**Supplemental Tables**

### Table S1. Phenotyping information for early- and normal-planted field trials.

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Phenotype** | **AB16\_b1*a*** | **AB16\_b2*a*** | **AB17*a*** | **MN17*a*** | **HA17*a*** | **HA18*a*** | **AB18*b*** | **MN18*b*** |
| Planting | Early | Early | Early | Early | Early | Early | Normal | Normal |
| Chilling Stress | Yes | Yes | Yes | Yes | Yes | Yes | No | No |
| Planting Date | April 7 | April 15 | April 25 | May 8 | April 20 | April 17 | May 7 | June 5 |
| First Emergence | April 26 | April 28 | May 8 | May 14 | May 8 | May 2 | May 10 | June 11 |
| Emergence Count  | April 28 | May 2 | May 15 | May 17 | May 12 | May 10 | May 14 | June 19 |
| Seedling Vigor1  | May 10 | May 9 | May 18 | May 24 | May 16 | May 17 | - | June 19 |
| Damage Rating  | May 6 | - | - | May 26 | May 23 | - | - | - |
| Seedling Vigor2  | May 19 | May 23 | May 25 | June 5 | June 6 | May 25 | May 23 | June 26 |
| Seedling Vigor3  | May 25 | May 26 | June 7 | June 9 | May 31 | June 8 | June 6 | - |

***a***Early-planted field trials, represented using a two digit field location name (in Kansas) and last two digits of the year the field trial was conducted. Field locations: Ashland Bottoms (AB), Manhattan (MN), and Hays (HA).

***b***Normal-planted field trials conducted in 2018 at AB and MN.

### Table S2. Molecular markers for sequence variant analysis.

|  |  |  |
| --- | --- | --- |
| **Gene*a*** | **Forward primer** | **Reverse primer** |
| *CBF1* | 5’-TTCAGCAGGCAGCCAAGGT-3’ | 5’-TCATGTACGACGAGGAGGA-3’ |
|  | 5’-ACACCCGCCCTTCACCCAT-3’ | 5’-CTCCTGATCACGCTTCATC-3’ |
| *Tan1* | 5’-CGTTTACCCGCCGTTCGCA-3’ | 5’-TCGTAGACGATGGTGGAGTG-3’ |
|  | 5’-GGACCGCCTCCATCGACAC-3’ | 5’-CCTCATACCAATGTGCACG-3’ |

### a C-repeat binding factor1 (CBF1) and Tannin1 (Tan1) genes were amplified from the four NAM founders.Table S3. Composite interval mapping (CIM) with early-planted field phenotype BLUPs

