**Fig.S1** Metabolism overview of the differentially expressed genes in the cob (A) and florets (B) under LN. The red and blue colors indicated up- or down-regulated expression under LN compared to control, respectively.
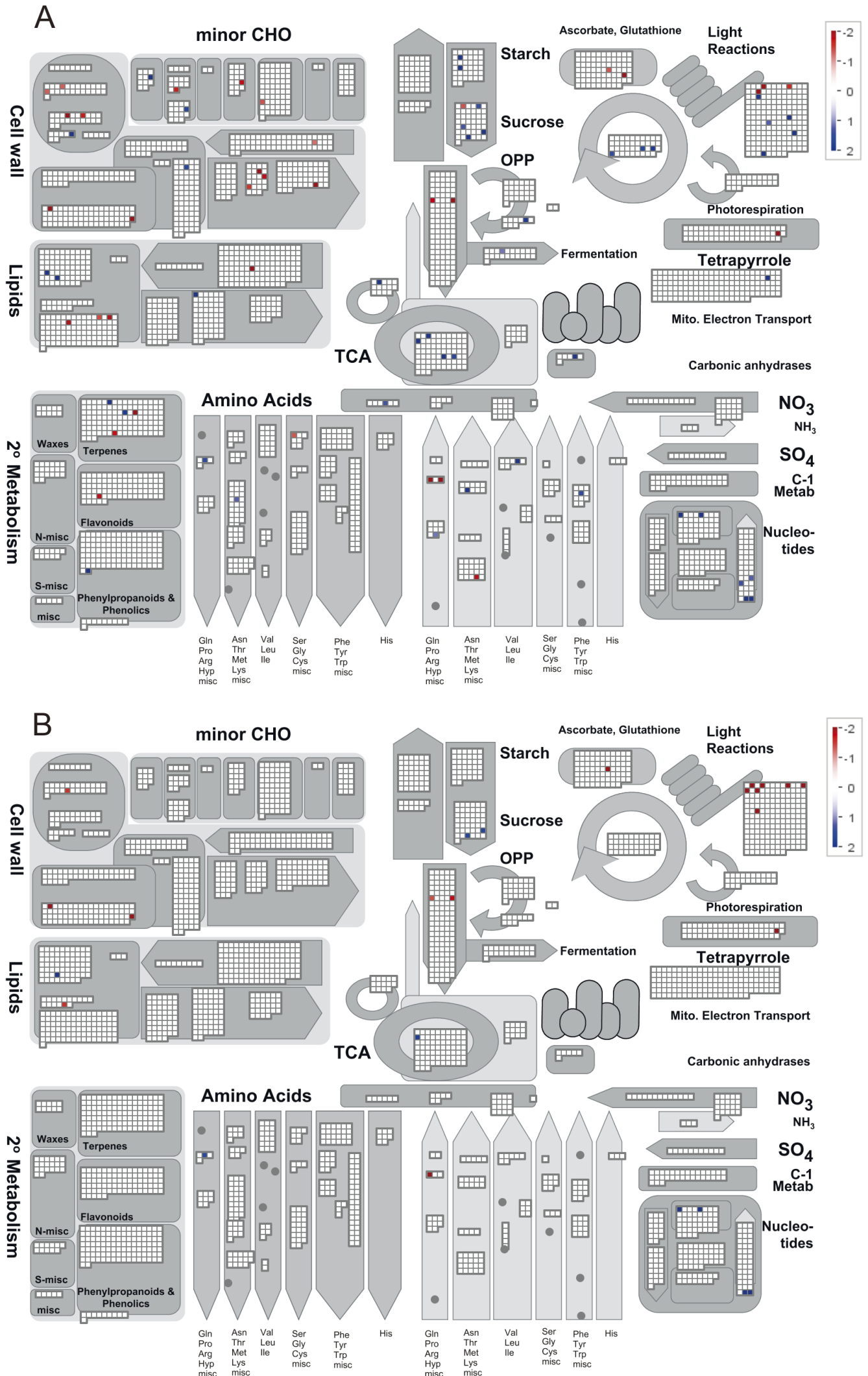
**Fig.S2** Transcription levels of *ZmAAP4* and *ZmVAAT3* in gene overexpression lines, with *AtTub4* as the control (Pfaffl, 2001). #1, #2, and #3 denoted three independent transgenic lines.
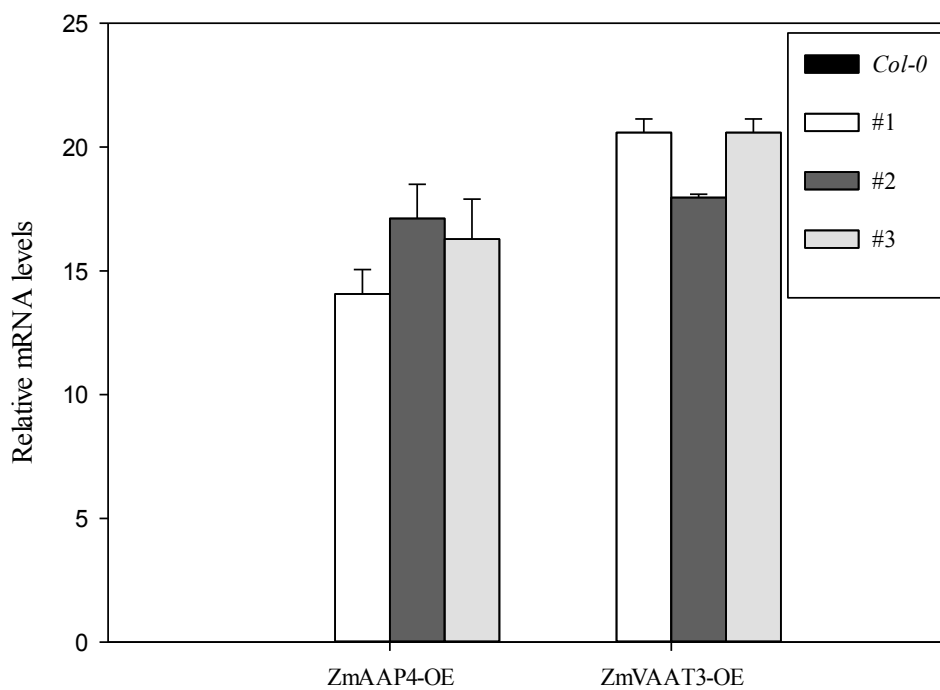
**Table S1.** *Fertilization practice among different nutrient treatments.*

| Treatment | Before sowing | | | V8 | V12 | | Total (kg ha⁻¹) | | |
|---|---|---|---|---|---|---|---|---|---|
| | N | P₂O₅ | K₂O | N | N | K₂O | N | P₂O₅ | K₂O |
| ON | 60 | 135 | 80 | 120 | 70 | 40 | 250 | 135 | 120 |
| LN |  | 135 | 80 |  |  | 40 | 0 | 135 | 120 |
| LP | 60 |  | 80 | 120 | 70 | 40 | 250 |  | 120 |
| LK | 60 | 135 |  | 120 | 70 |  | 250 | 135 |  |

