

BcAMT1;5* Mediates Nitrogen Uptake and Assimilation in Flowering Chinese Cabbage and Improves Plant Growth When Overexpressed in *Arabidopsis

Table S1. The primers were used in the study

| Primer name | Sequence (5'–3') | Usage |
|-------------------------|---|--|
| BcAMT1;5 | F: GCAGGTCGACTCTAGAAATGTCTGGAGCTATAACATGC R: ACGAGCTCGGTACCCGGGTCAAAGAGCTGGAGGATCAG | Cloning <i>BcAMT1;5</i> into pCAMBIA3301 vector using <i>Xba</i> I and <i>Sma</i> I sites |
| pYES-AMT1;5 | F: CAGTGTGCTGGAATTCATGTCTGGAGCTATAACATGC R: ATGCGGCCCTCTAGATCAAAGAGCTGGAGGATCAG | Cloning <i>BcAMT1;5</i> into pYES2 vector using <i>EcoR</i> I and <i>Xba</i> I sites |
| pBI-AMT1;5 | F: CACGGGGGACTCTAGAATGTCTGGAGCTATAACATGC R: TCCTTTACCCATCCCGGAAGAGCTGGAGGATCAG | Cloning <i>BcAMT1;5</i> into pBI121 vector using <i>Xba</i> I and <i>Sma</i> I sites |
| BcAMT1;5 _{pro} | F: CCGGCGCGCCAAGCTTTGTCAATGTCCACAATTCC R: GGGATCCGTCGACCTGCAGTTGAGAGACCTTAGAAGGAAAGC | Cloning <i>BcAMT1;5_{pro}</i> into pCAMBIA1391 vector using <i>Hind</i> III and <i>Pst</i> I sites |
| q-BcAMT1;5 | F: AGTGCCACAATGGGAACAC R: GAGCTGGAGGATCAGAACG | qPCR for the cDNA of flowering Chinese cabbage |
| GADPH | F: CAGGTTTGGAAATTGTCGAGG R: GAGCTGTGGAAGCACCTTTC | |
| Actin8 | F: AGGAGCCTCGGTAAGAAGAAC R: TTAGCAACTGGGATGACATGG | |
| q-AtGLN1;1 | F: CAACCTTAACCTCTCAGACTCCACT R: CAGCTGCAACATCAGGGTTGCTA | qPCR for the cDNA of <i>Arabidopsis</i> |
| q-AtGLN1;2 | F: TAACCTTGACATCTCAGACAACAGT R: TCAGCAATAACATCAGGGTTAGCA | |
| q-AtGLN2 | F: CCAACATGTCAGATGAGAGTGCC R: CCAGGTGCTTGACCGGTACTCG | |
| q-AtGDH2 | F: CCAACATGTCAGATGAGAGTGCC R: CCAGGTGCTTGACCGGTACTCG | |
| q-AtGLT1 | F: GGTCTTCCATGGGAAGTGGG R: TAGCAATTCACAGGGCAG | |
| q-ACTIN2 | F: TCGGTGGTTCCATTCTTGCT R: GCTTTTAAGCCTTTGATCTTGAGAG | |

Table S2. The sequence information for each AMT protein used for the phylogenetic analysis.

| Species | Protein name | Accession numbers |
|---|--------------|-------------------|
| Flowering Chinese cabbage (<i>Brassica campestris</i> L. ssp. <i>chinensis</i> var. <i>utilis</i> Tsen et Lee) | BcAMT1;5 | MF966940 |
| <i>Arabidopsis thaliana</i> | AtAMT1;1 | NP_193087.1 |
| | AtAMT1;2 | NP_176658.1 |
| | AtAMT1;3 | NP_189073.1 |
| | AtAMT1;4 | NP_194599.1 |
| | AtAMT1;5 | NP_189072.1 |
| | AtAMT2 | NP_181363 |
| <i>Brassica rapa</i> | BrAMT1;1 | XP_009124614 |
| | BrAMT1;2 | XP_009111404.1 |
| | BrAMT1;3 | XP_009102522.1 |
| | BrAMT1;4 | XP_009129394.1 |
| | BrAMT1;5 | XP_009135885.1 |
| | BrAMT2 | XP_009143404.1 |
| | BrAMT2like | XP_009141731.1 |
| <i>Oryza sativa</i> | OsAMT1;1 | XP_015636241.1 |
| | OsAMT1;2 | XP_015623207.1 |
| | OsAMT1;3 | XP_015624850.1 |
| | OsAMT2;1 | XP_015639562.1 |
| | OsAMT2;2 | XP_015643018.1 |
| | OsAMT2;3 | XP_015621584.1 |
| | OsAMT3;1 | XP_015622013.1 |
| | OsAMT3;2 | XP_015630045.1 |
| | OsAMT3;3 | XP_015626434.1 |
| | OsAMT4;1 | Q10CV4.1 |
| <i>Populus trichocarpa</i> | PtrAMT1;1 | XP_002314518.2 |
| | PtrAMT1;2 | XP_002325790.1 |
| | PtrAMT1;4 | XP_002301837.1 |
| | PtrAMT2 | XP_002309151.1 |
| | PtrAMT2;2 | XP_002323600.2 |
| | PtrAMT3;1 | XP_002324271.3 |
| | PtrAMT4;1 | XM_002302048.1 |

