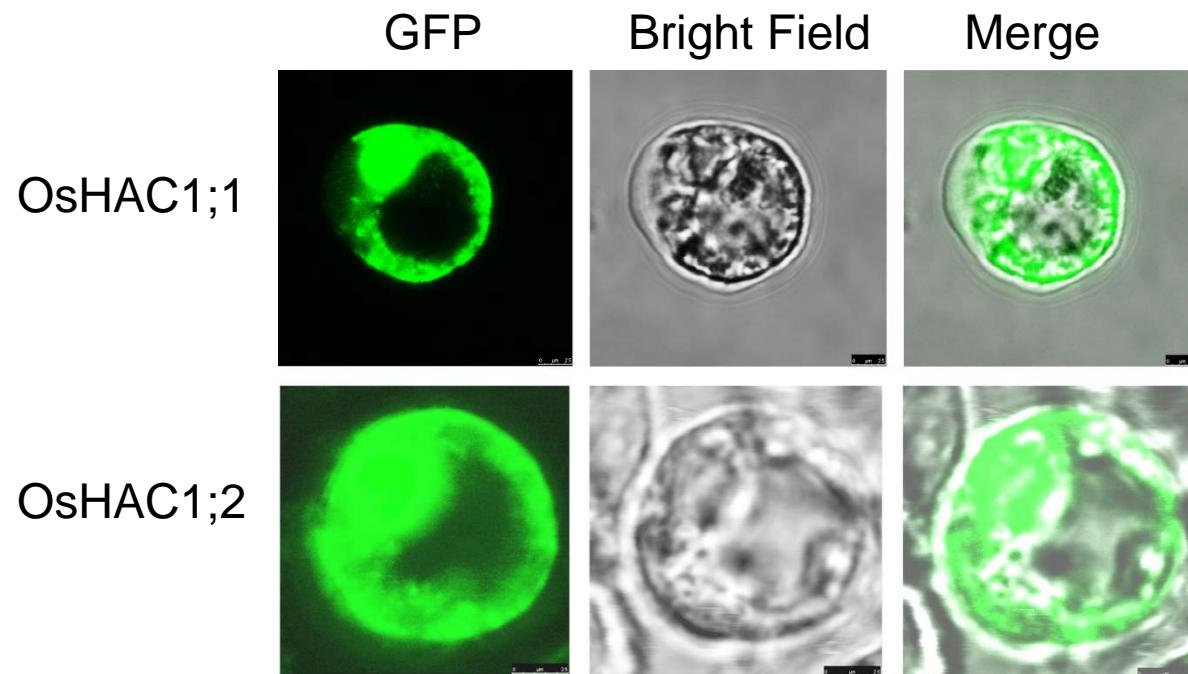
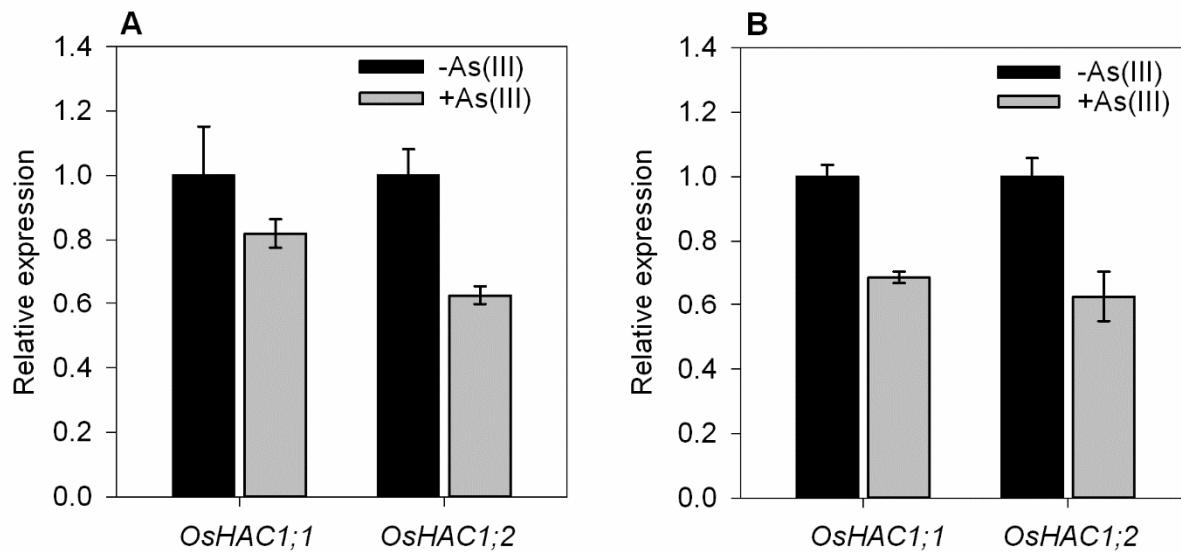


OsHAC1-1: Loc_Os02g01220
 OsHAC1-2: Loc_Os04g17660
 OsHAC2: Loc_Os06g50930
 OsHAC3: Loc_Os12g24020
 OsHAC4: Loc_Os02g06290
 OsHAC5: Loc_Os02g56050
 OsHAC6: Loc_Os02g38240
 OsHAC7: Loc_Os09g36040
 OsHAC8: Loc_Os03g18020
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 OsHAC10: Loc_Os03g64350
