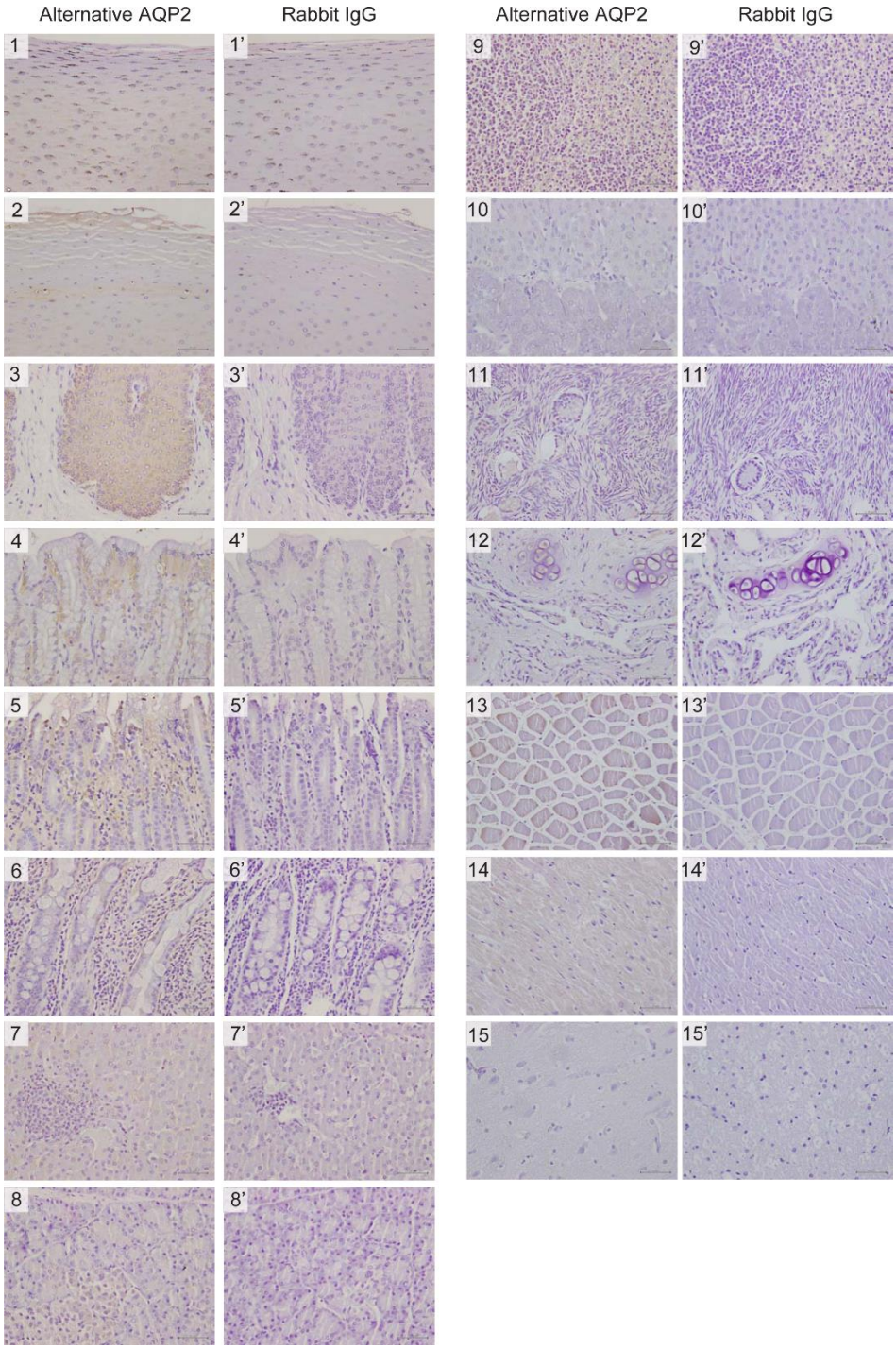

Fig. S1: Immunohistochemistry for alternative AQP2 in 15 organs of bottlenose dolphin. 1: epidermis, 2: esophagus, 3: forestomach, 4: main stomach, 5: pyloric stomach, 6: small intestine, 7: liver, 8: pancreas, 9: spleen, 10: adrenal gland, 11: ovary, 12: lung, 13: muscle, 14: heart, and 15: brain. Rabbit IgG was used instead of antibodies for a negative control (numbers with dash shows negative controls). Positive reaction was detected in many organs except adrenal grand and brain.

