

**New Phytologist Supporting Information**

Article title: **Broad spectrum developmental role of Brachypodium AUX1**

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**Supporting Information Methods S1 - DNA and oligonucleotide sequences**

Cas9 cassette: **FLAG-tag NLS Bd-optimized Cas9**

TATGGTACCATGATGATCGAC**TACAAGGACGACGACGACAAG**ATGGCCCCG**AAGAAGAAGCGCAAG**GTGGGC**ATGGACAAGAAGTA**  
**CTCCATCGGCCTCGACATCGGCACGAACTCCGTGGGCTGGGCCGTGATCACGGACGAGTACAAGGTGCCGTCCAAGAAGTTCAAGG**  
**TGCTCGGCAACACGGACCGCCACTCCATCAAGAAGAACCTCATCGGGCCCTCCTCTCGACTCCGGCGAGACGGCCGAGGCCACG**  
**CGCCTCAAGCGCACGGCCCGCCCGCTACACGCGCCGAAGAACCGCATCTGCTACCTCCAGGAGATCTTCTCCAACGAGATGGC**  
**CAAGGTGGACGACTCCTTCTCCACCGCCTCGAGGAGTCTTCTCGTGGAGGAGACAAGAAGCACGAGCGCCACCCGATCTTCG**  
**GCAACATCGTGGACGAGGTGGCCTACCACGAGAAGTACCCGACGATCTACCACCTCCGCAAGAAGCTCGTGGACTCCACGGACAAG**  
**GCCGACCTCCGCTCATCTACCTCGCCCTCGCCACATGATCAAGTTCGCGGCCACTTCCTCATCGAGGGCGACCTCAACCCGGA**  
**CAACTCCGACGTGGACAAGCTTTCATCCAGCTCGTGCAGACGTACAACCAGCTCTTCGAGGAGAACCCGATCAACGCCTCCGGCG**  
**TGGACGCCAAGGCCATCCTCTCCGCCCGCCTCTCCAAGTCCCGCCGCTCGAGAACCTCATCGCCAGCTCCCGGGCGAGAAGAAG**  
**AACGGCTCTTCGGCAACCTCATCGCCCTCTCCCTCGGCCTCACGCCGAACCTCAAGTCCAACCTTCGACCTCGCCGAGGACGCCAA**  
**GCTCCAGCTCTCCAAGGACACGTACGACGACGACCTCGACAACCTCCTCGCCAGATCGGGCAGCAGTACGCCGACCTCTTCCTCG**  
**CCGCCAAGAACCTCTCCGACGCCATCCTCCTCCGACATCCTCCGCGTGAACACGGAGATCACGAAGGCCCGCTCTCCGCCTCC**  
**ATGATCAAGCGCTACGACGAGCACCACCAGGACCTCACGCTCCTCAAGGCCCTCGTGCGCCAGCAGCTCCCGGAGAAGTACAAGGA**  
**GATCTTCTTCGACCAAGTCCAAGAACGGCTACGCCGCTACATCGACGGCGGCCCTCCAGGAGGAGTTCTACAAGTTCATCAAGC**  
**CGATCTTCGAGAAGATGGACGGCACGGAGGAGCTCTCGTGAAGTCAACCGCGAGGACCTCCTCCGCAAGCAGCGCAGTTCGAC**  