**Table S2.** *Primer sequences used for qRT- PCR.*

| Gene ID | Forward primer (5'-3') | Reverse primer (5'-3') |
| --- | --- | --- |
| GRMZM2G432926 | AACCAGCGAAAGGCGTGGGAGG | AAACTTGGAATGGAAGCCGATGAAA |
| GRMZM2G072322 | GCCACCAGAGTGATGTTTGA | GGGACGAGTAGCCGTTGAGC |
| GRMZM2G400156 | TGCTGTGGATGCTGGAGAAC | AAGAATGCCGAACGAAATGA |
| GRMZM2G062129 | TCCAGGGATACTGAGGATGA | AGGGAGCAGACTTCTATTTACAC |
| GRMZM2G142680 | GTACCATCGTCCATCAATCCC | CGACAACGACATCAACTACCG |
| GRMZM2G001799 | TGGACCAGATCGGCAAGGAATG | TGGCGACACGCACAACTCAAAA |
| GRMZM2G076345 | CGACTACTACGCCTGCTGAACTGC | AAATCAAACCAACGAGCCAAAGC |
| GRMZM2G437575 | TGAAGCCGTTCGTGATTCCAAA | TCGTTCGTCACCACATCCCACA |
| GRMZM2G318780 | GCGGCAGACGGTAGCCAATAA | CAAGGAGATGCTGGGCAACAGAG |
| GRMZM2G311182 | TGCCTGGTCTGTACCGAGTTG | GGAGAAGAGGATGGGCTTTGA |
| GRMZM2G081192 | CTCAGCGACGGCTACTCCAA | ATGAACACGGCGAACCCAAT |
| GRMZM2G051619 | CGTGGAGGGAGTACAAGAA | GATGTGGTGCGACGATAGA |
| GRMZM2G112377 | TCCTTTCTGTCTGGTGCTTGTG | ACGGTGACCTCCGGTTATTTAG |
| GRMZM2G161905 | ACGAGTATAGATGGCAAATGGGTCA | TTGGCAAGGAGACGAACATAAGAAT |

**Table S3.** *Total tags and genes generated by DGE sequencing.*

| | Protein coding | | Pseudogene | | Transposable element | |
|---|---|---|---|---|---|---|
| | Tags | Genes | Tags | Genes | Tags | Genes |
| Sense | 41,751,419 | 32,805 | 236,951 | 1,990 | 468,513 | 1,974 |
| Antisense | 10,589,158 | 29,065 | 828,149 | 1,976 | 226,330 | 1,700 |
| Total | 52,340,577 | 61,870 | 1,065,100 | 3,966 | 694,843 | 3,674 |

**Table S4.** *DGE data verification by qRT- PCR.*

| Gene ID | Fold change (Cob vs Floret) | | Annotation |
|---------|------|---------|------------|
| | **DGE** | **qRT-PCR** | |
| GRMZM2G134888 | 5.0 | 4.5 | Amino acid transporter family protein |
| GRMZM2G162893 | 15.2 | 16.3 | Ethylene-responsive protein -related |
| GRMZM2G413887 | 6.5 | 6.2 | GRAM domain-containing protein / ABA-responsive protein-related |
| GRMZM2G475683 | 8.9 | 8.8 | Auxin-responsive family protein |
| GRMZM2G108537 | 20 | 19.2 | Nodulin MtN21 family protein |
| GRMZM2G179349 | 43.1 | 46.6 | Nodulin MtN3 family protein |
| GRMZM2G045171 | 15.0 | 16.7 | SUS6, ATSUS6 | SUS6 (Sucrose Synthase 6) |
| GRMZM2G085199 | 8.5 | 8.7 | Ethylene-responsive protein -related |
| GRMZM2G078024 | 52.5 | 54.6 | PROT1, ATPROT1 | PROT1 (Proline Transporter 1); L-proline transmembrane transporter |
| GRMZM2G019974 | 38.6 | 41.4 | ATOCT2 (*Arabidopsis Thaliana* Organic Cation/Carnitine Transporter 2) |
| GRMZM2G432926 | 12.7 | 13.8 | PIP2B, PIP2;2 | PIP2B (Plasma Membrane Intrinsic Protein 2); water channel |
| GRMZM2G113167 | 9.3 | 9.1 | GRAM domain-containing protein / ABA-responsive protein-related |
| GRMZM2G175140 | 0.01 | 0.01 | ATAMT1;2, AMT1;2 | ATAMT1;2 (Ammonium Transporter 1;2) |
| GRMZM2G080843 | 0.01 | 0.01 | Amino acid transporter family protein |
| GRMZM2G453565 | 0.1 | 0.1 | AtGH9C3 | AtGH9C3 (Arabidopsis thaliana glycosyl hydrolase 9C3); carbohydrate binding |
| GRMZM2G088861 | 0.01 | 0.03 | Leucine-rich repeat protein, putative |
| GRMZM2G005284 | 0.01 | 0.02 | ARF10 | ARF10 (Auxin Response Factor 10) |
| GRMZM2G326707 | 0.1 | 0.12 | ATPT2, PHT1;4 | ATPT2 (Arabidopsis Thaliana Phosphate Transporter 2); carbohydrate transmembrane transporter |
| GRMZM2G008792 | 0.2 | 0.11 | ATCKX6, CKX6, ATCKX7 | CKX6 (Cytokinin oxidase/dehydrogenase 6) |
| GRMZM2G333980 | 0.2 | 0.18 | Polygalacturonase Inhibiting Protein 1 |

**Table S9.** *Phenotypes of ZmAAP4-OE seedlings 12 days after transfer onto ATS medium supplemented with different amino acids and 3 mM NO$_3^-$*