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Family*a*** | **Trait*b*** | **Peak SNP** | **QTL Interval (Mb)** | **LOD*c*** | **PVE*d*** | **Additive effect*e*** | **Known loci*f*** | **Distance to known loci** |
| HKZa | EPEC | S6\_53866236 | 53–55 | 5.8 | 21.9 | 0.20 |  |  |
|  | EPSV1 | S4\_60494734 | 58–62 | 4.3 | 16.9 | -0.14 | *Tan1* | 1.8 Mb |
|  | EPSV2 | S4\_473959 | 0.4–77 | 3.4 | 13.7 | 0.15 |  |  |
|  |  | S5\_69294666 | 68–69 | 3.9 | 15.3 | 0.13 |  |  |
|  | EPSV3 | S4\_597351 | 0.4–13 | 3.1 | 12.7 | 0.09 |  |  |
|  |  | S5\_69294666 | 68–70 | 3.9 | 15.7 | 0.11 |  |  |
|  |  | S7\_62611661 | 61–64 | 3.5 | 13.8 | 0.09 | *Dw3* | 3 Mb |
| Kao | EPEC | S4\_61061060 | 11–63 | 3 | 7.3 | -0.09 | *Tan1* | 1.2 Mb |
|  |  | S8\_52136076 | 7–54 | 3 | 6.1 | -0.09 |  |  |
|  | EPSV1 | S2\_10876295 | 9–13 | 4.5 | 9.1 | -0.11 | *Tan2* | 3 Mb |
|  |  | S9\_52692821 | 51–53 | 4 | 8 | -0.10 |  |  |
|  | EPSV2 | S2\_16848234 | 13–55 | 6.8 | 13.4 | -0.12 |  |  |
|  |  | S4\_61061060 | 11–62 | 4.5 | 9.1 | -0.09 | *Tan1* | 1.2 Mb |
|  | EPSV3 | S4\_62599717 | 61–63 | 3.9 | 7.9 | -0.09 | *Tan1* | 0.3 Mb |
| NSZ | EPEC | S2\_8451281 | 7–9 | 3.7 | 6.4 | -0.1 | *Tan2* | 0.5 Mb |
|  |  | S3\_72404691 | 72–74 | 4.1 | 7.1 | 0.09 |  |  |
|  |  | S4\_62882446 | 61–64 | 6.2 | 10.5 | -0.13 | *Tan1* | 0.6 Mb |
|  | EPSV1 | S1\_48528744 | 25–53 | 6.4 | 10.9 | -0.14 |  |  |
|  |  | S3\_72404691 | 11–73 | 3.2 | 5.6 | 0.09 |  |  |
|  |  | S4\_62882446 | 59–63 | 3.3 | 5.7 | -0.09 | *Tan1* | 0.6 Mb |
|  |  | S7\_59412395 | 58–60 | 4.9 | 8.4 | -0.12 | *Dw3* | 0.4 Mb |
|  |  | S9\_55625332 | 54–56 | 4.4 | 7.6 | -0.11 | *Dw1* | 1.4 Mb |
|  | EPSV2 | S1\_48528744 | 22–53 | 4.1 | 7.1 | -0.09 |  |  |
|  |  | S7\_2492149 | 1–3 | 3.2 | 5.5 | -0.08 |  |  |
|  | EPSV3 | S1\_48528744 | 21–53 | 4.1 | 7.1 | -0.1 |  |  |
|  |  | S7\_2492149 | 22–33 | 3.9 | 6.7 | -0.09 |  |  |

*a* The chilling NAM families: Hong Ke Zi (HKZa), Kaoliang (Kao), and NSZ, Niu Sheng Zui (NSZ).

*b* Early-planted emergence count (EPEC) and seedling vigor (EPSV1–3) were used for CIM.

*c* Logarithm of odds (LOD) score.

*d* Percentage of variation explained (PVE).

*e* Positive or negative effects of the BTx623 allele.

