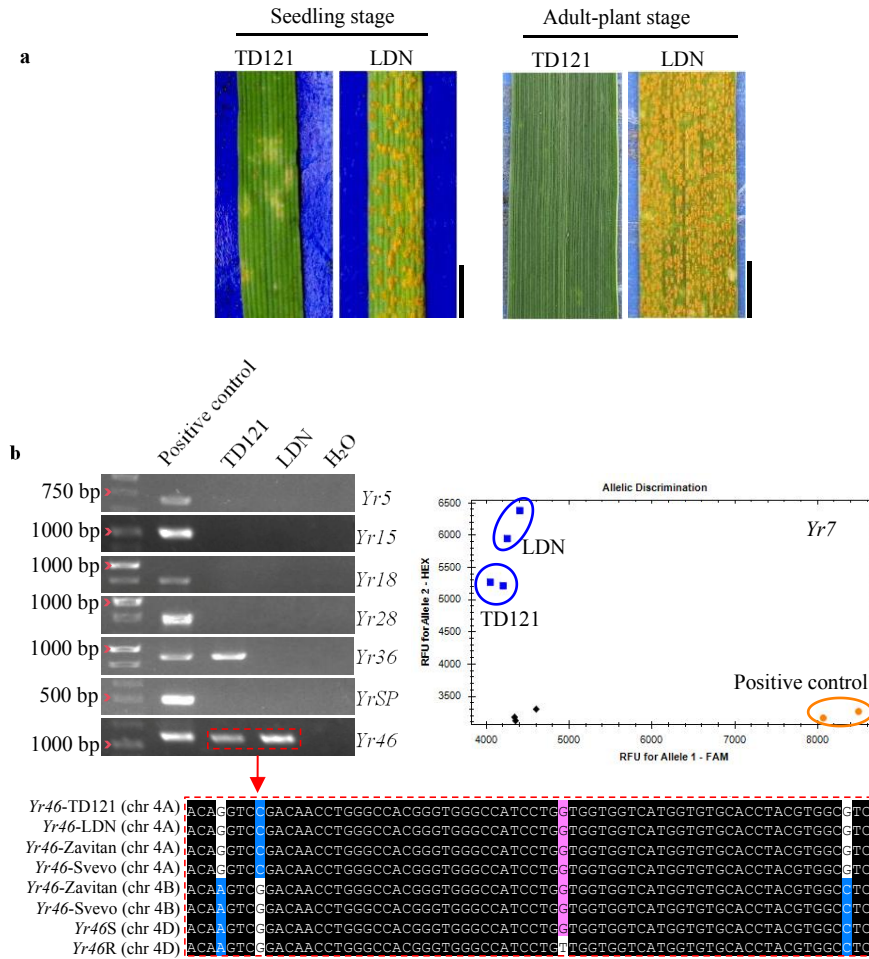


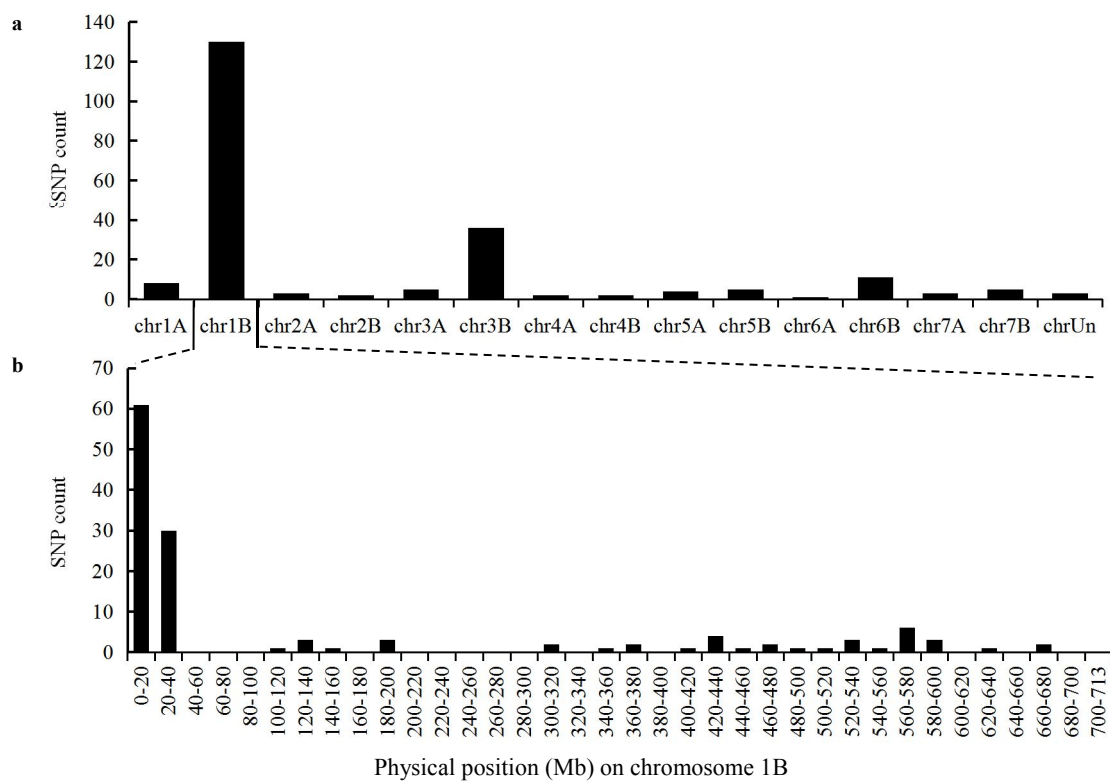
A head-to-head *NLR* gene pair from wild emmer confers stripe rust resistance in wheat