| Treatment | Primary root length (cm) | | | | Average rosette leaves per plant | | | |
|---|---|---|---|---|---|---|---|---|
| | *Col-0* | ZmAAP4-OE-1 | ZmAAP4-OE-2 | ZmAAP4-OE-3 | *Col-0* | ZmAAP4-OE-1 | ZmAAP4-OE-2 | ZmAAP4-OE-3 |
| 3 mM NO$_3^-$ | 9.78 ± 0.11 | 9.92 ± 0.12 | 9.87 ± 0.21 | 9.83 ± 0.08 | 10.72 ± 0.11 | 10.81 ± 0.13 | 10.91 ± 0.21 | 10.78 ± 0.14 |
| L-Gly (3 mM) | 5.22 ± 0.12 | 4.12 ± 0.10* | 4.32 ± 0.12* | 4.25 ± 0.14* | 7.94 ± 0.24 | 7.07 ± 0.13 * | 7.12 ± 0.17* | 6.94 ± 0.14* |
| L-Ala (3 mM) | 7.50 ± 0.12 | 8.10 ± 0.13 | 8.10 ± 0.12 | 8.28 ± 0.12 | 9.08 ± 0.14 | 7.24 ± 0.17** | 7.43 ± 0.13** | 7.33 ± 0.13** |
| L-Val (300 µM) | 6.71 ± 0.12 | 2.00 ± 0.13** | 2.10 ± 0.11** | 2.12 ± 0.12** | 9.12 ± 0.14 | 7.33 ± 0.17** | 7.25 ± 0.15** | 7.33 ± 0.13** |
| L-Leu (300 µM) | 3.52 ± 0.13 | 2.73 ± 0.12** | 2.63 ± 0.14** | 2.71 ± 0.14** | 9.33 ± 0.11 | 9.12 ± 0.12 | 9.22 ± 0.14 | 9.13 ± 0.15 |
| L-Ile (3 mM) | 3.0 ± 0.12 | 0.9 ± 0.0** | 1.1 ± 0.0** | 0.9 ± 0.0** | 11.11 ± 0.11 | 9.07 ± 0.15** | 9.33 ± 0.17** | 9.42 ± 0.15** |
| L-Ser (300 µM) | 5.13 ± 0.12 | 5.53± 0.11 | 5.61± 0.11 | 5.54± 0.13 | 9.15± 0.13 | 7.22± 0.13** | 7.32± 0.13** | 7.41± 0.13** |
| L-Cys (3 mM) | 6.10 ± 0.12 | 1.20 ± 0.0** | 1.24 ± 0.12** | 1.26 ± 0.0** | 9.24 ± 0.14 | 7.12 ± 0.17** | 7.16 ± 0.17** | 7.33 ± 0.17** |
| L-Thr (300 µM) | 4.82 ± 0.12 | 2.10 ± 0.12** | 2.00 ± 0.10** | 2.10 ± 0.10** | 8.12 ± 0.11 | 7.09 ± 0.13* | 7.23 ± 0.17* | 7.21 ± 0.15* |
| L-Met (300 µM) | 3.50 ± 0.12 | 1.00 ± 0.0** | 1.00 ± 0.0** | 1.00 ± 0.0** | 10.23 ± 0.14 | 11.12 ± 0.17* | 11.21 ± 0.14* | 11.33 ± 0.15* |
| L-Pro (100 µM) | 9.10 ± 0.12 | 8.32 ± 0.13 | 8.40 ± 0.12 | 8.28 ± 0.12 | 8.13 ± 0.11 | 7.33 ± 0.15** | 7.31 ± 0.17** | 7.15 ± 0.13** |
| L-Phe (300 µM) | 6.10 ± 0.12 | 1.72 ± 0.11** | 1.70 ± 0.11** | 1.60 ± 0.10** | 9.31 ± 0.14 | 7.24 ± 0.17** | 7.33 ± 0.13** | 7.13 ± 0.15** |
| L-Tyr (3 mM) | 5.12 ± 0.12 | 5.22 ± 0.15 | 5.23± 0.21 | 5.33 ± 0.15 | 6.12 ± 0.22 | 6.17 ± 0.12 | 6.24 ± 0.24 | 6.31 ± 0.22 |
| L-Trp (3 mM) | 2.32 ± 0.12 | 2.13 ± 0.12 | 1.94 ± 0.12 | 2.12 ± 0.12 | 6.11 ± 0.23 | 4.21 ± 0.22** | 4.05 ± 0.21** | 4.13 ± 0.16** |
| L-His (300 µM) | 4.22 ± 0.12 | 2.50 ± 0.12** | 2.57 ± 0.12** | 2.60 ± 0.10** | 9.12 ± 0.11 | 7.16 ± 0.15** | 7.32 ± 0.14** | 7.08 ± 0.14** |
| L-Lys (300 µM) | 2.30 ± 0.11 | 1.20 ± 0.08* | 1.23 ± 0.12* | 1.21 ± 0.21* | 8.33 ± 0.14 | 7.80 ± 0.17** | 7.92 ± 0.17** | 7.92 ± 0.15** |
| L-Arg (300 µM) | 7.21 ± 0.12 | 1.42 ± 0.14** | 1.52 ± 0.13** | 1.42 ± 0.12** | 9. 33 ± 0.14 | 7. 22 ± 0.14** | 7. 12 ± 0.14** | 7. 32 ± 0.14** |
| L-Asp (3 mM) | 7.10 ± 0.12 | 7.10 ± 0.10 | 7.12 ± 0.10 | 7.12 ± 0.13 | 6.13 ± 0.16 | 6.15 ± 0.132 | 6.25 ± 0.21 | 6.35 ± 0.22 |
| L-Glu (3 mM) | 12.10 ± 0.13 | 10.5 ± 0.10** | 10.61 ± 0.10** | 10.58 ± 0.13** | 8.24 ± 0.11 | 9.08 ± 0.12* | 9.16 ± 0.17* | 9.33 ± 0.15* |
| L-Asn (300 µM) | 8.40 ± 0.12 | 9.00 ± 0.13 | 9.10 ± 0.10 | 9.12 ± 0.13 | 9.24 ± 0.14 | 10.10 ± 0.17* | 10.12 ± 0.17* | 10. 00 ± 0.17* |
| L-Gln (3 mM) | 12.06 ± 0.12 | 10.00 ± 0.13** | 10.10 ± 0.12** | 10.08 ± 0.10** | 8.11 ± 0.12 | 7.12 ± 0.22* | 7.25 ± 0.12* | 7.33 ± 0.12* |
| GABA (3 mM) | 7.20 ± 0.12 | 6.7 0± 0.12 | 6.60 ± 0.10 | 6.70 ± 0.10 | 9.12 ± 0.14 | 7.12 ± 0.12** | 7.25 ± 0.00** | 7.08 ± 0.0-0** |
| L-Cit (100 µM) | 9.21 ± 0.21 | 6.32 ± 0.12** | 6.41 ± 0.15** | 6.62 ± 0.20** | 9.24 ± 0.08 | 9.15 ± 0.10 | 9.08 ± 0.12 | 9.10 ± 0.22 |