 OsHAC11: Loc_Os05g2578
 AtHAC1: Loc_At2g21045

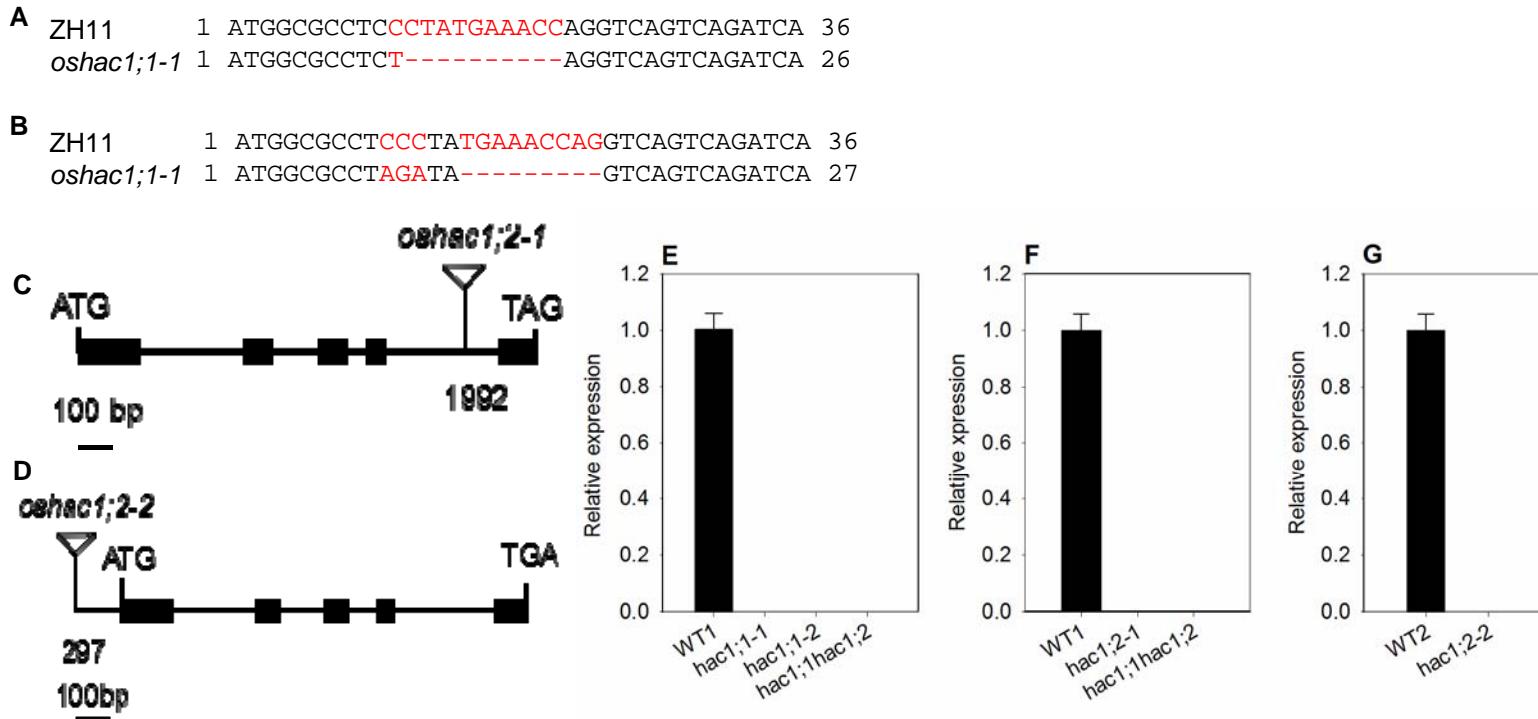
Supplemental Figure S1. Sequence analysis of *HAC* genes in rice and *Arabidopsis thaliana*. Phylogenetic analyses were conducted using MEGA version 6 (Tamura et al., 2013). Protein sequences were aligned using MUSCLE with UPGMB as clustering method. The tree was constructed using the Neighbor