In the format provided by the
authors and unedited



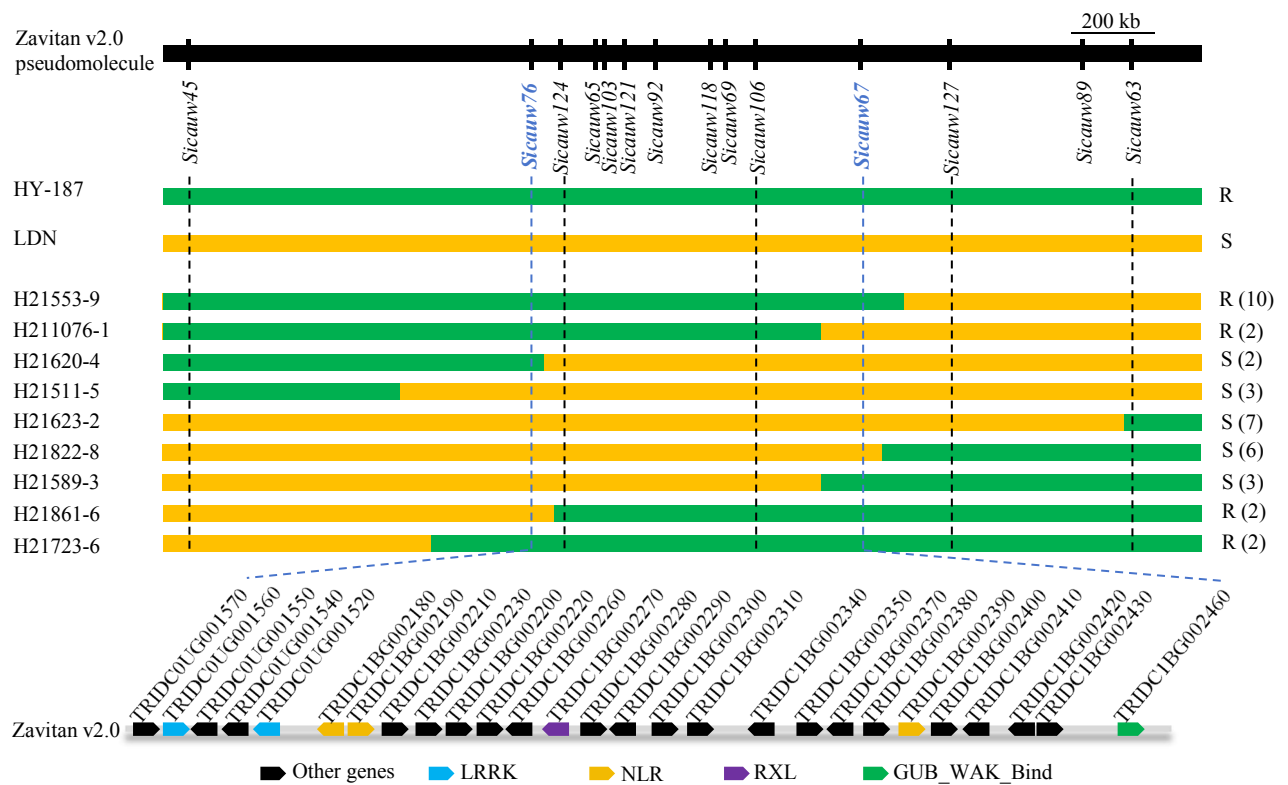
Supplementary Fig. 1 Characterization of stripe rust resistance in TD121.

a Responses of seedling and adult TD121 and LDN leaves following inoculation with *Pst*. Left panel, responses of seedling leaves to *Pst* race CYR34 at 14 days post inoculation (dpi). Right panel, responses of flag leaves to mixed Chinese *Pst* races in the field. Scale bar, 0.5 cm. **b** Molecular assays for the presence of eight cloned *Yr* genes in TD121. Genes *Yr5*, *Yr15*, *Yr18*, *Yr28*, *Yr36*, *Yr46*, and *YrSP* were assayed using gene-based primers. *Yr7* was characterized by KASP marker. Sanger sequencing showed that both TD121 and LDN carrying *Yr46* homologous sequences. *Yr46R*, the *Yr46* resistance gene (NCBI accession number KR604817); *Yr46S*, the *Yr46* susceptible allele (accession number KR604816). SNP (G/T) in purple shading results in critical amino acid change that distinguish the YR46S and YR46R proteins. Positive control, wheat line harboring the corresponding cloned *Yr* gene. LDN, susceptible control. The marker sequences can be found in Supplementary Table 2. All experiments were repeated three times with consistent results.



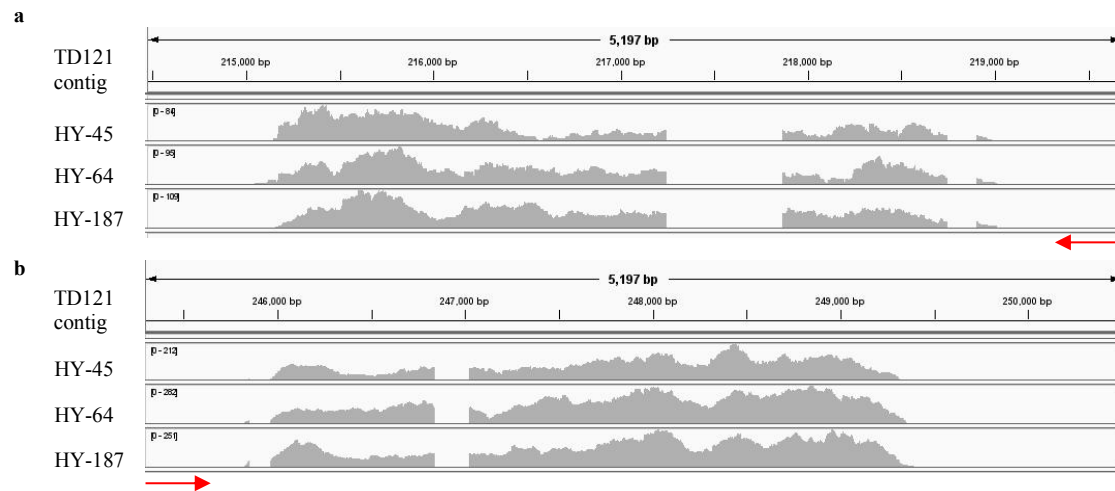
Supplementary Fig. 2 BSR-Seq analysis identifying SNPs putatively associated with *YrTD121*.

a Distribution of SNPs ($AFD \geq 0.7$) in resistant and susceptible DNA bulks on different wheat chromosomes. **b** Physical positions of SNPs on chromosome 1B based on Zavitan reference genome sequence v2.0.