| Treatment | Rosette diameter (cm) | | | | Fresh weight per plant (g) | | | |
|---|---|---|---|---|---|---|---|---|
| | *Col-0* | ZmAAP4-OE-1 | ZmAAP4-OE-2 | ZmAAP4-OE-3 | *Col-0* | ZmAAP4-OE-1 | ZmAAP4-OE-2 | ZmAAP4-OE-3 |
| 3 mM NO$_3^-$ | 2.3 ± 0.1 | 2.2 ± 0.1 | 2.3 ± 0.1 | 2.3 ± 0.1 | 0.0320 ± 0.001 | 0.0335 ± 0.0010 | 0.0348 ± 0.0010 | 0.0332 ± 0.0010 |
| L-Gly (3 mM) | 1.5± 0.1 | 0.9± 0.1* | 0.9± 0.1* | 0.9± 0.1* | 0.0120 ± 0.003 | 0.0067 ± 0.0003** | 0.0064 ± 0.0002** | 0.0063 ± 0.0003** |
| L-Ala (3 mM) | 2.1 ± 0.1 | 2.7 ± 0.1** | 2.6 ± 0.1** | 2.7 ± 0.1** | 0.0280 ± 0.001 | 0.0345 ± 0.0010** | 0.0348 ± 0.0010** | 0.0342 ± 0.0010** |
| L-Val (300 μM) | 1.6 ± 0.1 | 0.8 ± 0.0** | 0.8 ± 0.0** | 0.8 ± 0.0** | 0.0253 ± 0.001 | 0.0190 ± 0.001** | 0.0194 ± 0.001** | 0.0189 ± 0.001** |
| L-Leu (300 μM) | 2.2± 0.1 | 2.2± 0.1 | 2.2± 0.1 | 2.2± 0.1 | 0.0280 ± 0.001 | 0.0367± 0.002** | 0.0353± 0.002** | 0.0345± 0.001** |
| L-Ile (3 mM) | 2.3 ± 0.1 | 0.8 ± 0.0** | 0.8 ± 0.0** | 0.8 ± 0.0** | 0.0250 ± 0.001 | 0.0073 ±0.0003*** | 0.0070 ± 0.0004*** | 0.0075 ±0.0003*** |
| L-Ser (300 μM) | 2.2 ± 0.1 | 1.8 ± 0.1** | 1.7 ± 0.1** | 1.6 ± 0.1** | 0.0300 ± 0.001 | 0.0257 ± 0.001** | 0.0244± 0.001** | 0.0252± 0.001** |
| L-Cys (3 mM) | 2.0 ± 0.1 | 1.0 ± 0.0** | 1.0 ± 0.0** | 0.9 ± 0.0** | 0.0340 ± 0.001 | 0.0107 ± 0.002** | 0.0112 ± 0.001** | 0.0102 ± 0.002** |
| L-Thr (300 μM) | 1.0 ± 0.1 | 0.9± 0.0 | 0.9± 0.0 | 0.9± 0.0 | 0.0233 ± 0.001 | 0.0100 ± 0.001 ** | 0.0100 ± 0.001 ** | 0.0100 ± 0.001 ** |
| L-Met (300 μM) | 2.7 ± 0.1 | 1.6 ± 0.1** | 1.5 ± 0.1** | 1.6 ± 0.1** | 0.0267 ± 0.001 | 0.0143 ± 0.001 ** | 0.0142 ± 0.001 ** | 0.0146 ± 0.001** |
| L-Pro (100 μM) | 1.6 ± 0.1 | 1.6 ± 0.1 | 1.6 ± 0.1 | 1.5 ± 0.1 | 0.029 ± 0.001 | 0.020 ± 0.002 ** | 0.021 ± 0.001** | 0.022 ± 0.001 ** |
| L-Phe (300 μM) | 1.7 ± 0.1 | 1.2 ± 0.1* | 1.1 ± 0.1* | 1.2 ± 0.1* | 0.0213 ± 0.001 | 0.0100 ± 0.001** | 0.0120 ± 0.001** | 0.0110 ± 0.001** |
| L-Tyr (3 mM) | 1.6 ± 0.1 | 1.2± 0.1* | 1.2± 0.1* | 1.2± 0.1* | 0.0067± 0.0002 | 0.0020 ± 0.0001** | 0.0020 ± 0.0001** | 0.0020 ± 0.0001** |
| L-Trp (3 mM) | 0.8± 0.0 | 0.6 ± 0.0* | 0.6 ± 0.0* | 0.6 ± 0.0* | 0.0067± 0.0002 | 0.0020 ± 0.0001** | 0.0020 ± 0.0001** | 0.0020 ± 0.0001** |
| L-His (300 μM) | 1.7 ± 0.1 | 1.5 ± 0.1 | 1.5 ± 0.1 | 1.5 ± 0.1 | 0.0177 ± 0.001 | 0.0043 ±0.0003** | 0.0045 ±0.0003** | 0.0042 ±0.0003** |
| L-Lys (300 μM) | 1.4 ± 0.0 | 0.5 ± 0.0** | 0.5 ± 0.0** | 0.5 ± 0.0** | 0.0237± 0.0004 | 0.0057 ±0.0003** | 0.0059 ±0.0004** | 0.0060 ±0.0004** |
| L-Arg (300 μM) | 2.2 ± 0.1 | 0.8 ± 0.0** | 0.8 ± 0.0** | 0.8 ± 0.0** | 0.0333 ± 0.0004 | 0.0107 ± 0.0003** | 0.0114 ± 0.0004** | 0.0118 ± 0.0004** |
| L-Asp (3 mM) | 2.0 ± 0.1 | 2.0 ± 0.1 | 2.0 ± 0.1 | 1.9 ± 0.1 | 0.021 ± 0.001 | 0.022 ± 0.001 | 0.023 ± 0.001 | 0.023 ± 0.001 |
| L-Glu (3 mM) | 3.1 ± 0.1 | 2.5 ± 0.1** | 2.4 ± 0.1** | 2.5 ± 0.1** | 0.03533 ± 0.001 | 0.0233 ± 0.001** | 0.0244 ± 0.001** | 0.0221 ± 0.001** |
| L-Asn (300 μM) | 1.6 ± 0.1 | 1.6 ± 0.0 | 1.6 ± 0.0 | 1.6 ± 0.0 | 0.0183 ± 0.001 | 0.0187 ± 0.001 | 0.0185 ± 0.001 | 0.0183 ± 0.001 |
| L-Gln (3 mM) | 1.5 ± 0.1 | 1.5 ± 0.1 | 1.5 ± 0.1 | 1.5 ± 0.0 | 0.0086 ± 0.0002 | 0.0090 ± 0.0002 | 0.0092 ± 0.0001 | 0.0090 ± 0.0002 |
| GABA (3 mM) | 1.5± 0.0 | 1.1 ± 0.0** | 1.1 ± 0.0** | 1.0 ± 0.0** | 0.0123 ± 0.001 | 0.0080 ±0.0002* | 0.0080±0.0003* | 0.0080 ±0.0003* |
| L-Cit (100 μM) | 1.6 ± 0.1 | 1.4 ± 0.1 | 1.5 ± 0.1 | 1.4 ± 0.1 | 0.028 ± 0.001** | 0.023 ± 0.001** | 0.023 ±0.001** | 0.024 ± 0.002** |

Values are means ± SD; n = 12 individuals. Asterisks indicate significant differences as follows: * P < 0.05, ** P < 0.01