Joining (NJ) method. Bootstraps were carried out with 1,000 replications.



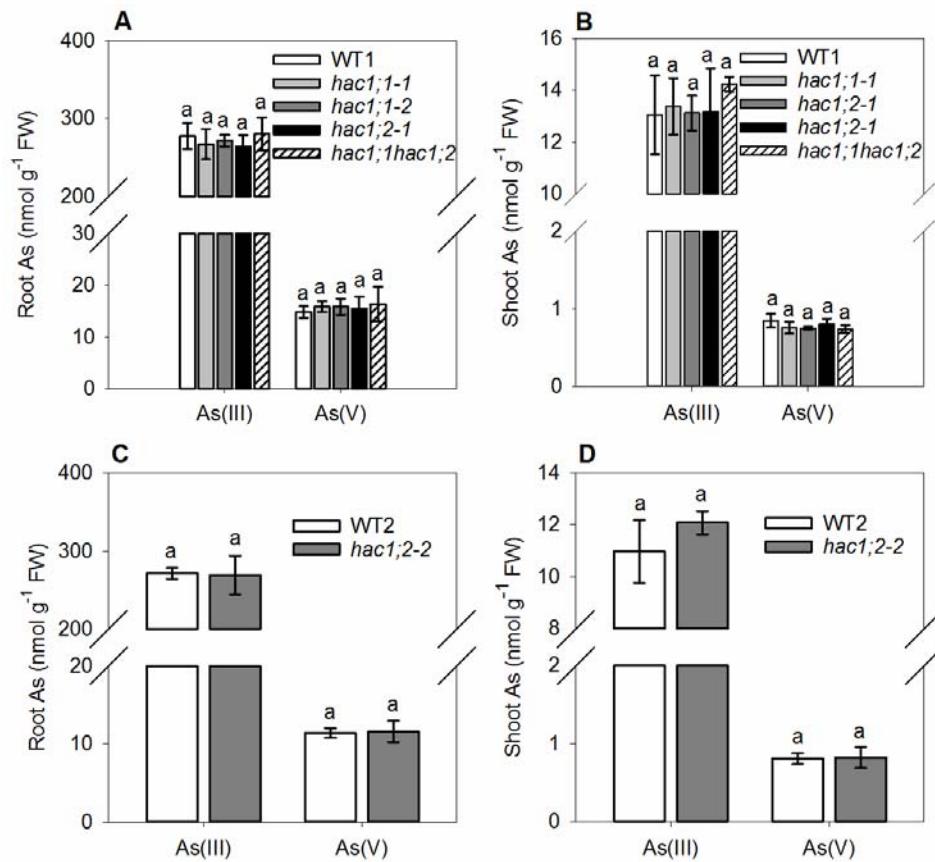
Supplemental Figure S2. Subcellular localization of OsHAC1;1 and OsHAC1;2. Protoplasts were isolated from the roots of transgenic rice plants expressing *pHAC1;1:OsHAC1;1-GFP* or *pHAC1;2:OsHAC1;2-GFP*. Top panel, scale bar=100 μ m. Bottom panel, scale bar = 25 μ m.