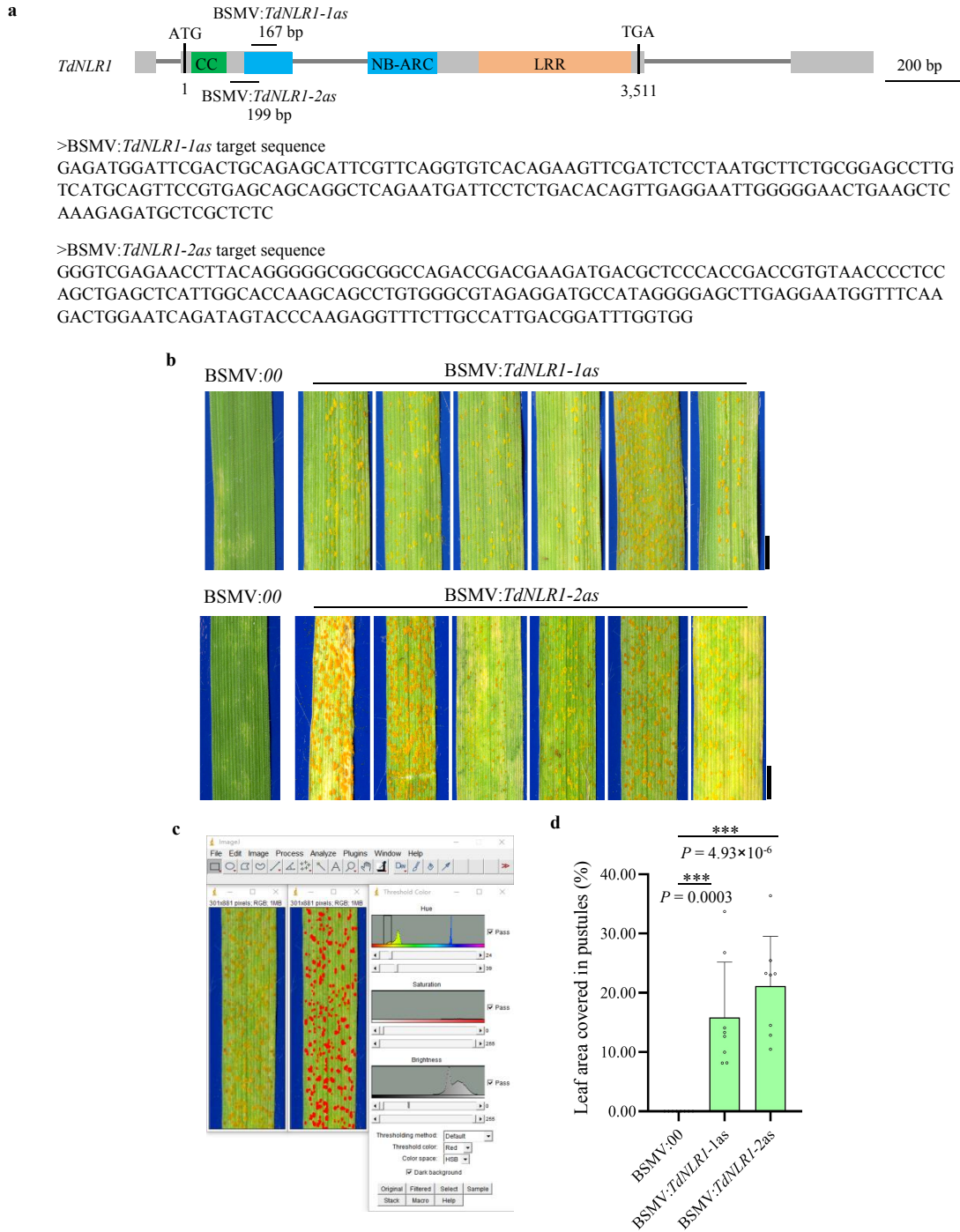
Supplementary Fig. 3 Graphical display of predicted genes on chromosome 1BS of Zavitan v2.0 corresponding to the *YrTD121* genomic region defined by nine recombination events.

Number of lines for each recombinant event is listed in parenthesis. Blue dashed lines indicate the region of the *YrTD121* locus as defined by fine mapping. S, susceptible, R, resistant.



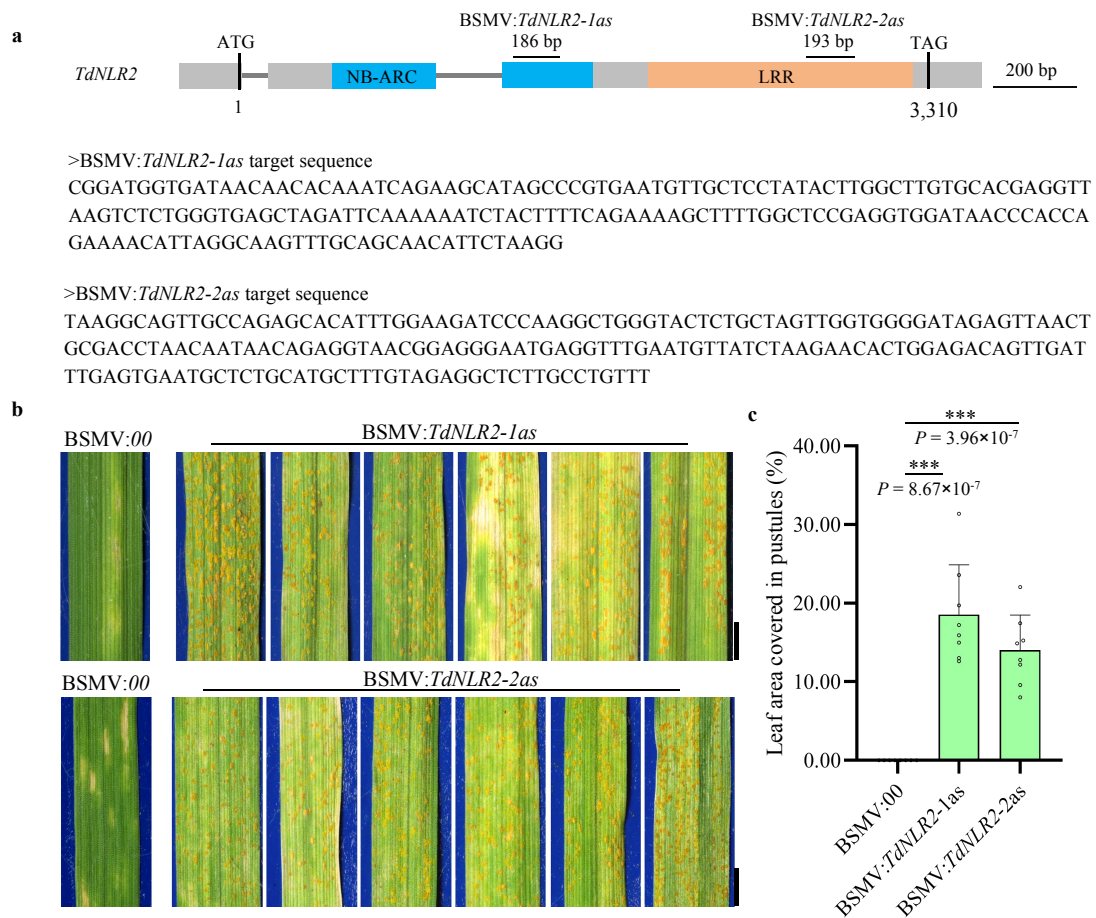
Supplementary Fig. 4 Identification of *TdNLR1* and *TdNLR2* full-length transcript sequences.