**AACGGCTCCATCCCGCACCAGATCCACCTCGGCGAGCTCCACGCCATCCTCCGCCGCCAGGAGGACTTCTACCCGTTCTCAAGGA**  
**CAACCGGAGAAGATCGAGAAGATCCTCACGTTCCGCATCCCGTACTACGTGGGCCGCTCGCCCGGGCAACTCCCGCTTCGCCT**  
**GGATGACCGCAAGTCCGAGGAGACGATCACGCCGTGGAACCTTCGAGGAGGTGGTGGACAAGGGCGCCTCCGCCAGTCTTCATC**  
**GAGCGCATGACGAACCTTCGACAAGAACCTCCCGAACGAGAAGGTGCTCCCGAACACTCCCTCCTTACGAGTACTTCACGGTGTG**  
**CAACGAGCTCACGAAGGTGAAGTACGTGACGAGGGCATGCGCAAGCCGGCCTTCTCTCCGGCGAGCAGAAGAAGGCCATCGTGG**  
**ACCTCCTTCAAGACGAACCGCAAGGTGACGGTGAAGCAGCTCAAGGAGGACTACTTCAAGAAGATCGAGTGCTTCGACTCCGTG**  
**GAGATCTCCGGCGTGGAGGACCGCTTCAACGCCCTCCTCGGCAGTACCACGACCTCCTCAAGATCATCAAGGACAAGGACTTCCT**  
**CGACAACGAGGAGAACGAGGACATCCTCGAGGACATCGTGCTCACGCTCACGCTCTTCGAGGACCGGAGATGATCGAGGAGCGCC**  
**TCAAGACGTACGCCACCTCTTCGACGACAAGGTGATGAAGCAGCTCAAGCGCCGCGCTACACGGGCTGGGGCCGCTCTCCCGC**  
**AAGTTCATCAACGGCATCCGCGACAAGCAGTCCGGCAAGACGATCCTCGACTTCTCAAGTCCGACGGCTTCGCCAACCGCAACTT**  
**CATGCAGCTCATCCACGACGACTCCCTCACGTTCAAGGAGGACATCCAGAAGGCCAGGTGTCCGGCCAGGGCGACTCCCTCCACG**  
**AGCACATCGCCAACCTCGCCGGCTCCCGGCCATCAAGAAGGGCATCCTCCAGACGGTGAAGGTGGTGGACGAGCTCGTGAAGGTG**  
**ATGGGCCGCCACAAGCCGGAGAACATCGTGATCGAGATGGCCCGGAGAACCAGACGACGCAGAAGGGCCAGAAGAAGTCCCGCGA**  
**GCGCATGAAGCGCATCGAGGAGGGCATCAAGGAGCTCGGCTCCAGATCCTCAAGGAGCACCCGTTGGAGAAACGCAGCTCCAGAA**  
**CGAGAAGCTTACTCTACTACCTCCAGAACGGCCGACATGTACGTGGACCAGGAGCTCGACATCAACCGCCTCTCCGACTACG**  
**ACGTGGACCACATCGTCCCGCAGTCTTCTCAAGGACGACTCCTTCGACAACAAGGTGCTCACGCGCTCCGACAAGAACCAGCGCC**  
**AAGTCCGACAACGTGCCGTCCGGGAGGTGGTGAAGAAGATGAAGAAGTACTGGCGCCAGCTCCTCAACGCCAAGCTCATCACGCAG**  
**CGCAAGTTCGACAACCTCACGAAGCCGAGCGCGCCGCTCTCCGAGCTCGACAAGGCCGGCTTCATCAAGCGCCAGCTCGTGGAA**  
**GACGCGCCAGATCACGAAGCACGTGGCCAGATCCTCGACTCCCGCATGAACACGAAGTACGACGAGAACGACAAGCTCATCCGCC**

