryza nivara</i>	4	ONIVA05G03230 ONIVA06G14580 ONIVA07G25170 ONIVA11G21550	1	ONIVA12G18960	2	ONIVA09G04150 ONIVA09G14830
<i>Oryza glaberrima</i>	4	ORGLA09G0109500 ORGLA07G0266400 ORGLA07G0198900 ORGLA05G0031000	1	ORGLA12G0176700	2	ORGLA09G0109500 ORGLA12G0176700
<i>Oryza barthii</i>	4	OBART05G03250 OBART06G12670 OBART07G25570 OBART11G21190	1	OBART12G20310	2	OBART09G03930 OBART09G14270
<i>Oryza longistaminata</i>	4	AMDW01040861.1_ F KN538696.1_FG089 KN539398.1_FG001 KN540366.1_FG005	1	KN538756.1_FG016	2	KN539227.1_FG004 KN538880.1_FG014
<i>Oryza glumipatula</i>	4	OGLUM05G03300 OGLUM06G13560 OGLUM07G25540 OGLUM11G20600	1	OGLUM12G22080	2	OGLUM09G04670 OGLUM09G14920
<i>Oryza punctata</i>	4	OPUNC05G03110 OPUNC06G12060 OPUNC07G23980 OPUNC11G17770	1	OPUNC12G18600	2	OPUNC09G12770
<i>Oryza brachyantha</i>	5	OB05G13170 OB06G21140 OB07G30930 OB11G25980 OB12G17900	2	OB09G20810 OB12G27070	5	OB0092G10010 OB04G14430 OB09G12460 OB09G12470 OB12G18280

Table S5. Identification and sequence analysis of NHX1 in representative *Oryza* species.

NHX1 species	Genome	Length (aa)	MW (kD)	PI
<i>O. sativa subsp. japonica</i>	AA	535	59.07	7.77
<i>O. sativa subsp. indica</i>	AA	498	54.85	8.34
<i>O. glaberrima</i>	AA	535	59.07	8.34
<i>O. nivara</i>	AA	535	59.07	8.34
<i>O. rufipogon</i>	AA	535	59.07	8.34
<i>O. meridionalis</i>	AA	536	59.16	8.34
<i>O. glumaepatula</i>	AA	513	56.52	8.84
<i>O. longistaminata</i>	AA	539	59.88	9.51
<i>O. barthii</i>	BB	475	52.70	9.56
<i>O. punctata</i>	BB	536	58.95	8.10
<i>O. brachyantha</i>	FF	477	52.74	9.27
<i>O. coarctata</i>	KKLL	539	59.45	7.85

Table S6. Salinity ranking of *Oryza* species based on the relative value of the physiological parameters.

Species	Genome	Gene Pool	RFW	RDW	RWC	Na ⁺ accumulation	ΔK ⁺	ΔNa ⁺	Overall	Tolerance Rank
<i>O. brachyantha</i>	FF	Tertiary	1	1	3	1	1	1	1.2	Susceptible
<i>O. sativa</i> cv. IR29	AA	Primary	2	3	4	2	2	2	3.4	Susceptible
<i>O. sativa</i> cv. Koshihikari	AA	Primary	3	2	7	3	3	3	3.6	Susceptible
<i>O. punctata</i>	BB	Secondary	4	4	1	5	4	5	3.9	Moderately Tolerant
<i>O. rufipogon</i>	AA	Secondary	5	5	2	4	5	4	3.9	Moderately Tolerant
<i>O. australiensis</i>	EE	Secondary	6	6	6	6	6	6	6	Tolerant
<i>O. sativa</i> cv Pokkali	AA	Primary	7	7	5	7	7	7	7.2	Tolerant
<i>O. latifolia</i>	CCDD	Secondary	8	8	9	8	8	8	7.4	Tolerant
<i>O. alta</i>	CCDD	Secondary	9	9	8	9	9	9	8.6	Tolerant
<i>O. coarctata</i>	KKLL	Tertiary	10	10	10	10	10	10	10	Highly Tolerant

Abbreviations: RFW- relative fresh weight, RDW- relative dry weight, RWC- relative water content, Na⁺ accumulation- relative mesophyll Na⁺ fluorescence, ΔK⁺- K⁺ flux difference(stress/control), ΔNa⁺- Na⁺ flux difference (stress/control).