**Table S10.** *Phenotypes of ZmVAAT3-OE seedlings 12 days after transfer onto ATS medium supplemented with different amino acids and 3 mM NO$_3^-$*

| Treatment | Primary root length (cm) | | | | Average rosette leaves per plant | | | |
|---|---|---|---|---|---|---|---|---|
| | *Col-0* | ZmVAAT3-OE-1 | ZmVAAT3-OE-2 | ZmVAAT3-OE-3 | *Col-0* | ZmVAAT3-OE-1 | ZmVAAT3-OE-2 | ZmVAAT3-OE-3 |
| 3 mM NO$_3^-$ | 10.12 ± 0.12 | 10.21 ± 0.13 | 10.22 ± 0.12 | 10.13 ± 0.12 | 8.42 ± 0.14 | 8.49 ± 0.22 | 8.33 ± 0.12 | 8.52 ± 0.08 |
| L-Gly (3 mM) | 5.11 ± 0.21 | 5.0 ± 0.22 | 5.12 ± 0.21 | 5.11 ± 0.22 | 8.22 ± 0.24 | 7.11 ± 0.13 * | 7.34 ± 0.17* | 7.23 ± 0.14* |
| L-Ala (3 mM) | 8.13 ± 0.11 | 13.22 ± 0.21** | 13.11 ± 0.2** | 13.0 1± 0.31** | 8 .33 ± 0.14 | 7.36 ± 0.12* | 7 .24 ± 0.16* | 7.33 ± 0.21* |
| L-Val (300 µM) | 3.51 ± 0.22 | 5.12 ± 0.21* | 5.21 ± 0.12* | 5.2 2± 0.22* | 8.11 ± 0.12 | 7 .22 ± 0.22* | 7.33 ± 0.14* | 7.14 ± 0.15* |
| L-Leu (300 µM) | 3.32 ± 0.21 | 10.52 ± 0.23** | 10.53 ± 0.23** | 10. 43 ± 0.24** | 8.23 ± 0.12 | 8.31 ± 0.13 | 8.25 ± 0.17 | 8.35 ± 0.13 |
| L-Ile (3 mM) | 3.71 ± 0.21 | 3.61 ± 0.12 | 3.61 ± 0.31 | 3.51 ± 0.22 | 7.52 ± 0.22 | 5.34 ± 0.23** | 5.32 ± 0.24** | 5.31 ± 0.14** |
| L-Ser (300 µM) | 5.81 ± 0.21 | 3.52 ± 0.12** | 3.42 ± 0.16** | 3.51 ± 0.21** | 8.41 ± 0.14 | 6.32 ± 0.22** | 6.53 ± 0.24** | 6.42± 0.14** |
| L-Cys (3 mM) | 6.22 ± 0.22 | 5.51 ± 0.23* | 5.42 ± 0.24* | 5.52 ± 0.16* | 8.21 ± 0.11 | 7.24 ± 0.12* | 7.33 ± 0.15* | 7.50 ± 0.19* |
| L-Thr (300 µM) | 4.22 ± 0.12 | 3.53 ± 0.13* | 3.52 ± 0.12* | 3.43 ± 0.12* | 6.32 ± 0.22 | 5.25 ± 0.14** | 5.33 ± 0.12** | 5.25± 0.17** |
| L-Met (300 µM) | 3.81 ± 0.21 | 9.53 ± 0.22** | 9.41 ± 0.22** | 9.52 ± 0.13** | 9.21 ± 0.21 | 7.33 ± 0.14* | 7.41 ± 0.13* | 7.34 ± 0.12* |
| L-Pro (100 µM) | 10.62 ± 0.22 | 10.71 ± 0.21 | 10.61 ± 0.23 | 10.72 ± 0.22 | 10.52 ± 0.22 | 8.23 ± 0.12 | 8.32 ± 0.23 | 8.18 ± 0.13 |
| L-Phe (300 µM) | 8.21 ± 0.22 | 11.12 ± 0.22** | 11.32 ± 0.21** | 11.41 ± 0.22** | 8.62 ± 0.21 | 9.73 ± 0.23* | 9.81 ± 0.17* | 9.64 ± 0.12* |
| L-Tyr (3 mM) | 5.32 ± 0.14 | 5.32 ± 0.13 | 5.43± 0.23 | 5.43 ± 0.23 | 6.24 ± 0.22 | 6.32 ± 0.12 | 6.34 ± 0.24 | 6.61 ± 0.22 |
| L-Trp (3 mM) | 2.42 ± 0.12 | 2.03 ± 0.13 | 1.92 ± 0.13 | 2.02 ± 0.13 | 6.51 ± 0.23 | 4.43 ± 0.22** | 4.33 ± 0.21** | 4.25 ± 0.16** |
| L-His (300 µM) | 3.00 ± 0.10 | 2.51 ± 0.22* | 2.62 ± 0.12* | 2.42 ± 0.13* | 8.53 ± 0.21 | 6.32 ± 0.22** | 6.38 ± 0.13** | 6.51 ± 0.17** |
| L-Lys (300 µM) | 2.10 ± 0.13 | 2.03 ± 0.12 | 2.03 ± 0.12 | 1.90 ± 0.12 | 7.23 ± 0.12 | 5.32 ± 0.23** | 5.37 ± 0.22** | 5.38 ± 0.12** |
| L-Arg (300 µM) | 8.31 ± 0.21 | 8.82 ± 0.12 | 8.9 2± 0.12 | 8.82 ± 0.12 | 8.33 ± 0.12 | 8.24 ± 0.21 | 8.12 ± 0.11 | 8.37 ± 0.13 |
| L-Asp (3 mM) | 7.72 ± 0.12 | 7.62 ± 0.23 | 7.73 ± 0.23 | 7.71 ± 0.13 | 6.33 ± 0.16 | 6.25 ± 0.12 | 6.32 ± 0.21 | 6.33 ± 0.22 |
| L-Glu (3 mM) | 8.52 ± 0.18 | 8.62 ± 0.25 | 8.72 ± 0.25 | 8.64 ± 0.25 | 9.25 ± 0.22 | 7.09 ± 0.12** | 7.33 ± 0.12** | 7.42 ± 0.15** |
| L-Asn (300 µM) | 8.62 ± 0.22 | 11.56 ± 0.23** | 11.65 ± 0.34** | 11.44 ± 0.24** | 8.23 ± 0.16 | 8.12± 0.19 | 8.09± 0.21 | 8.0± 0.13 |
| L-Gln (3 mM) | 13.12 ± 0.21 | 11.61 ± 0.22* | 11.52 ± 0.21* | 11.51 ± 0.23* | 7.22 ± 0.12 | 8.32 ± 0.22* | 8 .12 ± 0.12* | 8.22 ± 0.12* |
| GABA (3 mM) | 9.12 ± 0.12 | 7.24 ± 0.11** | 7.13 ± 0.11** | 7.32 ± 0.12** | 8 .25 ± 0.15 | 8.25 ± 0.21 | 8.33 ± 0.13 | 8.66 ± 0.12 |
| L-Cit (100 µM) | 9.22 ± 0.22 | 6.61 ± 0.21** | 6.57 ± 0.22** | 6.73 ± 0.22** | 8.24 ± 0.13 | 6.23 ± 0.12** | 6.27 ± 0.13** | 6.21 ± 0.17** |