AGGTGAAGGTGATCACGCTCAAGTCCAAGCTCGTGTCCGACTTCCGCAAGGACTTCCAGTTCTACAAGGTGCGCGAGATCAACAAC  
TACCACCACGCCACGACGCCTACCTCAACGCCGTGGTGGCAGCGCCCTCATCAAGAAGTACCCGAAGCTCGAGTCCGAGTTCGT  
GTACGGCGACTACAAGGTGTACGACGTGCGCAAGATGATCGCCAAGTCCGAGCAGGAGATCGGCAAGGCCACGGCCAAGTACTTCT  
TCTACTCCAACATCATGAACCTTCTCAAGACGGAGATCACGCTCGCCAACGGCGAGATCCGCAAGCGCCCGCTCATCGAGACGAAC  
GGCGAGACGGGCGAGATCGTGTGGGACAAGGGCCGCGACTTCGCCACGGTGCGCAAGGTGCTCTCCATGCCGCAGGTGAACATCGT  
GAAGAAGACGGAGGTGCAGACGGGCGGCTTCTCCAAGGAGTCCATCTCCCGAAGCGCAACTCCGACAAGCTCATCGCCCGCAAGA  
AGGACTGGGACCCGAAGAAGTACGGCGGCTTCGACTCCCCGACGGTGGCCTACTCCGTGCTCGTGGTGGCCAAGGTGGAGAAGGGC  
AAGTCCAAGAAGCTCAAGTCCGTGAAGGAGCTCCTCGGCATCACGATCATGGAGCGCTCCTCCTTCGAGAAGAACCCGATCGACTT  
CCTCGAGGCCAAGGGTACAAGGAGGTGAAGAAGGACCTCATCATCAAGCTCCCGAAGTACTCCCTCTTCGAGCTCGAGAACGGCC  
GCAAGCGCATGCTCGCTCCGCCGGGAGCTCCAGAAGGGCAACGAGCTCGCCCTCCCGTCCAAGTACGTGAACCTCTCTACCTC  
GCCTCCCACTACGAGAAGCTCAAGGGCTCCCGGAGGACAACGAGCAGAAGCAGCTCTTCGTGGAGCAGCACAAGCACTACCTCGA  
CGAGATCATCGAGCAGATCTCCGAGTCTCCAAGCGGTGATCTCGCCGACGCCAACCTCGACAAGGTGCTCTCCGCCTACAACA  
AGCACCGCGACAAGCCGATCCGCGAGCAGGCCGAGAACATCATCCACCTCTTCACGCTCACGAACCTCGGCGCCCCGGCCGCCTTC  
AAGTACTTCGACACGACGATCGACCGCAAGCGCTACACGTCCACGAAGGAGGTGCTCGACGCCACGCTCATCCACCAGTCCATCAC  
GGGCCTCTACGAGACGCGCATCGACCTCTCCAGCTCGGCGGGCGACTAGCTGCTTTAATGAGATATGCGAGACGCCATGATCGCA  
TGATATTTGCTTTCAATTCTGTTGTGCACGTTGTAAAAACCTGAGCATGTGTAGCTCAGATCCTTACCGCCGGTTTCGGTTCATTC  
TAATGAATATATACCCGTTACTATCGTATTTTTATGAATAATATCTCCGTTCAATTTACTGATGTACCCTACTACTTATATGT  
ACAATATTTAAAATGAAAACAATATATTTGTGCTGAATAGGTTTATAGCGACATCTATGATAGAGCGCCACAATAACAAAACAATTGCC  
TTTTATTATTACAAATCCAATTTTCGGGGATCCTCTAGAGTGCACCTGCAGGCATGCAAGCTTGGCACGGTTACCAAGCGCGCAA