*f* Previously characterized genes colocalizing with the mapped QTL.

### Table S4. Composite interval mapping (CIM) with normal-planted field phenotype BLUPs.

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Family*a*** | **Trait*b*** | **Peak SNP** | **QTL Interval (Mb)** | **LOD*c*** | **PVE*d*** | **Additive effect*e*** | **Known loci*f*** | **Distance to known loci** |
| HKZa | NPEC | - | - | - | - | - |  |  |
|  | NPSV1 | S1\_58632047 | 58–60 | 3.4 | 13.8 | -0.13 |  |  |
|  |  | S7\_55055485 | 52–57 | 3.7 | 14.6 | -0.16 | *Dw3* | 4.8 Mb |
|  | NPSV2 | S5\_69294666 | 65–70 | 3.3 | 13.3 | 0.13 |  |  |
|  | NPSV3 | S2\_4944067 | 4–6 | 3.4 | 13.5 | -0.13 |  |  |
|  |  | S5\_69720862 | 68–70 | 4.2 | 16.5 | 0.13 |  |  |
| Kao | NPEC | S7\_57372955 | 56–58 | 4.1 | 8.2 | -0.10 | *Dw3* | 2.4 Mb |
|  |  | S10\_3575834 | 18–54 | 3.1 | 6.4 | 0.10 |  |  |
|  | NPSV1 | S1\_68309753 | 64 –71 | 3.3 | 6.7 | -0.10 |  |  |
|  |  | S2\_63030650 | 62–65 | 3.6 | 7.3 | -0.16 |  |  |
|  |  | S3\_13167621 | 12–14 | 11.3 | 21.2 | 0.28 |  |  |
|  |  | S7\_56368290 | 55–57 | 5.4 | 10.8 | -0.18 | *Dw3* | 3.4 Mb |
|  | NPSV2 | S1\_2859329 | 2–6 | 3.7 | 7.5 | -0.15 |  |  |
|  |  | S2\_16406623 | 13–18 | 6.1 | 12 | -0.18 |  |  |
|  |  | S3\_13097936 | 12–14 | 9.8 | 18.6 | 0.24 |  |  |
|  |  | S10\_3240955 | 17–53 | 3.5 | 7 | 0.15 |  |  |
|  | NPSV3 | S2\_16406623 | 15–55 | 3.7 | 7.6 | -0.12 |  |  |
|  |  | S3\_13167621 | 12–14 | 13.2 | 24.2 | 0.22 |  |  |
|  |  | S7\_57460966 | 13–62 | 3.3 | 6.7 | -0.1 | *Dw3* | 2.3 Mb |
|  |  | S10\_3536844 | 0.3–60 | 3.3 | 6.7 | 0.12 |  |  |
| NSZ | NPEC | S7\_39531433 | 12–56 | 2.9 | 5 | -0.07 |  |  |
|  | NPSV1 | S1\_58632047 | 56–59 | 7 | 11.9 | -0.16 |  |  |
|  |  | S7\_58340519 | 57–59 | 5.4 | 9.1 | -0.15 | *Dw3* | 1.5 Mb |
|  |  | S9\_56657651 | 56–58  | 3 | 5.3 | -0.11 | *Dw1* | 0.4 Mb |
|  | NPSV2 | S1\_26240607 | 21–52 | 7.5 | 12.5 | -0.17 |  |  |
|  |  | S2\_66319995 | 58–70 | 3 | 5.2 | -0.1 |  |  |
|  |  | S3\_73329070 | 1 –73 | 4 | 6.9 | 0.12 |  |  |
|  |  | S7\_27327629 | 14–53 | 3.5 | 6 | -0.12 |  |  |
|  | NPSV3 | S1\_26240607 | 21–54 | 3.3 | 5.7 | -0.1 |  |  |
|  |  | S3\_73329070 | 0.5 –73 | 3 | 5.3 | 0.10 |  |  |
|  |  | S9\_50757859 | 0.5–50 | 2.9 | 5.1 | -0.1 |  |  |