RNA sequencing reads generated from HY-45, HY-64, and HY-187 mapped against TD121 PacBio contigs and viewed using IGV software. **a** Alignment of *TdNLR1* transcripts. **b** Alignment of *TdNLR2* transcripts. Red arrows indicate the direction of transcription.



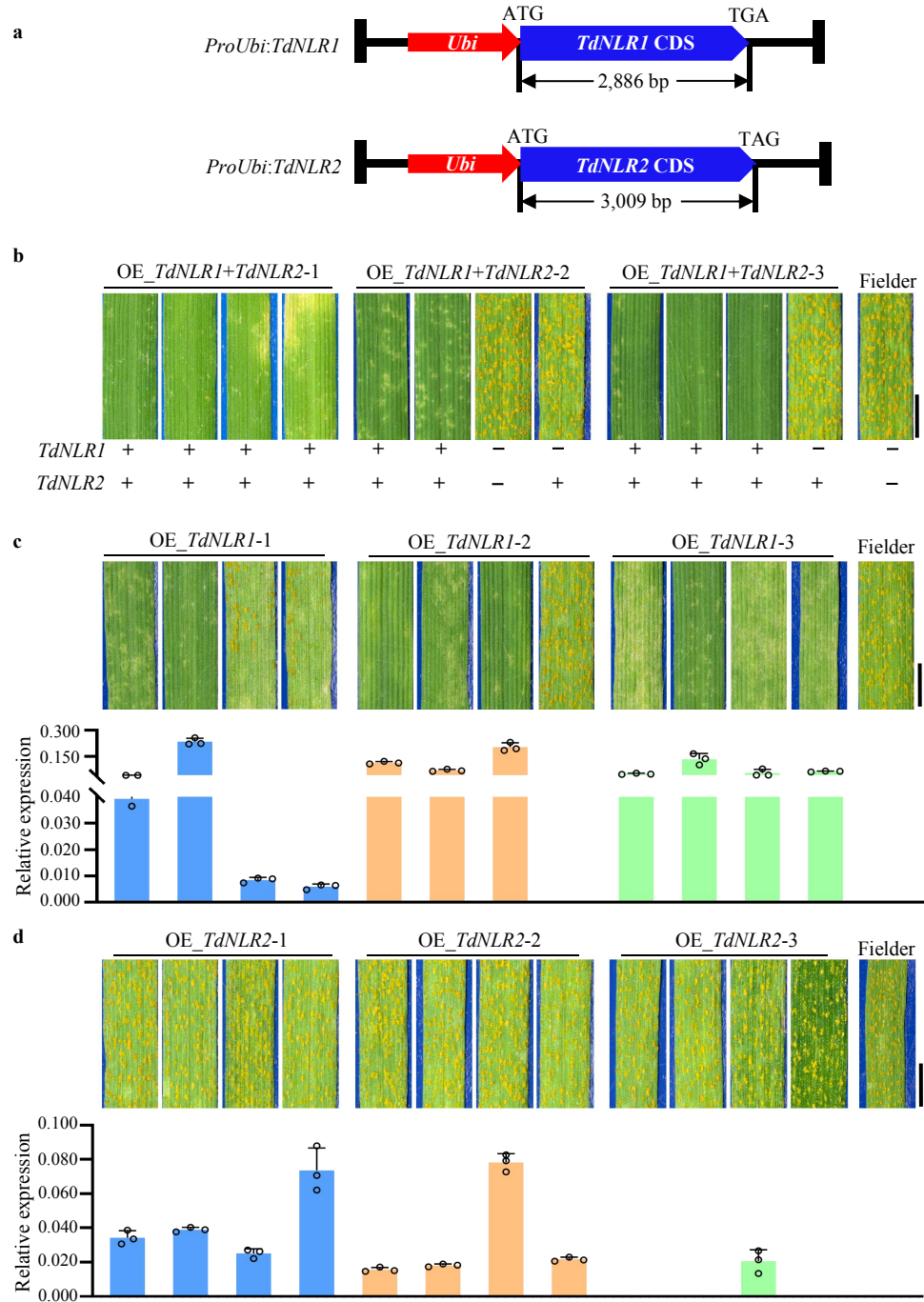
Supplementary Fig. 5 Validation of *TdNLR1* using VIGS.

a Target positions and sequences of *TdNLR1* in BSMV-VIGS experiments. **b** Phenotypes of HY-45 16 dpi with either BSMV:*TdNLR1-1as* or BSMV:*TdNLR1-2as* silencing constructs. Scale bar, 0.5 cm. **c** Screenshot of the color transformation step in ImageJ software (<https://imagej.net/ij/>) used to identify stripe rust pustules. **d** Mean percentage (%) leaf area covered in uredinia at 16 dpi; *TdNLR1*-silenced leaves of HY-45 were inoculated with *Pst* race CYR34. Error bars, mean $\pm$ SD ($n = 8$ biologically independent samples). Statistical significance, as indicated by P -value, was determined using a two-tailed Student's t -test (***, $P < 0.001$).