| Treatment | Rosette diameter (cm) | | | | Fresh weight per plant (g) | | | |
|---|---|---|---|---|---|---|---|---|
| | *Col-0* | ZmVAAT3-OE -1 | ZmVAAT3-OE -2 | ZmVAAT3-OE -3 | *Col-0* | ZmVAAT3-OE -1 | ZmVAAT3-OE -2 | ZmVAAT3-OE -3 |
| 3 mM NO$_3^-$ | 1.7 ±0.00 | 1.7 ±0.00 | 1.7 ±0.00 | 1.7 ±0.00 | 0.0213 ± 0.001 | 0.0227 ± 0.001 | 0.0210 ± 0.001 | 0.0228 ± 0.002 |
| L-Gly (3 mM) | 1.4 ± 0.1 | 1.1 ±0.0** | 1.1 ±0.0** | 1.1 ±0.0** | 0.0100 ± 0.003 | 0.0097 ± 0.0003 | 0.0094 ± 0.0002 | 0.0093 ± 0.0003 |
| L-Ala (3 mM) | 1.2 ± 0.10 | 1.6 ± 0.10** | 1.6 ± 0.10** | 1.7 ± 0.10** | 0.0210 ± 0.002 | 0.0223 ± 0.003 | 0.0225 ± 0.003 | 0.0224± 0.004 |
| L-Val (300 µM) | 1.8 ± 0.1 | 1.5 ± 0.0** | 1.5 ± 0.0** | 1.5 ± 0.0** | 0.0153 ± 0.002 | 0.0155 ± 0.002 | 0.0152 ±0.003 | 0.0155 ± 0.003 |
| L-Leu (300 µM) | 1.6 ± 0.1 | 1.5 ± 0.10 | 1.5 ± 0.10 | 1.5 ± 0.10 | 0.0253 ± 0.001 | 0.0257 ± 0.001 | 0.0240 ± 0.001 | 0.0248 ± 0.002 |
| L-Ile (3 mM) | 1.6 ± 0.20 | 0.5 ± 0.00** | 0.5 ± 0.00** | 0.5 ± 0.00** | 0.0220 ± 0.003 | 0.004 ± 0.001** | 0.004 ± 0.001** | 0.004 ± 0.001** |
| L-Ser (300 µM) | 1.5 ± 0.1 | 1.1 ± 0.1** | 1.1 ± 0.0** | 1.1 ± 0.1** | 0.0127 ± 0.0004 | 0.0057 ± 0.0003** | 0.0056 ±0.0002** | 0.0053 ± 0.0003** |
| L-Cys (3 mM) | 1.8 ± 0.10 | 1.9 ± 0.10 | 1.9 ± 0.10 | 2.0 ± 0.10 | 0.0303 ± 0.0003 | 0.0190 ± 0.0002** | 0.0188 ± 0.0002** | 0.0190 ± 0.0002** |
| L-Thr (300 µM) | 1.5 ± 0.10 | 1.4 ± 0.10 | 1.4 ± 0.10 | 1.4 ± 0.10 | 0.0173 ± 0.0001 | 0.0157 ± 0.0001 | 0.0156 ± 0.0002 | 0.0157 ± 0.0001 |
| L-Met (300 µM) | 1.5 ± 0.1 | 1.3 ± 0.1 | 1.3 ± 0.10 | 1.3 ± 0.10 | 0.0103 ± 0.0003 | 0.0173 ± 0.0002** | 0.0162 ± 0.0003** | 0.0170 ± 0.0003** |
| L-Pro (100 µM) | 1.8 ± 0.2 | 1.9 ± 0.1 | 1.9 ± 0.1 | 1.8 ± 0.1 | 0.018 ± 0.001 | 0.027 ± 0.001** | 0.025 ± 0.002** | 0.027 ± 0.001** |
| L-Phe (300 µM) | 1.3 ± 0.10 | 1.3 ± 0.0 | 1.3 ± 0.10 | 1.2 ± 0.10 | 0.0123 ± 0.001 | 0.0163 ±0.001* | 0.0162 ±0.001* | 0.0161 ±0.001* |
| L-Try (3 mM) | 2.1 ± 0.10 | 1.8 ± 0.10 | 1.8 ± 0.20 | 1.8 ± 0.10 | 0.0117 ±0.0003 | 0.0114 ±0.0003 | 0.0115 ±0.0003 | 0.0111 ±0.0003 |
| L-Trp (3 mM) | 0.7 ± 0.00 | 0.3 ± 0.00** | 0.3 ± 0.00** | 0.3 ± 0.00** | 0.0067± 0.0002 | 0.0020 ± 0.0001** | 0.0020 ± 0.0001** | 0.0020 ± 0.0001** |
| L-His (300 µM) | 1.1 ± 0.1 | 0.8 ± 0.0* | 0.8 ± 0.0* | 0.8 ± 0.0* | 0.0111 ± 0.001 | 0.0053 ± 0.0002** | 0.0050 ± 0.0002** | 0.0058 ± 0.0001** |
| L-Lys (300 µM) | 1.0 ± 0.10 | 0.7 ± 0.00** | 0.7 ± 0.00** | 0.7 ± 0.00** | 0.029 ± 0.001** | 0.006 ± 0.0** | 0.006 ± 0.0** | 0.006 ± 0.0** |
| L-Arg (300 µM) | 2.0 ± 0.10 | 2.0 ± 0.10 | 2.0 ± 0.10 | 1.8 ± 0.10 | 0.0200 ± 0.0002 | 0.0183 ± 0.0002 | 0.0181 ± 0.0002 | 0.0184 ± 0.0002 |
| L-Asp (3 mM) | 1.8 ± 0.10 | 1.6 ± 0.20 | 1.6 ± 0 .10 | 1.6 ± 0.10 | 0.024 ± 0.001 | 0.022 ± 0.001 | 0.023 ± 0.001 | 0.023 ± 0.001 |
| L-Glu (3 mM) | 2.3 ± 0.20 | 1.8 ± 0.1** | 1.8 ± 0.10** | 1.8 ± 0.10** | 0.0277 ± 0.0002 | 0.0190 ± 0.0002** | 0.0182 ± 0.0002** | 0.0193 ± 0.0002** |
| L-Asn (300 µM) | 1.4 ± 0.10 | 1.5 ± 0.10 | 1.5 ± 0.10 | 1.5 ± 0.10 | 0.0140 ± 0.0002 | 0.0173 ± 0.0003* | 0.0171 ± 0.0002* | 0.0178 ± 0.0002* |
| L-Gln (3 mM) | 2.0 ± 0.20 | 1.5 ± 0.10** | 1.5 ± 0.10** | 1.5 ± 0.10** | 0.0136 ± 0.0002 | 0.0100 ± 0.0002* | 0.0102 ± 0.0001* | 0.0100 ± 0.0002* |
| GABA (3 mM) | 1.9 ± 0.10 | 1.9 ± 0.20 | 1.8 ± 0.10 | 1.8 ± 0.10 | 0.0170 ± 0.0003 | 0.0130 ± 0.0003* | 0.0130 ± 0.003* | 0.01300 ± 0.0003* |
| L-Cit (100 µM) | 1.4 ± 0.1 | 0.9 ± 0.10** | 0.9 ± 0.00** | 0.9 ± 0.10** | 0.0113 ± 0.0004 | 0.0120 ± 0.0002 | 0.0094 ± 0.0002 | 0.0092 ± 0.0002 |