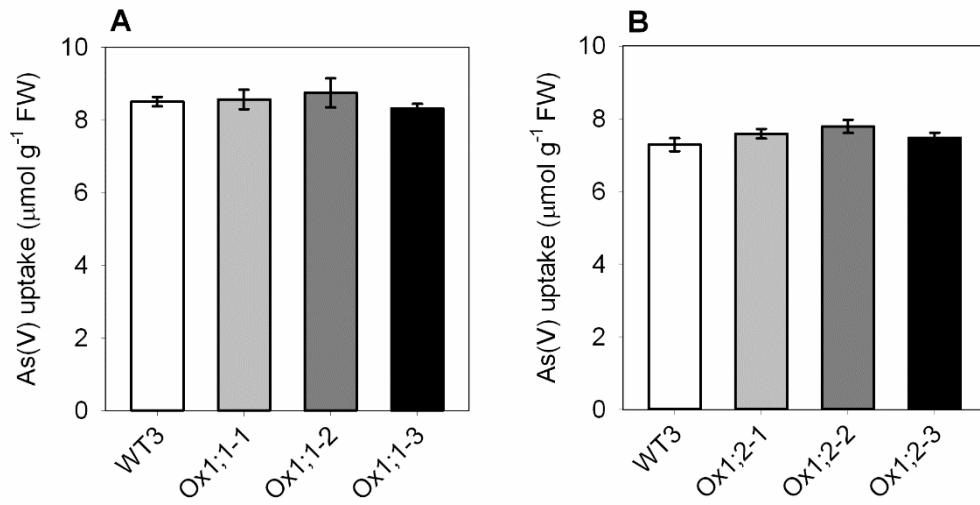
Supplemental Figure S3. Exposure to As(III) decreases the expression of *OsHAC1;1* and *OsHAC1;2*. The expression of *OsHAC1;1* and *OsHAC1;2* in roots (A) and shoots (B) after plants (cv Nipponbare) were exposed to 10 μ M As(III) for 24 h. The expression levels were determined using quantitative real-time PCR with *Actin* as the internal standard. Data represents means \pm S.D. ($n=3$ biological replicates).