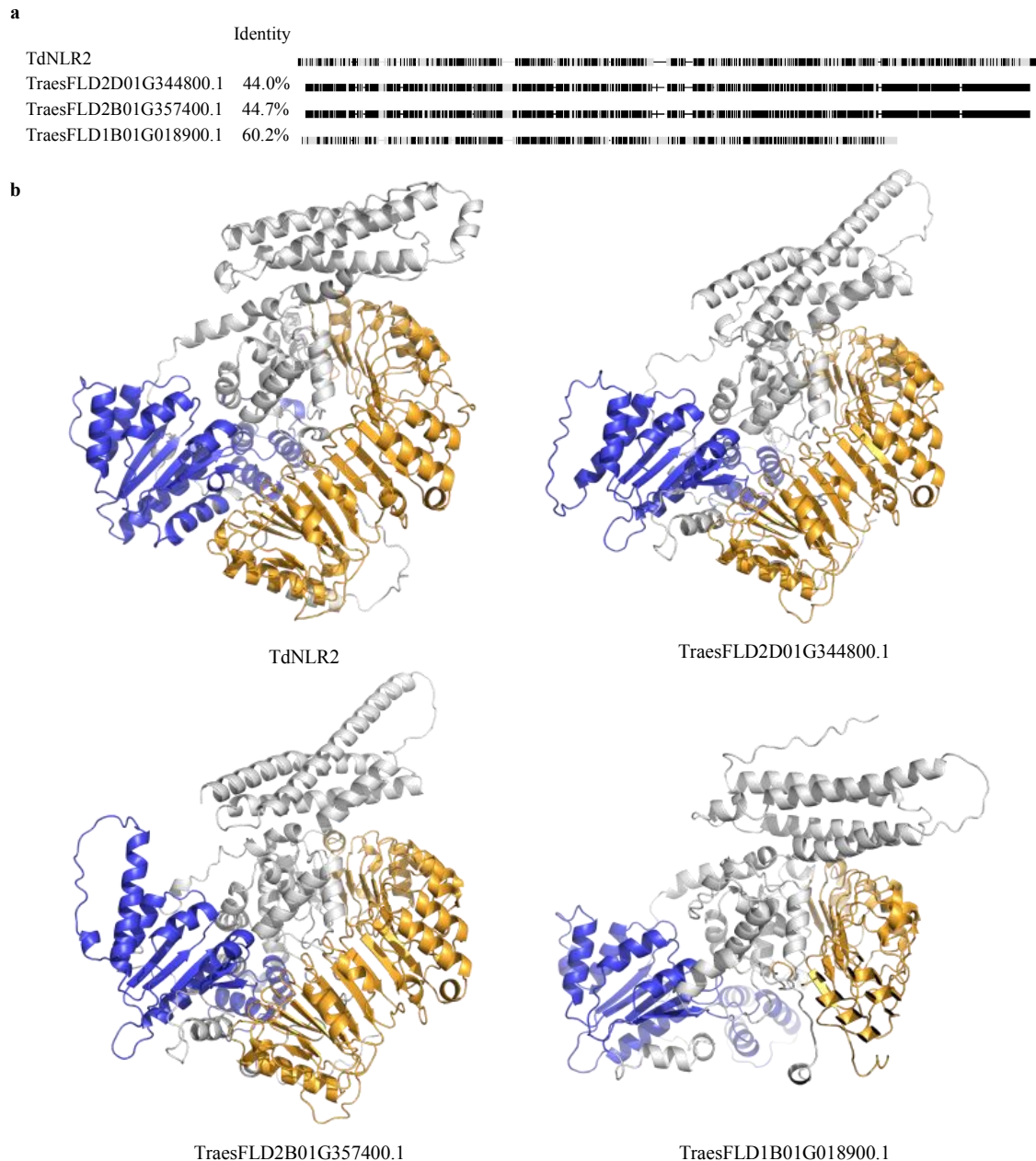
Supplementary Fig. 6 Validation of *TdNLR2* using VIGS.

a Target positions and sequences of *TdNLR2* in BSMV-VIGS experiments. **b** Phenotypes of HY-45 16 dpi with either BSMV:*TdNLR2-1as* or BSMV:*TdNLR2-2as* silencing constructs. Scale bar, 0.5 cm. **c** Mean percentage (%) leaf area covered in uredinia at 16 dpi; *TdNLR2*-silenced leaves were inoculated with *Pst* race CYR34. Error bars, mean $\pm$ SD ($n = 8$ biologically independent samples). Statistical significance, as indicated by *P*-value, was determined using a two-tailed Student's *t*-test (***, $P < 0.001$).



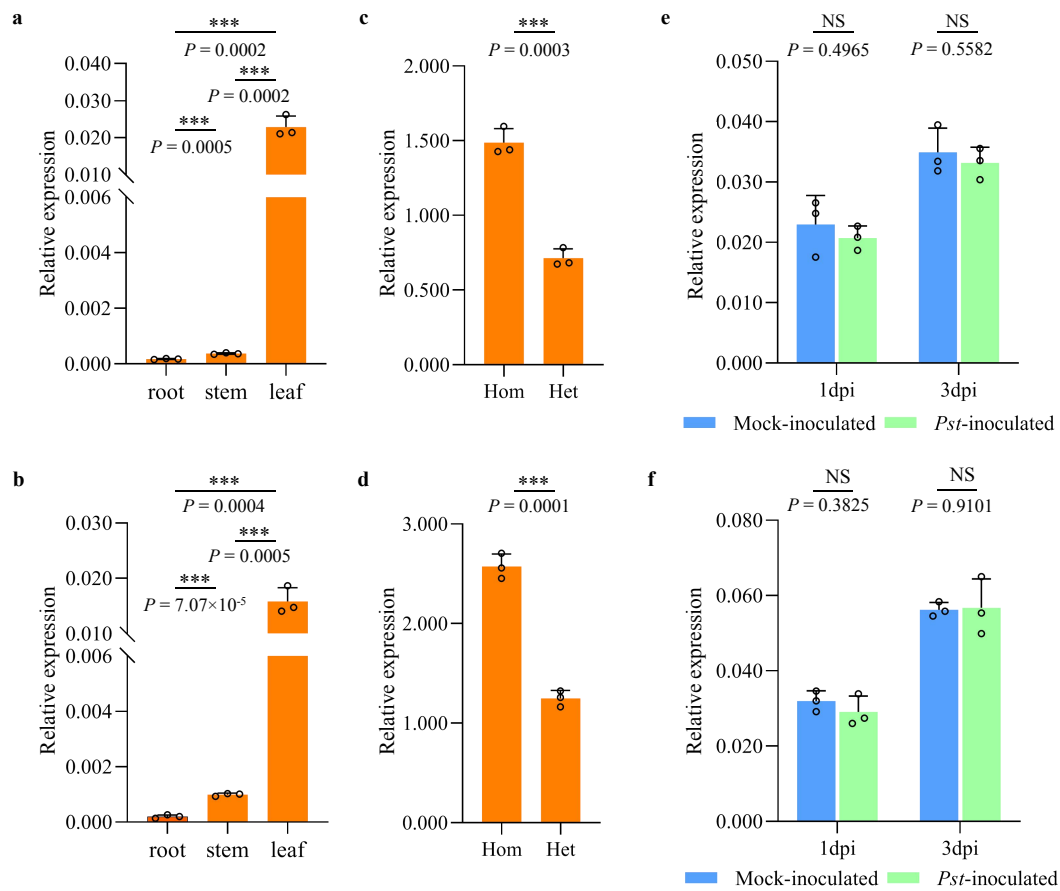
Supplementary Fig. 7 Validation of *TdNLR1* and *TdNLR2* by genetic transformation.