Values are means ± SD; n = 12 individuals. Asterisks indicate significant differences as follows: * P < 0.05, ** P < 0.01

**Table S11**. *Effects of different amino acids on growth parameters of ZmAAP4-OE and ZmVAAT3-OE arabidopsis seedlings*

| Parameter | Effect | Amino acids | |
| --- | --- | --- | --- |
| | | **ZmAAP4-OE** | **ZmVAAT3-OE** |
| Root length | Positive | - | Leu, Ala, Val, Phe, Met, Arg, Asn, Glu |
| | Negative | Gly, Val, Cys, Thr, Cit, Leu, Ile, Met, Pro, Trp, Phe, His, Lys, Arg, Glu, Gln, GABA | Ser, Cys, Thr, Trp, Lys, Gln, Cit, GABA, His |
| Rosette leaf number | Positive | Met, Tyr, Asp, Glu Asn | Leu, Phe, Tyr, Gln, |
| | Negative | Gly, Ala, Val, Leu, Ile, Ser, Cys, Thr, Pro, Phe, Trp, His, Lys, Arg, Gln, Cit, GABA | Gly, Ala, Val, Ile, Ser, Cys, Thr, Met, Pro, Trp, His, Lys, Arg, Glu, Asn, Cit |
| Rosette diameter | Positive | Ala | Ala |
| | Negative | Gly, Val, Ile, Ser, Cys, Met, Phe, Tyr, Trp, Lys, Arg, Glu, GABA | Gly, Val, Ile, Ser, Tyr, Trp, His, Lys, Glu, Gln, Cit |
| Fresh weight | Positive | Ala, Leu | Met, Pro, Phe, Asn |
| | Negative | Gly, Val, Ile, Ser, Cys, Thr, Met, Pro, Phe, Tyr, Trp, His, Lys, Arg, Glu, GABA, Cit | Ile, Ser, Cys, Trp, His, Lys, Arg, Glu, Gln, GABA |

**Supplementary dataset 1. Coding sequences and functional domains of ZmAAP4 and ZmVAAT3**

Coding sequences of two amino acid transporters (the sequence from the start codon "ATG" to the stop codon "TGA" or "TAG" were subcloned into the overexpression vector for gene overexpression) and illustration of their functional domains (Tusnády and Simon, 1998; 2001).

***ZmAAP4* (GRMZM2G110195_T01)**:
ATGGCGGAGAACAACGTCGTGGCCACGTACTACTACCCGACGGCAGCGCC
GGCGGCCATGGAGGTCTGCGGCGCGGAGCTCGGCCAGGGCAAGCCCGAC
AAGTGCTTCGACGACGATGGCCGCCCCAAGCGCAATGGGACGATGTGGAC
GGCGAGCGCGCACATCATCACGGCGGTGATCGGCTCCGGGGTGCTCTCGC
TGGGGTGGGCCATCGCGCAGCTCGGCTGGGTGGCCGGACCCGTCGTCATG
CTGCTCTTCTCGCTCGTCACCTACTACACCTCGTCGCTGCTCGCAGACTGC
TACCGCTCCGGCGACCCCAGCACCGGCAAGCGGAACTACACCTACATGGA
CGCCGTCAACGCGAACCTCAGTGGCATCAAGGTCCAGATCTGCGGGTTCC
TGCAGTACGCCAACATCGTGGGCGTGGCCATCGGCTACACCATCGCTGCC
TCCATTAGCATGCTCGCGATCAGGAGGGCCAACTGCTTCCACCAGAAGGG
ACACGGCAACCCCTGCAAGATCTCCAGCACGCCCTACATGATCATCTTCG
GCGTGGCGGAGATCTTCTTCTCGCAGATCCCGGACTTCGACCAGATCTCCT
GGCTCTCCATCCTCGCCGCCGTCATGTCCTTCACCTACTCCTCCATTGGGC
TCGGCCTGGGCGTCGTCCAAGTCATCGCGAACAGAGGCGTGCAGGGCAGC
CTGACCGGCATCACCATCGGCGTGGTGACCCCGATGGACAAGGTGTGGCG
CAGCCTCCAGGCGTTCGGCGACGTCGCCTTCGCCTACTCCTACTCCCTCAT
CCTGATCGAGATCCAGGACACCATCCGGGCGCCGCCGCCGTCGGAGTCGA
CGGTGATGAAGCGCGCCACGGTGGTGAGCGTGGCGGTCACCACGCTCTTC
TACATGCTGTGCGGCTGCATGGGGTACGCGGCGTTCGGCGACGGCGCGCC
CGGGAACCTCCTCACGGGCTTCGGCTTCTACGAGCCCTTCTGGCTCCTGGA
CGTGGCCAACGCCGCCATCGTGGTCCACCTGGTCGGCGCCTACCAGGTCT
ACTGCCAGCCGCTGTTCGCCTTCGTGGAGAAGTGGGCCGCGCAGCGGTGG
CCGGACTCGGCGTACATCACCGGGGAGGTCGAGGTCCCGCTCCCGCTCCC
GGCGAGCCGGCGGCGGTGCTGCAAGGTGAACCTGTTCCGGGCGACGTGGC
GGACGGCGTTCGTCGTGGCCACGACGGTCGTGTCCATGCTGCTGCCCTTCT
TCAACGACGTGGTGGGCTTCCTGGGCGCGCTCGGCTTCTGGCCGCTCACC
GTCTACTTCCCCGTCGAGATGTACGTGGTGCAGAAGAAGGTGCCGCGGTG
GAGCTCCCGGTGGGTGTGCCTGCAGATGCTCAGCCTCGGCTGCCTCGTCAT
CTCCATCGCCGCCGCAGCCGGGTCCATCGCCGGCATCGCGTCCGACCTCA
AAGTCTACCGCCCGTTCAAGTCCTACTGA

**Illustration of 12 transmembrane domains of ZmAAP4.** Locations of trans-membrane helices (from left to right): 51-67, 74-90, 139-155, 174-190, 199-223, 230-246, 255-271, 290-309, 336-360, 403-419, 424-440, 459-483.