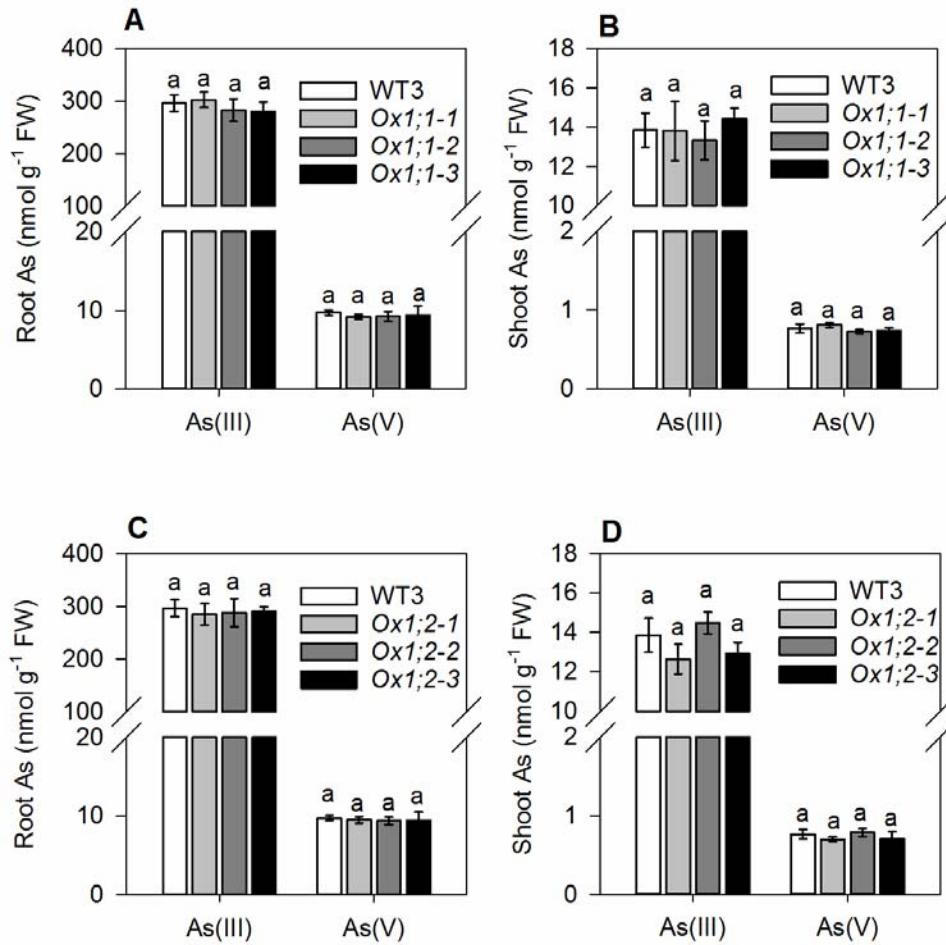
Supplemental Figure S4. Knockout mutants of *OsHAC1;1* (*hac1;1-1*, *hac1;1-2*), *OsHAC1;2* (*hac1;2-1*, *hac1;2-2*) and double mutant of *OsHAC1;1* and *OsHAC1;2* (*hac1;1:hac1;2*). (A, B) Two independent knockout mutants of the *OsHAC1;1* in the cv. Zhonghua 11 (ZH11) background generated by CRISPR-Cas9. (C, D) Two independent T-DNA insertion mutants of *OsHAC1;2* in the Zhonghua 11 (C) or Dongjin (D) background. The black boxes stand for exons, the lines stand for introns, and the triangle indicates the T-DNA insertion. (E, F, G) The expression levels of *OsHAC1;1* (E) and *OsHAC1;2* (F, G) in wild-type (WT1, Zhonghua 11; WT2, Dongjin) and mutants by quantitative real-time PCR with *Actin* as the reference gene.



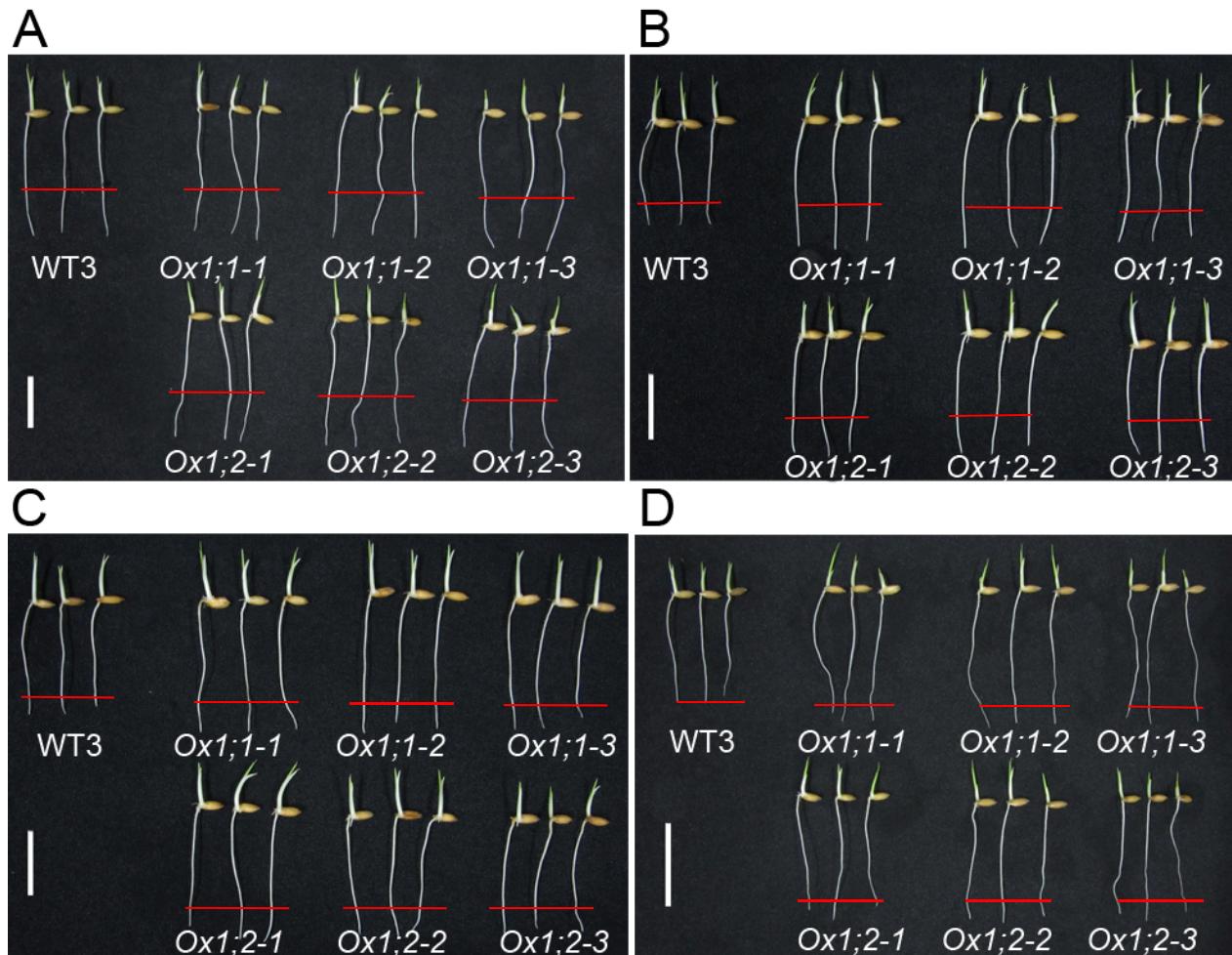
Supplemental Figure S5. Knocking out *OsHAC1;1* or *OsHAC1;2* has no significant effect on As(III) uptake and As accumulation in rice. A-D, As speciation in roots (A, C) and shoots (B, D) after wild-type and knockout mutant plants were exposed to 10 μ M As(III) for 48 h. WT1, cv Zhonghua 11; WT2, cv Dongjin. Data are means \pm S.E. ($n=4$ biological replicates).



Supplemental Figure S6. Overexpression of *OsHAC1;1* or *OsHAC1;2* has no significant effect on As(V) uptake. As(V) uptake by overexpressing lines of *OsHAC1;1* (A) and *OsHAC1;2* (B) and wild-type (Nipponbare) plants from nutrient solution containing 10 μM As(V) for 48 h. *Ox1;1-1*, *Ox1;1-2*, *Ox1;1-3* represent independent overexpression lines of *OsHAC1;1*. *Ox1;2-1*, *Ox1;2-2*, *Ox1;2-3* represent independent overexpression lines of *OsHAC1;2*. Data represents means \pm S.E. ($n=4$ biological replicates).