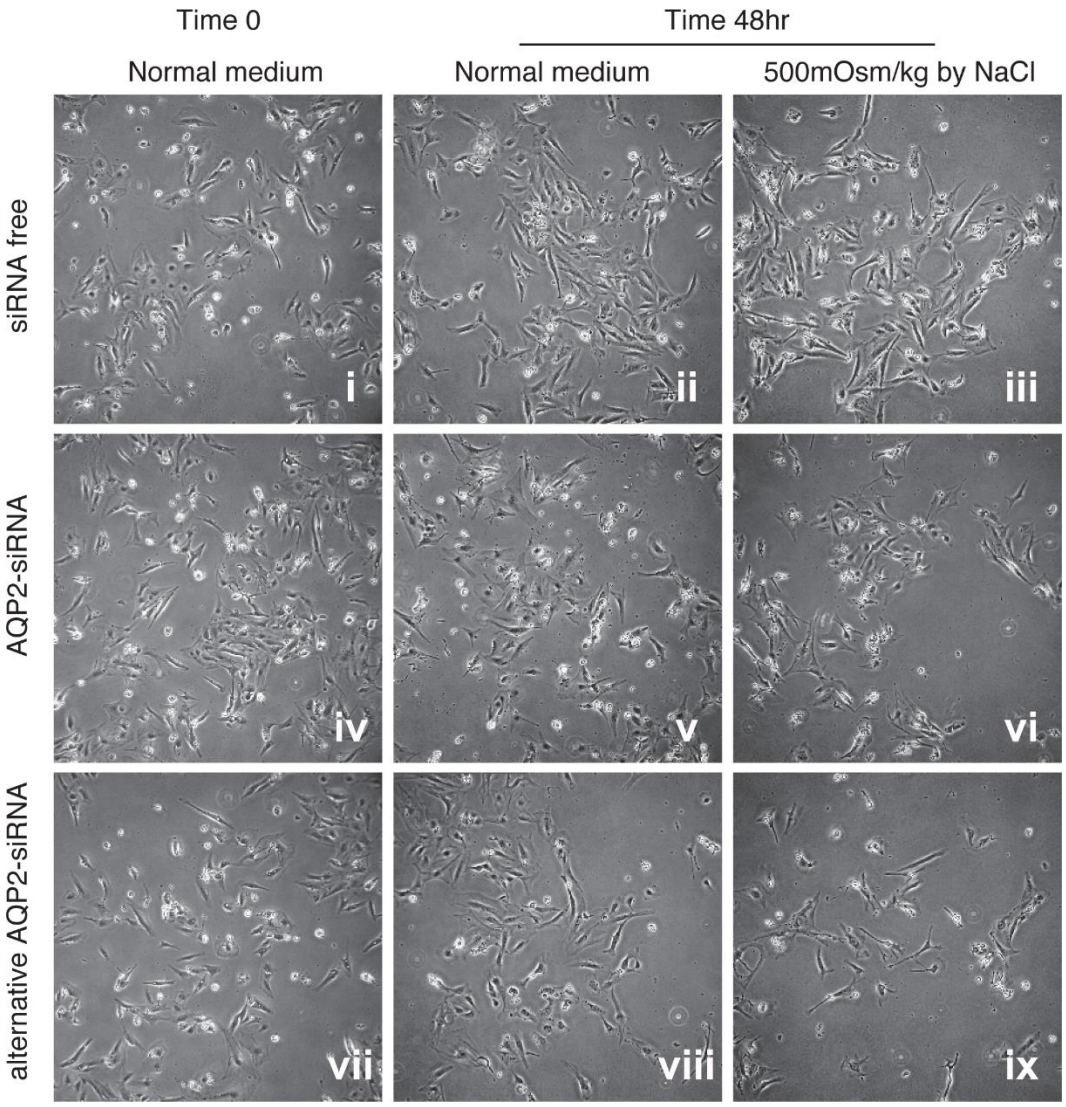

Fig. S2. Effect of siRNA knockdown of AQP2 and alternative AQP2 in cells exposed to hyperosmotic medium (500 mOsm/kg by addition of NaCl). Time 0 indicates the time point just before cell exposure to hyperosmotic medium (i, iv, and vii). Compared with the normal medium (255 mOsm/kg; ii, v, and viii), hyperosmolality caused severe cell shrinkage and death not in siRNA free cells but in cells transfected with AQP2 siRNA (vi) and alternative AQP2 siRNA (ix).