*a* The chilling NAM families: Hong Ke Zi (HKZa), Kaoliang (Kao), and NSZ, Niu Sheng Zui (NSZ).

*b* Normal-planted emergence count (NPEC) and seedling vigor (NPSV1–3) were used for CIM.

*c* Logarithm of odds (LOD) score.

*d* Percentage of variation explained (PVE).

*e* Positive or negative effects of the BTx623 allele.

*f* Previously characterized genes colocalizing with the mapped QTL.

### Table S5. Joint linkage mapping (JLM) with normal-planted field phenotype BLUPs.

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| **Trait*a*** | **QTL** | **QTL\_SNP** | **PVE*b*** | **Additive effect*c*** | **Known loci*d*** | **Distance to known loci** | **QTL name*e*** |
| NPEC | *qSbNPEC\_7-39* | S7\_39135105 | 4.3 | -0.05 |  |  |  |
|  | *qSbNPEC\_5-65* | S5\_65643264 | 3.5 | 0.09 |  |  |  |
| NPSV1 | *qSbNPSV1\_3-13* | S3\_13282550 | 6.8 | 0.09 |  |  |  |
|  | *qSbNPSV1\_1-57* | S1\_57941435 | 4.2 | -0.14 |  |  | *qSbCT01.57* |
|  | *qSbNPSV1\_7-57* | S7\_57053050 | 5.1 | -0.09 | *Dw3* | 2.8 Mb | *qSbCT07.59* |
|  | *qSbNPSV1\_9-53* | S9\_53265143 | 1 | -0.04 | *Dw1* | 3.8 Mb | *qSbCT09.57* |
|  | *qSbNPSV1\_2-62* | S2\_62962601 | 4.2 | -0.07 |  |  |  |
|  | *qSbNPSV1\_1-05* | S1\_5754842 | 4.3 | -0.07 |  |  | *qSbCT01.06* |
|  | *qSbNPSV1\_10-03* | S10\_3699364 | 2.7 | 0.14 |  |  |  |
|  | *qSbNPSV1\_1-11* | S1\_11764852 | 4.8 | -0.10 |  |  | *qSbCT01.13* |
| NPSV2 | *qSbNPSV2\_1-07* | S1\_7625404 | 6 | -0.08 |  |  | *qSbCT01.06* |
|  | *qSbNPSV2\_7-30* | S7\_30184611 | 4.5 | -0.07 |  |  |  |
|  | *qSbNPSV2\_1-57* | S1\_57941435 | 4.1 | -0.13 |  |  |  |
|  | *qSbNPSV2\_10-03* | S10\_3686338 | 3.1 | 0.16 |  |  |  |
|  | *qSbNPSV2\_2-64* | S2\_64314153 | 3.2 | -0.06 |  |  |  |
|  | *qSbNPSV2\_2-25* | S2\_25130132 | 3.8 | -0.06 |  |  |  |
|  | *qSbNPSV2\_3-13* | S3\_13759646 | 3.7 | 0.09 |  |  |  |
|  | *qSbNPSV2\_1-14* | S1\_14774558 | 3 | -0.08 |  |  | *qSbCT01.13* |
|  | *qSbNPSV2\_9-53* | S9\_53265132 | 0.5 | -0.03 | *Dw1* | 3.7 Mb | *qSbCT09.57* |
|  | *qSbNPSV2\_3-13* | S3\_13245510 | 5 | 0.07 |  |  |  |
| NPSV3 | *qSbNPSV4\_3-13* | S3\_13282550 | 6.2 | 0.07 |  |  |  |
|  | *qSbNPSV4\_2-64* | S2\_64314153 | 3.7 | -0.05 |  |  |  |
|  | *qSbNPSV4\_10-04* | S10\_4652526 | 2.6 | 0.14 |  |  |  |
|  | *qSbNPSV4\_7-41* | S7\_41996273 | 4.4 | -0.06 |  |  |  |
|  | *qSbNPSV4\_7-60* | S7\_60102977 | 3.6 | -0.08 | *Dw3* | 0.2 Mb |  |
|  | *qSbNPSV4\_1-07* | S1\_7784945 | 3.9 | -0.06 |  |  | *qSbCT01.06* |
|  | *qSbNPSV4\_1-11* | S1\_11801181 | 4.8 | -0.08 |  |  | *qSbCT01.13* |
|  | *qSbNPSV4\_4-57* | S4\_57456389 | 2 | 0.1 | *Tan1* | 4 Mb |  |
|  | *qSbNPSV4\_1-57* | S1\_57941435 | 2.1 | -0.1 |  |  |  |

*a* Normal-planted emergence count (EPEC) and seedling vigor (EPSV1–3) BLUPS were used for JLM.

*b* Percentage of variation explained (PVE).

*c* Positive or negative effects of the BTx623 allele.

*d* Previously characterized genes colocalizing with the mapped QTL.

*e* QTL in 2 Mb interval, detected with different seedling traits, were assigned a common name.

### Table S6. Composite interval mapping (CIM) of agronomic traits and grain tannins.

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Family*a*** | **Trait*b*** | **Peak SNP** | **QTL Interval (Mb)** | **LOD*c*** | **PVE*d*** | **Additive effect*e*** | **Known loci*f*** | **Distance to known loci** |
| HKZa | PHT | S1\_68318606 | 67–71 | 4.5 | 18 | 17 |  |  |
|  |  | S7\_60216181 | 60–61 | 12 | 41 | -30 | *Dw3* | 0.3 Mb |
|  |  | S9\_57804067 | 56–58 | 6 | 23 | -18 | *Dw1* | 0.7 Mb |
|  | FT | S3\_13889853 | 1–15 | 4 | 16 | 5 |  |  |
|  |  | S10\_56467046 | 56–57 | 3 | 12 | -3 |  |  |
|  | Tannins | S4\_61061060 | 60–62 | 34 | 77 | -0.8 | *Tan1* | 1.2 Mb |
| NSZ | PHT | S7\_59953003 | 59–60 | 35 | 46 | -38 | *Dw3* | 0.1 Mb |
|  |  | S9\_56657651 | 56–57 | 22 | 32 | -29 | *Dw1* | 0.4 Mb |
|  | FT | S1\_59369684 | 58–61 | 4 | 6 | -3 |  |  |
|  |  | S2\_63186100 | 60–65 | 5 | 8 | 4 |  |  |
|  |  | S8\_61537016 | 59–62 | 3 | 6 | 2 |  |  |
|  |  | S9\_58670296 | 58–59 | 4 | 7 | -3 |  |  |
|  | Tannins | S4\_61212673 | 60–62 | 23 | 34 | -0.8 | *Tan1* | 1. 1 Mb |
| Kao | PHT | S7\_56844702 | 56–64 | 6 | 11 | -28 | *Dw3* | 3 Mb |
|  |  | S9\_51987531 |  51–53  | 10 | 19 | -36 |  |  |
|  | FT | S2\_66179497 | 65–67 | 5 | 10 | 3 |  |  |
|  |  | S10\_54171866 | 53–55 | 5 | 10 | -3 |  |  |
|  | Tannins | S4\_62599717 | 61–63 | 1450 | 100 | -0.92 | *Tan1* | 0.8 Mb |