Supplemental Figure S7. Overexpression of *OsHAC1;1* or *OsHAC1;2* has no significant effect on As(III) uptake and As accumulation in rice. A-D, As speciation in roots (A, C) and shoots (B, D) after wild-type and knockout mutant plants were exposed to 10 μ M As(III) for 48 h. WT3, cv Nipponbare. *Ox1;1-1*, *Ox1;1-2*, *Ox1;1-3* represent independent overexpression lines of *OsHAC1;1*. *Ox1;2-1*, *Ox1;2-2*, *Ox1;2-3* represent independent overexpression lines of *OsHAC1;2*. Data represents means \pm S.E. ($n=4$ biological replicates).



Supplemental Figure S8. Root elongation of *OsHAC1;1* and *OsHAC1;2* overexpression lines and wild-type plants exposed to different concentrations of As(V): 0 µM (A), 2.5 µM (B), 3 µM (C) and 4 µM (D), for 24 h. WT3 represent wild-type plants (Nipponbare). *Ox1;1-1*, *Ox1;1-2*, *Ox1;1-3* represent independent overexpression lines of *OsHAC1;1*. *Ox1;2-1*, *Ox1;2-2*, *Ox1;2-3* represent independent overexpression lines of *OsHAC1;2*. The red line indicates root length at the beginning of As(V) treatment. Scale bars = 2 cm.

Supplemental Table S1. The primers used in this study.

Gene	Forward (5'-3')	Reverse (3'-5')
<i>T-DNA(NTLB)</i>	AATCCAGATCCCCCGAATT	
<i>RMD_03Z11FF65(oshac;2-1)</i>	CATTGTCTGACGGGTGAAAG	ACAAATCAAATTAACACCCAGTC
<i>T-DNA(pGA2715)</i>	AACGCTGATCAATTCCACAG	
<i>PFG_3A-02094(oshac1;2-2)</i>	ACAGGAGCACATCACACAGC	TGACGTCCAAGTAGCTGTGG
<i>CAS9-OsHAC1;1</i>	GTGTTGGCGCCTCCCTATGAAACC	AAACGGTTCATAGGGAGGCGCCA
<i>HAC1;1-CAS9S</i>	TTCTAGGCTAGCACTGCACG	GGAGTTGTAGATGCCCT
<i>OsHAC1;1-N</i>	TTGGTACCATGGCGCCTCCCTATGAAACCAGCG	TTGGATCCAATACATTGTATGAGGAGGAGTGT
<i>OsHAC1;2-N</i>	TTGGTACCATGATGATGGTGAGATTGCCT	TTGGATCCAGAGCTTCTTGCCTAGAGGTTA
<i>qActin(AB047313)</i>	CAACACCCCTGCTATGTACG	CATCACAGAGTCCAACACAA
<i>qUbq(Os03g13170)</i>	GACGGACGCACCCCTGGCTGACTAC	TGCTGCCAATTACCATATACCAACGAC
<i>OsHAC1;1-qPCR</i>	TGAACAAGGGCCATCTACAC	GACGAGAACTGCTCCACAAA
<i>OsHAC1;2-qPCR</i>	TAGCATCTGCCGATCTCATA	GAGGTTTATTACCGCAAGG
<i>OsHAC1;1-29a</i>	GATATCGGATCCATGGCGCCTCCCTATGAAAC	GTGGTGCTCGAGATACATTGTATGAGGAGGAG
<i>OsHAC1;2-29a</i>	GATATCGGATCCATGATGATGATGGTGAGATT	GTGGTGCTCGAGGAGCTTCTTGCCTAGAG
<i>pET29a</i>	TTCCATGGCTGATATCGGATCC	GTGGTGGTGGTGGTCTCGAG
<i>OsHAC1;1-Ox</i>	GGATCCATGGCGCCTCCCTATGAAACCAGCG	ACTAGTTAACATTGTATGAGGAGGAGTG
<i>OsHAC1;2-Ox</i>	GGATCCATGATGATGATGGTGAGATTGCCTG	ACTAGTCTAGAGCTTCTTGCCTAGAGGT