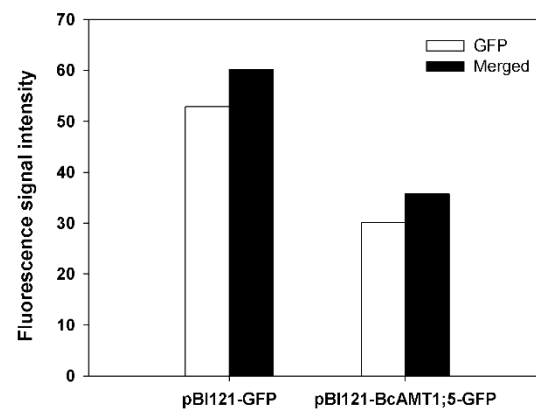


Figure S1. The fluorescence signal intensity of pBI121-GFP and pBI121-BcAMT1;5-GFP in onion epidermal cells.

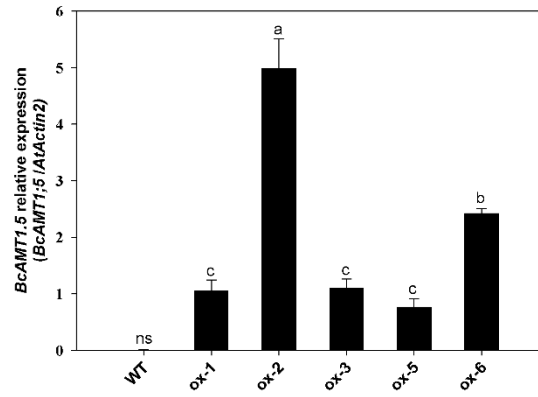


Figure S2. Identification of qPCR in different *BcAMT1;5*-overexpressing lines. WT: wild-type; ox-1, ox-2, ox-3, ox-5, and ox-6 represent overexpressing *BcAMT1;5* lines 1, 2, 3, 5, and 6, respectively. Bars indicate mean \pm SE ($n = 3$), different letters above the bars indicate significant differences at $p < 0.05$.

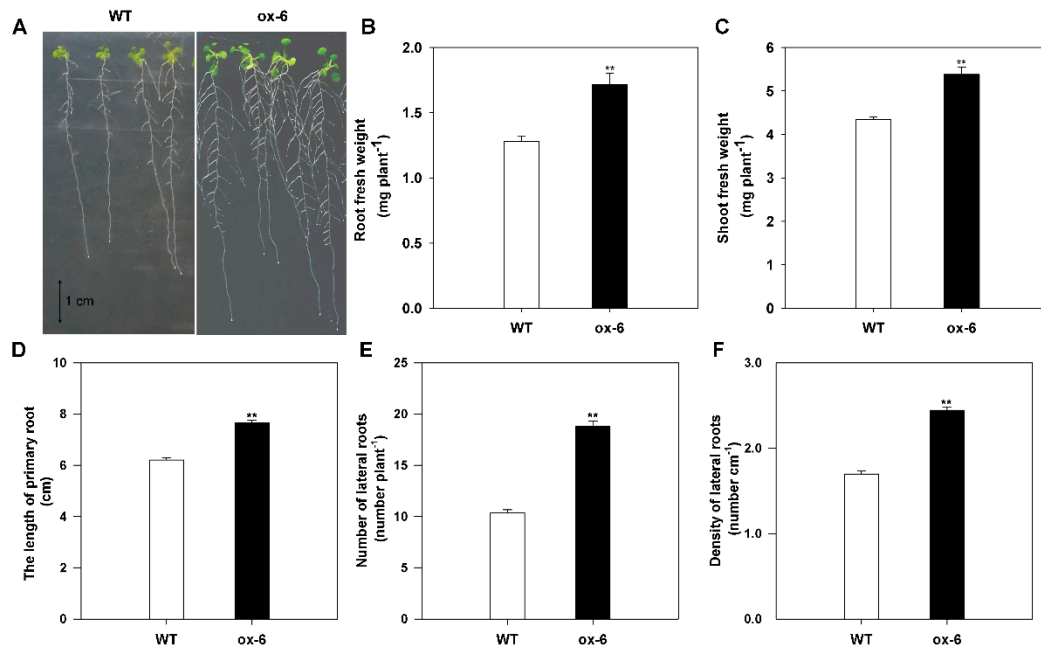


Figure S3. Overexpression *BcAMT1;5* in *Arabidopsis* improves plant growth on the mixture of 0.0625 mmol L⁻¹ NH₄⁺ and 0.1875 mmol L⁻¹ NO₃⁻ as N source. (A) The growth phenotype of T₄ transgenic lines together with wildtype and vector on low NH₄⁺ concentration. Seedlings were grown vertically on solid medium containing the mixture of 0.0625 mmol L⁻¹ NH₄⁺ and 0.1875 mmol L⁻¹ NO₃⁻ for 10 d after a 4 d pre-culture on 4 mmol L⁻¹ NO₃⁻. (B, C) Root and shoot fresh weight of wildtype, and the lines of overexpressing *BcAMT1;5*. (D, E, F) The length of primary root, the number of lateral roots, and density of lateral roots of wildtype in wildtype and the lines of overexpressing *BcAMT1;5*. Data represent mean ± SE (*n*=10), different letters above the bars indicate significant differences at *p* < 0.05, and ** in the histogram shows significant differences at *p* < 0.01.