*a* The NAM population families referred by the alternate NAM founder.

*b* Plant height (PHT), flowering time (FT), and grain tannin phenotypes were used for CIM.

*c* Logarithm of odds (LOD).

*d* Percentage of variation explained (PVE).

*e* Positive or negative effects of the BTx623 allele.

*f* Previously characterized genes colocalizing with the mapped QTL.

### Table S7. Comparison of chilling tolerance QTL with previous mapping studies.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Chr*a*** | **Position*b*** | **Trait*c*** | **Tagging marker** | **Interval (Mb)*d*** | **Reference*e*** | **NAM QTL*f*** |
| 1 | 66.04 | SV | Xtxp58 | 19–59 | Knoll *et al.* 2007 | *qSbCT01.57* |
| 1 | 57.36 | Emer, SV | Xtxp43 | 57–59 | Knoll *et al.* 2007 | *qSbCT01.57* |
| 2 | 4.98 | Emer, SV | Xtxp211 | 4–27 | Knoll *et al.* 2007 | *qSbCT02.08* |
| 4 | 61.86 | Emer, SV | Xtxp51 | 61–68 | Knoll *et al.* 2007 | *qSbCT04.62* |
| 5 | 51.77 | ESV | Xtxp14 | - | Knoll *et al.* 2007 | *-* |
| 9 | 4.2 | Emer, SV | Xtxp287 | - | Knoll *et al.* 2007 | *-* |
| 1 | 4.05 | FEarlyGerm1.1 | Xsbarslbk1.04 | 0–7 | Burow *et al.* 2010 | *qSbCT01.06* |
| 1 | 68.33 | FEarlyGerm1.2 | Xtxp279 | 65–71 | Burow *et al.* 2010 | *-* |
| 2 | 56.01 | Germ12C-2.1 | Xsbarslbk\_2.56 | 44–60 | Burow *et al.* 2010 | *-* |
| 4 | 61.86 | FEarlyGerm4.1 | Xtxp212 | 59–65 | Burow *et al.* 2010 | *qSbCT04.62* |
| 4 |  | FearlyVigor4.1 | Xtxp327 | - | Burow *et al.* 2010 |  |
| 7 |  | FEarlyGerm7.1 | Xtxp218 | 7–57 | Burow *et al.* 2010 | *qSbCT07.59* |
| 7 |  | FEarlyGerm7.1 | Xtxp159 | 7–57 | Burow *et al.* 2010 | *qSbCT07.59* |
| 9 | 7.07 | FEarlyGerm9.1 | Xsbarslbk9.07 | 2–44 | Burow *et al.* 2010 | *-* |
| 9 | 47.75 | FEarlyGerm9.2 | Xtxp010 | 44–57 | Burow *et al.* 2010 | *qSbCT09.57* |
| 9 | 52.85 | FEarlyGerm9.3 | Xsbarslbk\_9.53 | 52–57 | Burow *et al.* 2010 | *qSbCT09.57* |
| 7 | 56.99 | Emer (GWAS) | S7\_56998511 |  | Upadhyaya *et al.* 2015 | *qSbCT07.59* |
| 4 | 49.4 | Emer | sPb-4851 | 13–40 | Fiedler *et al.* 2012 | *-* |
| 1 | 57.36 | Emer | Xtxp43 | 55–57 | Fiedler *et al.* 2012 | *qSbCT01.57* |

*a* Chromosomes with previously reported chilling tolerance QTL.

*b* Genetic distance of the previously mapped QTL on the BTx623 reference genome *SbV3.1.*

*c* Seedling traits mapping the previous QTL.

*d* QTL interval derived using flanking markers of previously mapped QTL.