a Schematics of *TdNLR* constructs used for transformation. *ProUbi:TdNLR1*, coding sequence of *TdNLR1* driven by the maize *ubiquitin* (*Ubi*) promoter. *ProUbi:TdNLR2*, coding sequence of *TdNLR2* driven by the *Ubi* promoter. **b** T₁ transgenic plants expressing both *ProUbi:TdNLR1* and *ProUbi:TdNLR2* were resistant to *Pst* race CYR34. “+” and “-” signs below each leaf indicate the presence or absence of TD121-derived *TdNLR1* and *TdNLR2* transgenes. **c** *ProUbi:TdNLR1* T₁ transgenic plants were resistant to *Pst* race CYR34. Transcript levels of *TdNLR1* in transgenic plants determined by RT-qPCR. **d** *ProUbi:TdNLR2* T₁ transgenic plants were susceptible to *Pst* race CYR34. Transcript levels of *TdNLR2* in transgenic plants were quantified by RT-qPCR. Error bars in **c** and **d**, mean $\pm$ SD ($n=3$ technical replicates from a single plant). Scale bar, 0.5 cm.



Supplementary Fig. 8 Identification of *TdNLR2*-like homologs in Fielder reference genome.

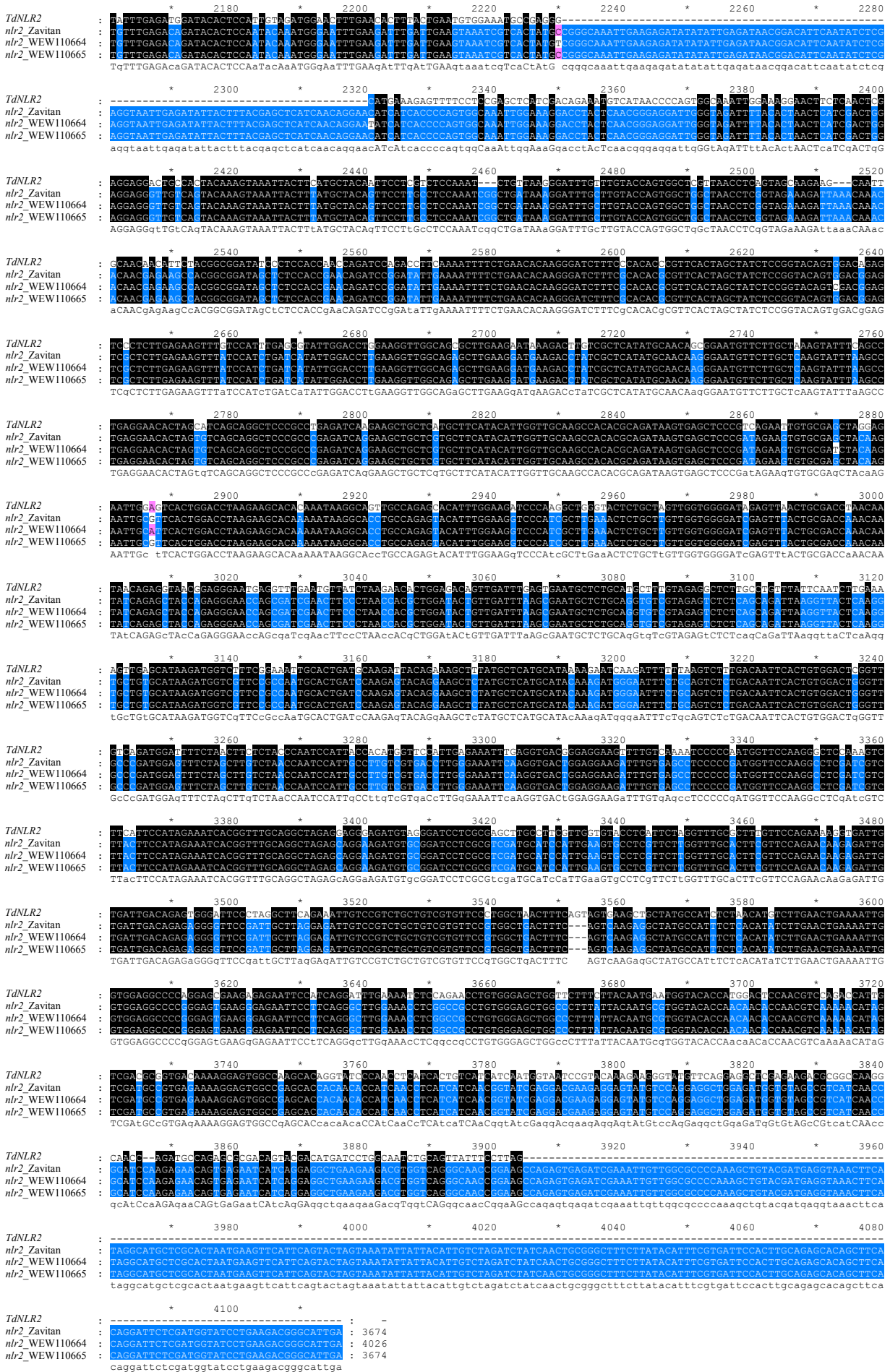
a Three *TdNLR2*-like homologous sequences (TraesFLD2D01G344800.1, TraesFLD2B01G357400.1, and TraesFLD1B01G018900.1) were identified in the Fielder reference genome. Amino acid identity was determined by comparing the homologous sequences with that of TdNLR2. **b** 3D structures of TdNLR2, TraesFLD2D01G344800.1, TraesFLD2B01G357400.1, and TraesFLD1B01G018900.1 proteins predicted using AlphaFold3 (<https://golgi.sandbox.google.com/>) and visualized by PyMOL (<https://pymol.org/2/>). Gray, blue, and yellow represent unknown function (Un), NB-ARC, and LRR domains.



Supplementary Fig. 9 Tissue-specific expression patterns of *TdNLR1* and *TdNLR2*.

a–b Transcript levels of *TdNLR1* (a) and *TdNLR2* (b) in root, stem, and leaf tissues of HY-187, estimated using RT-qPCR. **c–d** Transcript levels of *TdNLR1* (c) and *TdNLR2* (d) in plants homozygous (Hom) or heterozygous (Het) for *YrTD121*. **e–f** Transcript levels of *TdNLR1* (e) and *TdNLR2* (f) in mock-inoculated or *Pst*-inoculated HY-187 plants. Each bar represents the mean $\pm$ SD ($n = 3$ biologically independent samples, each sample has three technical replicates). Statistical significance, as indicated by P -value, was determined using a two-tailed Student's t -test (***, $P < 0.001$; NS, not significant).