**BdAUX1 k.o. sgRNA cassette: *BdU6 promoter* *tracrRNA* *OsU6 promoter* *BsaI* or *BtgZI* sites**

AAAGGATCCGGCGCGCCCTACTTGGGCTGTTGCTCTCTACTGGGTTGGGCCGCATGGACTGACACAGGCCACGCGGGTCTCTTAC  
GAGCCTGGGGCTGGCCTGATCCGATGGTGTGCTGATCAAGGCAACAGGCTAGAAAGTTAGTCCCACCTCGCGAGATGAAGGATAGT  
TTGACTAGATTATAAACATTCTGCTACCACCCTTCTGagagaccgagctcggctctcaGTTTTAGAGCTAGAAATAGCAAGTTAAAA  
TAAGGCTAGTCCGTTATCAACTTGAAAAAGTGGCACCGAGTCGGTGTCTTTTTTTGAGATTTCCAACCAGGTCCCTGGAGCCCAT  
GTCTAGTAACGGCCCGCAGTGTGCTGGAATTGCCCTTGATCATGAACCAACGGCCTGGCTGTATTTGGTGGTGTGTAGGGAGAT  
GGGGAGAAGAAAAGCCGATTCTCTTCTGTGATGGCTGGATGCATGCGGGGGAGCGGGAGGCCAAGTACGTGCACGGTGAGCG  
GCCCCACAGGGCGAGTGTGAGCGCGAGAGGCGGGAGGAACAGTTTACTACCACATTGCCAGCTAACTCGAACCGGACCAACTTATA  
AACCCGCGCGCTGTGCTTGTGTacaattgcatcatcgagtcagcgatgagtagacagcaaGTTTTAGAGCTAGAAATAGCAAGTTA  
AAATAAGGCTAGTCCGTTATCAACTTGAAAAAGTGGCACCGAGTCGGTGTCTTTTTTTGAGATTTTGCACCCGGTCTGCAGGGTC  
AAGCTTAAA

**List of primers:**

pUbil-For	5'-GAG CTC CAG CTT GCA TGC CTG CAG TG-3'
pUbil-Rev	5'-GAG CTC TCT AGA GTC GAC CTG CAG AA-3'
sgRNA-BdAux1a F2	5'-TCT CGT CAC CAG CTT CCT CTG GCA-3'
sgRNA-BdAux1a R	5'-AAA CTG CCA GAG GAA GCT GGT GAC-3'
sgRNA-BdAux1b F	5'-GTG TGA TCC GGT AGT TGT GGA AGG-3'
sgRNA-BdAux1b R	5'-AAA CCC TTC CAC AAC TAC CGG ATC-3'
BdAUX1L-WT-F	5'-GTG AAC TTT CCA CAC TGA GC-3'
BdAUX1L-WT-R	5'-TCA CAA GAG CTG GGC AAT GG-3'
pJJ2LBA-RB-F2	5'-CAG GAA TTC ATG CCG ACA GC-3'
KanR-XhoI-For	5'-CCA CTC GAG GAT CTC CAC TCT AGT CGA G-3'
KanR-XhoI-Rev	5'-TGT CTC GAG TTG AAC GAT CGG GGA TCC-3'
promBdAUX1F2SacI	5'-CTA GAG CTC TGG ACG TGG TTT TGT CCT AG-3'
promBdAUX1R2SalI	5'-ACG CGT CGA CAT CTC TTC AAC GCG CTG TC-3'
pCAMBIA-promBdAUX1F	5'-CAT GAT TAC GAA TTC GAG CTC GTC ACT TAA TCT CGT C-3'
promBdAUX1seq6R	5'-CGA ATT TCC TCT CTG TCT CC-3'
promBdAUX1seq6F	5'-GGA GAC AGA GAG GAA ATT CG-3'

genBdAUX1seq5R	5'-TGG AAC GAT GAG GTG CAT TG-3'
genBdAUX1seq5F	5'-CAA TGC ACC TCA TCG TTC CA-3'
genBdAUX1R3-pCAMBIA	5'-GGA AAT TCG AGC TGG TCA CCT AGC AAG CAT TAC TGG GTT-3'
BdAUX1p-XmaI F	5'-GCG ACT GTG CCA ACA CCC-3'
BdAUX1p-sGFP R	5'-GCC CTT GCT CAC CAT CTC TTC AAC GCG CTG TCC TC-3'
sGFP(S65T) F	5'-ATG GTG AGC AAG GGC GAG G-3'
sGFP+linker R	5'-ATC CTC TAG AGT CGA CCT TGT ACA GCT CGT CCA TGC-3'
sGFPlink-AUX1g F	5'-GTC GAC TCT AGA GGA TCC ATG GTG CCG CGC GAG CAT-3'
AUX1g-PacI-pCAM R	5'-TTT TTC CTC GGG TTA GTT AAT TAA TTC-3'