*e* Previous biparental mapping studies to map early-season chilling tolerance.

*f* Overlapping chilling tolerance QTL between the chilling NAM and previous biparental populations.

### Table S8. Inadvertent selection of chilling-sensitive alleles

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Genotype*a*** | **EPEC*b*** | **EPSV1*c*** | **EPSV2*d*** | **EPSV3*e*** | **Height (cm)** | ***Dwarfing allelesf*** |
| 14FS125\_Kao | 2.5 | 2.2 | 2.5 | 2.8 | 144 | *dw1 dw3* |
| 14FS155\_Kao | 2.5 | 1.8 | 2 | 2.1 | 117 | *dw1 dw3* |
| 14FS169\_Kao | 1.7 | 2.3 | 2.5 | 2.7 | 123 | *dw1 dw3* |
| 14FS197\_Kao | 2.3 | 2.4 | 2.7 | 3 | 118 | *dw1 dw3* |
| 14FS205\_Kao | 2.1 | 2.3 | 2.7 | 2.9 | 129 | *dw1 dw3* |
| 14FS208\_Kao | 3 | 2.6 | 3 | 3.4 | 172 | *dw1 dw3* |
| 14FS256\_Kao | 1.9 | 2.8 | 3 | 3.1 | 165 | *dw1 dw3* |
| 14FS261\_Kao | 2.2 | 2.1 | 1.9 | 2.2 | 93 | *dw1 dw3* |
| 14FS273\_Kao | 2.6 | 2.8 | 2.9 | 3 | 168 | *dw1 dw3* |
| 14FS292\_Kao | 2 | 2.2 | 2.3 | 2 | 123 | *dw1 dw3* |
| 14FS306\_Kao | 1.4 | 1.8 | 1.9 | 2.3 | 111 | *dw1 dw3* |
| 14FS314\_Kao | 2.1 | 2.3 | 2.8 | 2.7 | 119 | *dw1 dw3* |
| 14FS336\_Kao | 2.5 | 2.2 | 2.5 | 2.7 | 126 | *dw1 dw3* |
| 15FS005\_NSZ | 2.8 | 2.9 | 3.2 | 3.3 | 168 | *dw1 dw3* |
| 15FS032\_NSZ | 1.8 | 2.1 | 2.6 | 2.7 | 65 | *dw1 dw3* |
| 15FS036\_NSZ | 2.4 | 2.4 | 2.5 | 2.7 | 121 | *dw1 dw3* |
| 15FS063\_NSZ | 2.1 | 2.7 | 2.6 | 3 | 111 | *dw1 dw3* |
| 15FS068\_NSZ | 2.4 | 2.6 | 2.6 | 3 | 116 | *dw1 dw3* |
| 15FS083\_NSZ | 3.1 | 2.5 | 2.7 | 3 | 126 | *dw1 dw3* |
| 15FS105\_NSZ | 2.2 | 2.5 | 2.6 | 2.9 | 116 | *dw1 dw3* |
| 15FS142\_NSZ | 2 | 2.3 | 2.5 | 2.4 | 115 | *dw1 dw3* |
| 15FS152\_NSZ | 2 | 2.1 | 2.7 | 2.9 | 115 | *dw1 dw3* |
| 15FS202\_NSZ | 1.6 | 1.4 | 1.5 | 1.7 | 128 | *dw1 dw3* |
| 15FS218\_NSZ | 2.9 | 2.7 | 3.1 | 3.4 | 128 | *dw1 dw3* |
| 15FS609\_HKZa | 2.2 | 2.6 | 2.7 | 2.7 | 97 | *dw1 dw3* |
| 15FS642\_HKZa | 2.3 | 2.4 | 2.5 | 2.7 | 100 | *dw1 dw3* |
| 15FS653\_HKZb | 2.2 | 2.3 | 2.9 | 2.8 | 101 | *dw1 dw3* |
| 15FS654\_HKZa | 1.8 | 2.3 | 2.5 | 2.8 | 109 | *dw1 dw3* |
| 15FS666\_HKZb | 2.8 | 2.3 | 2.8 | 2.9 | 154 | *dw1 dw3* |
| 15FS679\_HKZb | 1.5 | 2.2 | 2.3 | 2.9 | 95 | *dw1 dw3* |
| 15FS682\_HKZa | 1.8 | 2.1 | 2 | 2.7 | 97 | *dw1 dw3* |
| 15FS698\_HKZb | 2.9 | 2.9 | 3 | 3.3 | 121 | *dw1 dw3* |
| 15FS705\_HKZa | 2.7 | 2.7 | 2.5 | 2.8 | 115 | *dw1 dw3* |
| 15FS709\_HKZb | 2.9 | 2.6 | 2.9 | 3.1 | 122 | *dw1 dw3* |
| 15FS715\_HKZb | 2.3 | 2.7 | 2.5 | 3.1 | 112 | *dw1 dw3* |
| 15FS761\_HKZa | 2.4 | 2.7 | 2.7 | 3.2 | 132 | *dw1 dw3* |
| 15FS794\_HKZa | 2.5 | 3 | 2.8 | 3.1 | 127 | *dw1 dw3* |
| 15FS820\_HKZb | 2.1 | 2.1 | 2.2 | 2.4 | 97 | *dw1 dw3* |
| BTx623\_P | 2.5 | 2.6 | 2.7 | 2.9 | 137 | *dw1 dw3* |
| HKZ\_P | 3.5 | 3.9 | 3.9 | 4.1 | 269 | *Dw1 Dw3* |
| Kao\_P | 3.4 | 3.4 | 3.3 | 3.6 | 278 | *Dw1 Dw3* |
| NSZ\_P | 3.6 | 4 | 3.6 | 3.8 | 246 | *Dw1 Dw3* |