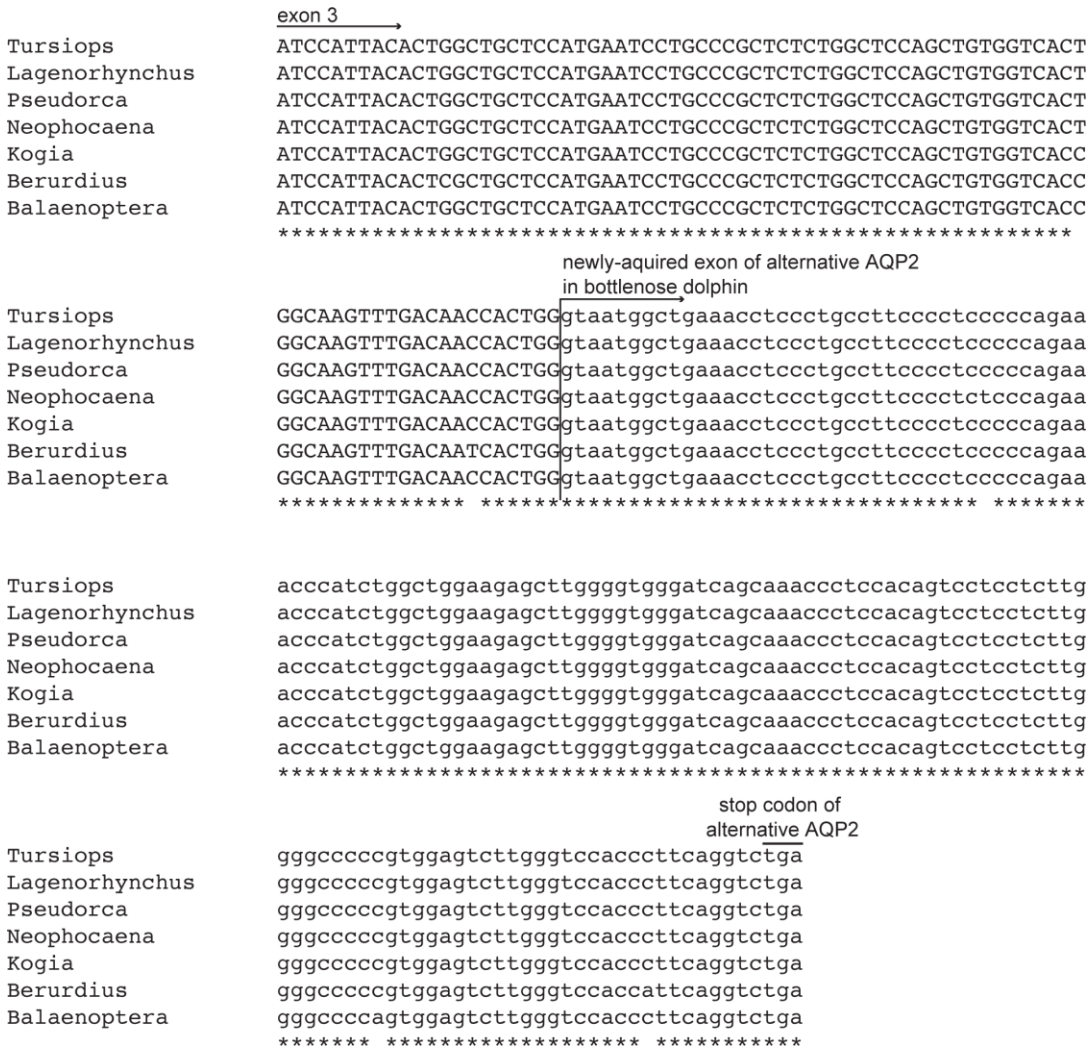

Fig. S3. DNA sequence alignment of the region through exon 3 to the position of the stop codon of alternative AQP2 between AQP2 gene of 7 cetacean species in 5 families. Upper case characters indicate the bases of the original exon 3 of AQP2, and lower case characters indicate the intron of conventional AQP2 that is alternatively spliced to yield an elongated exon in the bottlenose dolphin. Asterisk indicates positions that have a single, fully-conserved base.