[illegible]



Supplementary Fig. 10 Alignments of *nlr2* allelic sequences.

Identical sequences are highlighted in shading.

TdNLR1 : ATGGAACCTCGTCTGCGGCGCTTCaAGCAACGCCGTAAATCaCTcGtGGCAAGCTGGAAGGCTCCTCGCCAGGAGTACACCTaATCCAGGcGTCTCCAGcGACATCCAGTACATC : 120
nrl1_Zavitan : ATGGAACCTCGTCTGCGGCGCTTCaAGCAACGCCGTAAATCaCTcGtGGCAAGCTGGAAGGCTCCTCGCCAGGAGTACACCTaATCCAGGcGTCTCCAGcGACATCCAGTACATC : 120
nrl1_WEW110596 : ATGGAACCTCGTCTGCGGCGCTTCaAGCAACGCCGTAAATCaCTcGtGGCAAGCTGGAAGGCTCCTCGCCAGGAGTACACCTaATCCAGGcGTCTCCAGcGACATCCAGTACATC : 120
nrl1_WEW110776 : ATGGAACCTCGTCTGCGGCGCTTCaAGCAACGCCGTAAATCaCTcGtGGCAAGCTGGAAGGCTCCTCGCCAGGAGTACACCTaATCCAGGcGTCTCCAGcGACATCCAGTACATC : 120
nrl1_WEW110869 : ATGGAACCTCGTCTGCGGCGCTTCaAGCAACGCCGTAAATCaCTcGtGGCAAGCTGGAAGGCTCCTCGCCAGGAGTACACCTaATCCAGGcGTCTCCAGcGACATCCAGTACATC : 120

TdNLR1 : AACGAcGAGCTcGGAGCATcCAAGCCTTCATCAACAAGcAAGCGAGAGcCGcGGCCACAGcATcCAGAGcGATcGCTGGATGAAGcGGTGGcGAGGTGTCTATGAATGA : 240
nrl1_Zavitan : AACGAcGAGCTcGGAGCATcCAAGCCTTCATCAACAAGcAAGCGAGAGcCGcGGCCACAGcATcCAGAGcGATcGCTGGATGAAGcGGTGGcGAGGTGTCTATGAATGA : 237
nrl1_WEW110596 : AACGAcGAGCTcGGAGCATcCAAGCCTTCATCAACAAGcAAGCGAGAGcCGcGGCCACAGcATcCAGAGcGATcGCTGGATGAAGcGGTGGcGAGGTGTCTATGAATGA : 237
nrl1_WEW110776 : AACGAcGAGCTcGGAGCATcCAAGCCTTCATCAACAAGcAAGCGAGAGcCGcGGCCACAGcATcCAGAGcGATcGCTGGATGAAGcGGTGGcGAGGTGTCTATGAATGA : 237
nrl1_WEW110869 : AACGAcGAGCTcGGAGCATcCAAGCCTTCATCAACAAGcAAGCGAGAGcCGcGGCCACAGcATcCAGAGcGATcGCTGGATGAAGcGGTGGcGAGGTGTCTATGAATGA : 237

TdNLR1 : GATTGCATCGAcGAGCTcCAACcACCGcCTCGGCCcAGAGcCGCGGGGAAGCGcAGCTGATtTACTCTCGGAAGAAGTGTACTCTCTCAAAcGCTATGcCGCGCGcGATCGCT : 360
nrl1_Zavitan : GATTGCATCGAcGAGCTcCAACcACCGcCTCGGCCcAGAGcCGCGGGGAAGCGcAGCTGATtTACTCTCGGAAGAAGTGTACTCTCTCAAAcGCTATGcCGCGCGcGATCGCT : 357
nrl1_WEW110596 : GATTGCATCGAcGAGCTcCAACcACCGcCTCGGCCcAGAGcCGCGGGGAAGCGcAGCTGATtTACTCTCGGAAGAAGTGTACTCTCTCAAAcGCTATGcCGCGCGcGATCGCT : 357
nrl1_WEW110776 : GATTGCATCGAcGAGCTcCAACcACCGcCTCGGCCcAGAGcCGCGGGGAAGCGcAGCTGATtTACTCTCGGAAGAAGTGTACTCTCTCAAAcGCTATGcCGCGCGcGATCGCT : 357
nrl1_WEW110869 : GATTGCATCGAcGAGCTcCAACcACCGcCTCGGCCcAGAGcCGCGGGGAAGCGcAGCTGATtTACTCTCGGAAGAAGTGTACTCTCTCAAAcGCTATGcCGCGCGcGATCGCT : 357

TdNLR1 : GCGCAcATCCGAACCTcCAAGcTCCGGGGcGAGcAGCTcGAGcGACCGCGcCACCAGTtGGGCTcGAGAACCTTtAGGGGGcGGcGGCAGcAGcGAAGATGAcCTCCcAGcGAG : 480
nrl1_Zavitan : GCGCAcATCCGAACCTcCAAGcTCCGGGGcGAGcAGCTcGAGcGACCGCGcCACCAGTtGGGCTcGAGAACCTTtAGGGGGcGGcGGCAGcAGcGAAGATGAcCTCCcAGcGAG : 471
nrl1_WEW110596 : GCGCAcATCCGAACCTcCAAGcTCCGGGGcGAGcAGCTcGAGcGACCGCGcCACCAGTtGGGCTcGAGAACCTTtAGGGGGcGGcGGCAGcAGcGAAGATGAcCTCCcAGcGAG : 471
nrl1_WEW110776 : GCGCAcATCCGAACCTcCAAGcTCCGGGGcGAGcAGCTcGAGcGACCGCGcCACCAGTtGGGCTcGAGAACCTTtAGGGGGcGGcGGCAGcAGcGAAGATGAcCTCCcAGcGAG : 471
nrl1_WEW110869 : GCGCAcATCCGAACCTcCAAGcTCCGGGGcGAGcAGCTcGAGcGACCGCGcCACCAGTtGGGCTcGAGAACCTTtAGGGGGcGGcGGCAGcAGcGAAGATGAcCTCCcAGcGAG : 471