*a* The chilling NAM RILs selected based on agronomic suitability.

*b* Early-planted emergence count (EPEC) rating from AB16\_b1 field trial.

*c-e* Early-planted seedling vigor (EPSV1–3) rating from AB16\_b1 field trial.

*f* Alleles of the *Dw1* and *Dw3* genes in the selected NAM RILs. The four founders were included as controls.

**Literature cit**ed

Burow G., J. J. Burke, Z. Xin, and C. D. Franks, 2010 Genetic dissection of early-season cold tolerance in sorghum (Sorghum bicolor (L.) Moench). Mol. Breed. 28: 391–402. https://doi.org/10.1007/s11032-010-9491-4

Fiedler K., W. A. Bekele, W. Friedt, R. Snowdon, H. Stützel, *et al.*, 2012 Genetic dissection of the temperature dependent emergence processes in sorghum using a cumulative emergence model and stability parameters. Theor. Appl. Genet. 125: 1647–1661. https://doi.org/10.1007/s00122-012-1941-4

Knoll J., N. Gunaratna, and G. Ejeta, 2007 QTL analysis of early-season cold tolerance in sorghum. Theor. Appl. Genet. 116: 577–587. https://doi.org/10.1007/s00122-007-0692-0

Upadhyaya H. D., Y.-H. Wang, D. V. S. S. R. Sastry, S. L. Dwivedi, P. V. V. Prasad, *et al.*, 2015 Association mapping of germinability and seedling vigor in sorghum under controlled low-temperature conditions. Genome 59: 137–145. https://doi.org/10.1139/gen-2015-0122

## *Note on tannin genotypes*

As early-planted seedling trait QTL colocalized with tannin genes, we took advantage of Mendelian genetics to infer the NAM founders tannin genotypes. Given that tannin sorghums must be homozygous dominant at the two complementary genes (Stephens 1946; Wu *et al.* 2012), the Chinese founders with grain tannin contains the *Tan1Tan1Tan2Tan2* genotype. By contrast, nontannin BTx623 with a non-functional *tan1-b* allele has the *tan1tan1*\_ \_ genotype [(Wu *et al.* 2012)](https://www.zotero.org/google-docs/?kJMdeE). Based on the tannin genotypes of the founders, each family segregates for tannins (nontannin versus tannin) as 1:1 when BTx623 contains a *Tan2* allele, and as 3:1 when BTx623 contains a *tan2* allele. Chi-square goodness of fit test supported that tannins were segregating as 1:1 in the HKZa, NSZ, and Kao families, as expected when BTx623 contains a *Tan2* allele, and rejected the same for the HKZb family. Tannin segregation as 1:1 in individual families indicates that BTx623 has the *tan1tan1Tan2Tan2* genotype. After dropping the HKZb RILs from JLM, the *Tan2* gene was not detected (Figure S22).