Table S1. Primer sequences

Primers	
AQP2 FW1	5'–atgtgggaactccggtccatagccttctc–3'
AQP2 FW2	5'–tggccagcatgtgggaactccggtcc–3'
AQP2 FW3	5'–atctcctcgggatccattac–3'
AQP2 RV1	5'–tcaggccttgctgccccgaggcaggctc–3'
AQP2 RV2	5'–tcctcaaggcaagggatctgccac–3'
AQP2 RV3	5'–aacaggatgtagttgtagag–3'
alternative AQP2 FW	5'–ctagcccacagaagtcttcgaagcatgtg–3'
alternative AQP2 FW2	5'–caacaactcgacagctggccaggccg–3'
alternative AQP2 RV1	5'–ttacgtagtataaacattgccatttatcgag–3'
alternative AQP2 RV2	5'–tccaaagcaaccactgctctt–3'
alternative AQP2 RV3	5'–ttccagccagatgggtttct–3'
AQP2 GRFW	5'–cactgggtcttctgggtcggacccttggtc–3'
AQP2 GRRV	5'–ccttcagcacggccaggcgctccgacag–3'
alternative AQP2 GRFW1	5'–cccagaaacccatctggctggaagagct–3'
alternative AQP2 GRFW2	5'–gtggaatcagcaaaccctccacagtcctc–3'
alternative AQP2 GRRV	5'–agctcttccagccagatgggtttctggg–3'
GAPDH FW	5'–atcaccatcttccaggagcg–3'
GAPDH RV	5'–gcattgctgacgatcttgag–3'
TonEBP FW1	5'–ctatgatcttctcccaaaggagttacag–3'
TonEBP FW2	5'–tgtacaacctgagacccagcaca–3'
TonEBP RV1	5'–cagttaaaaggagccagttaagttgttc–3'
TonEBP RV2	5'–tgctcggatcaaggccaacttc–3'
5' primer	5'–cgactggagcacgaggacactga–3'
3' primer	5'–gctgtcaacgatacgctacgtaacg–3'

Table S2. siRNA sequences

siRNA	
d–AQP2 siRNA (1) sense	5'–cccuccucuacaacuacautt–3'
d–AQP2 siRNA (1) antisense	5'–auguaguuguagaggagggtt–3'
d–AQP2 siRNA (2) sense	5'–gaauggggcaucagguuuutt–3'
d–AQP2 siRNA (2) antisense	5'–aaaaccugaugccccauuctt–3'
d–AQP2 siRNA (3) sense	5'–caauggcuggcccccuuuutt–3'
d–AQP2 siRNA (3) antisense	5'–aaaaggggccagccauugtt–3'
d–alternative AQP2 siRNA (1) sense	5'–cacuggguaauggcugaaatt–3'
d–alternative AQP2 siRNA (1) antisense	5'–uuucagccauuacccagugtt–3'
d–alternative AQP2 siRNA (2) sense	5'–caucuggcuggaagagcuutt–3'
d–alternative AQP2 siRNA (2) antisense	5'–aagcucuuccagccagaugtt–3'
d–alternative AQP2 siRNA (3) sense	5'–gugguugcuuuggcuuuuutt–3'
d–alternative AQP2 siRNA (3) antisense	5'–aaaaagccaaagcaaccactt–3'
d–GAPDH siRNA (1) sense	5'–gaacggauuuggccguauutt–3'
d–GAPDH siRNA (1) antisense	5'–aauacggccaaauccguuctt–3'
d–GAPDH siRNA (2) sense	5'–cacccacggcaaguuccautt–3'
d–GAPDH siRNA (2) antisense	5'–auggaacuugccgugggugtt–3'
d–GAPDH siRNA (3) sense	5'–gccuccugcaccaccaacutt–3'
d–GAPDH siRNA (3) antisense	5'–aguugguggugcaggaggctt–3'