Extracellular

Cytoplasm
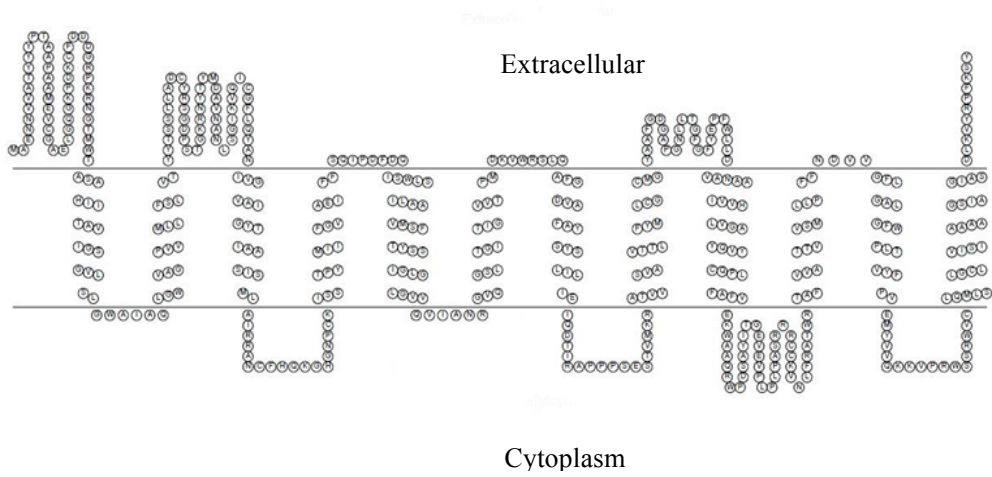
*ZmVAAT3* (GRMZM2G080843_T01):

ATGGCGGCGGCGGAGGAGGAGGAGGAGTGGGAGGATGGGCGTTCGGTGA
TGCCGGAGGAGGAGGACGGAGACATCGAGGACGTCCCACCGCCGCCGCT
GCTAGGCAGGAACTTCGGCAGCGAGGACGAGGACGAGGACGAGGCGGGC
TCGTTGAACTCCCAGTCCCAGTGGTGGCCGCGGAGCTTCCGGGAAGCGAC
TGACACCTATACCATTGCCGCGCCCCGGGCCTCCGTCACCTCGTCGTCGG
TGACTGCAATGGCCTGGGCTCTGACCTGAAGCTGCCTCTTCTGTCGGACAA
GCTAGATGGGAGGCAAGATTCATCCAACAACTTGCTGCCAGTAGCAGACC
CCTTCGGTTCAGTGCTAAGCGACGGGAGATTCTCAGATAGAGTGCAACAA
GCCCCAACCACTCCAGGATGCAATCTGACGCAAACCGTGTTCAATGGGGT
CAACGTGCTTGCTGGCATTGGCATTTTCTCTGCTCCATACACGATTCGTGA
GGCCGGATGGGCCAGCCTTGTGGTTCTGGCTTTTTTCGCTGTGGTGTGCTG
CTACACTGGAGTTCTTCTAAAGTACTGCTTTGAGAGCAAAGATGGTGTCA
AAACTTTCCCAGACATTGGGGAGCTTGCCTTTGGAAGGATCGGCCGGTTT
CTCATTTCTATAGTTCTGTACACCGAGCTCTATTCCTTTTGTGTGGAGTTCG
TTATTCTGGAAGGTGATAACCTGGCATCAATTTTCACGAGCACCACTTTCG
ATTGGAATGGGATTCATGCCGATGGCAGACATTTCTTTGGGATTCTATTTG
CTCTTGTTGTCTTACCCAGCGTGTGGTTGCGAGATCTTCGAGTAATATCTT
ACCTTTCAGTGGGTGGTGTGTTTGCAACTCTTCTGGTTTTCCTATCTGTTGG
TGTAGTTGGTGCTACTGGCAACGTAGGGTTCCACTTGGCAGGGAAAGCAG
TGAAGTGGGATGGTATACCATTTGCAATTGGCATTTATGGTTTTTGCTACG
CTGGACACTCAGTGTTTCCAAATATCTATCAGTCAATGTCCGATCGCACCA
AATTCAACAAGGCACTCTACATATGTTTTGCAATATGCACGACAATTTATG
GCGCTATAGCTGTTATTGGCTACCTCATGTTTGGAGATAAGACTCTCTCAC
AAATAACTTTGAATCTTCCGAAAGATTCATTTGTGGCAAAAGTTGCACTGG
GGACCACGGTGATCATTCCTTTCACAAAATATTCTTTGGTGATAAACCCAT
TGGCTCGGAGTATAGAAGAGTTGCGTCCAGAAGGGTTTCTAACCGATAGA
CTCTTCGCTGTTATGCTGCGTACTGCCATTGTTGCATCAACTCTATGTGTTG
CCTTTCTCCTGCCATTCTTTGGTCTTGTGATGGCTCTCATTGGGTCTCTTCT
CAGTATACTTGTGGCGCTCATAATGCCTGCCCTGTGCTTCCTGAAGATCGC
TCGGAACAAAGCGACGCGCTTGCAGGTCATCGCAAGCGTCGCGACCGTGG
TGCTGGGCTCCGTCTGCGCAGTTCTTGGGACGTACAACTCTATCGCCAAGA
TAGCCGAAAGCTACTAG

**Illustration of 11 transmembrane domains of ZmVAAT3.** Locations of trans-membrane helices (from left to right): 146-165, 170-190, 217-237, 262-278, 287-306, 327-346, 359-381, 402-422, 439-459, 464-487, 500-522.

Extracellular



Cytoplasm
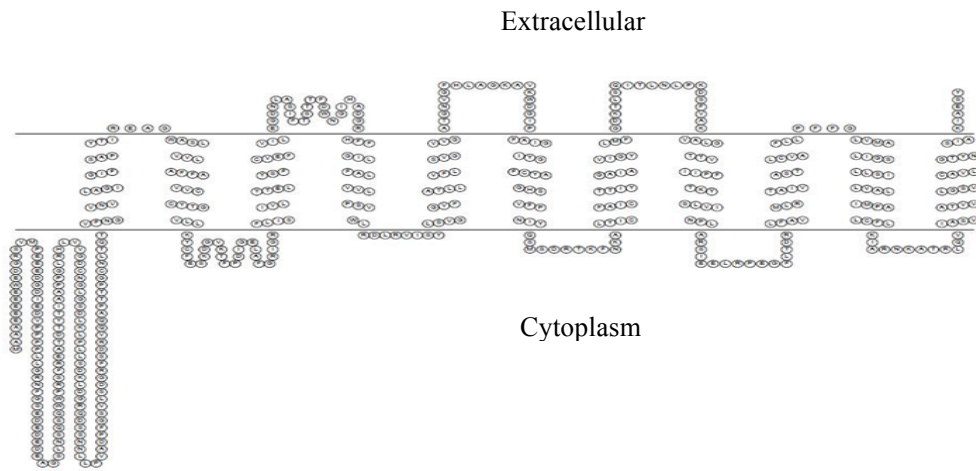
*References:*

**Tusnady GE, Simon I.** 1998. Principles governing amino acid composition of integral membrane proteins: application to topology prediction. *Journal of Molecular Biology* **283**, 489-506.

**Tusnady GE, Simon I.** 2001. The HMMTOP transmembrane topology prediction server. *Bioinformatics* **17**, 849-850.