TdNLR1 : CGTGTAACCCCTcCAGcCTGAGCTcATTGcGACCAAGcAGcCTGTGGcGTAGAGGATGcCAATGGcGAGCTTGGAATGGTTTCAAGAGGGGAATcAGATcAGATtACCCcAAGAGGTTCT : 600
nrl1_Zavitan : CGTGTAACCCCTcCAGcCTGAGCTcATTGcGACCAAGcAGcCTGTGGcGTAGAGGATGcCAATGGcGAGCTTGGAATGGTTTCAAGAGGGGAATcAGATcAGATtACCCcAAGAGGTTCT : 588
nrl1_WEW110596 : CGTGTAACCCCTcCAGcCTGAGCTcATTGcGACCAAGcAGcCTGTGGcGTAGAGGATGcCAATGGcGAGCTTGGAATGGTTTCAAGAGGGGAATcAGATcAGATtACCCcAAGAGGTTCT : 588
nrl1_WEW110776 : CGTGTAACCCCTcCAGcCTGAGCTcATTGcGACCAAGcAGcCTGTGGcGTAGAGGATGcCAATGGcGAGCTTGGAATGGTTTCAAGAGGGGAATcAGATcAGATtACCCcAAGAGGTTCT : 588
nrl1_WEW110869 : CGTGTAACCCCTcCAGcCTGAGCTcATTGcGACCAAGcAGcCTGTGGcGTAGAGGATGcCAATGGcGAGCTTGGAATGGTTTCAAGAGGGGAATcAGATcAGATtACCCcAAGAGGTTCT : 588

TdNLR1 : GCCATTGAcGATTGGTGGCGCTcGGcAAGACcACCGCTCGCCCTGGAGTTGTACCcGAAGTtGGAGATGGATTtGACTGcAGAGcGcTCTGTcGAGGTCTcGcAGAAGTTCGATCTcGTA : 720
nrl1_Zavitan : GCCATTGAcGATTGGTGGCGCTcGGcAAGACcACCGCTCGCCCTGGAGTTGTACCcGAAGTtGGAGATGGATTtGACTGcAGAGcGcTCTGTcGAGGTCTcGcAGAAGTTCGATCTcGTA : 708
nrl1_WEW110596 : GCCATTGAcGATTGGTGGCGCTcGGcAAGACcACCGCTCGCCCTGGAGTTGTACCcGAAGTtGGAGATGGATTtGACTGcAGAGcGcTCTGTcGAGGTCTcGcAGAAGTTCGATCTcGTA : 708
nrl1_WEW110776 : GCCATTGAcGATTGGTGGCGCTcGGcAAGACcACCGCTCGCCCTGGAGTTGTACCcGAAGTtGGAGATGGATTtGACTGcAGAGcGcTCTGTcGAGGTCTcGcAGAAGTTCGATCTcGTA : 708
nrl1_WEW110869 : GCCATTGAcGATTGGTGGCGCTcGGcAAGACcACCGCTCGCCCTGGAGTTGTACCcGAAGTtGGAGATGGATTtGACTGcAGAGcGcTCTGTcGAGGTCTcGcAGAAGTTCGATCTcGTA : 708

TdNLR1 : ATGCTTTcGcGAGcCTTtCATcCAGTTCcGTGAGcAGcAGGCTcAcATGATTtCTCTGcAcAGTtGAGGAATtGGGAATtGGGAATtGAGTcCAAAcAGATGTCTGCTcGcCAATTtAAAG : 840
nrl1_Zavitan : ATGCTTTcGcGAGcCTTtCATcCAGTTCcGTGAGcAGcAGGCTcAcATGATTtCTCTGcAcAGTtGAGGAATtGGGAATtGGGAATtGAGTcCAAAcAGATGTCTGCTcGcCAATTtAAAG : 828
nrl1_WEW110596 : ATGCTTTcGcGAGcCTTtCATcCAGTTCcGTGAGcAGcAGGCTcAcATGATTtCTCTGcAcAGTtGAGGAATtGGGAATtGGGAATtGAGTcCAAAcAGATGTCTGCTcGcCAATTtAAAG : 828
nrl1_WEW110776 : ATGCTTTcGcGAGcCTTtCATcCAGTTCcGTGAGcAGcAGGCTcAcATGATTtCTCTGcAcAGTtGAGGAATtGGGAATtGGGAATtGAGTcCAAAcAGATGTCTGCTcGcCAATTtAAAG : 828
nrl1_WEW110869 : ATGCTTTcGcGAGcCTTtCATcCAGTTCcGTGAGcAGcAGGCTcAcATGATTtCTCTGcAcAGTtGAGGAATtGGGAATtGGGAATtGAGTcCAAAcAGATGTCTGCTcGcCAATTtAAAG : 828

TdNLR1 : GAGAAGAGGTATAAATTTTGcCTcTACTAGcTAATtTACcATTTTcTcATcTCTGcACGAcATtCTGCTtTCTAGTtTACtCCCGcCAATcATATtTtGACACAcATcTTAGcACAA : 960
nrl1_Zavitan : GAGAAGAGGTATAAATTTTGcCTcTACTAGcTAATtTACcATTTTcTcATcTCTGcACGAcATtCTGCTtTCTAGTtTACtCCCGcCAATcATATtTtGACACAcATcTTAGcACAA : 947
nrl1_WEW110596 : GAGAAGAGGTATAAATTTTGcCTcTACTAGcTAATtTACcATTTTcTcATcTCTGcACGAcATtCTGCTtTCTAGTtTACtCCCGcCAATcATATtTtGACACAcATcTTAGcACAA : 947
nrl1_WEW110776 : GAGAAGAGGTATAAATTTTGcCTcTACTAGcTAATtTACcATTTTcTcATcTCTGcACGAcATtCTGCTtTCTAGTtTACtCCCGcCAATcATATtTtGACACAcATcTTAGcACAA : 947
nrl1_WEW110869 : GAGAAGAGGTATAAATTTTGcCTcTACTAGcTAATtTACcATTTTcTcATcTCTGcACGAcATtCTGCTtTCTAGTtTACtCCCGcCAATcATATtTtGACACAcATcTTAGcACAA : 947

TdNLR1 : GATTAAAGAGGTATAAATTTTGcCTcTACTAAcTtTccATTTcATTtTctcTaaaaacattgtATgTCTGgTAgTcctTTCaTataTtGAAcTcctCTCAAAcAGATGTCTGCTcGcCAATTtAqAA : 1080
nrl1_Zavitan : GATTAAAGAGGTATAAATTTTGcCTcTACTAAcTtTccATTTcATTtTctcTaaaaacattgtATgTCTGgTAgTcctTTCaTataTtGAAcTcctCTCAAAcAGATGTCTGCTcGcCAATTtAqAA : 1063
nrl1_WEW110596 : GATTAAAGAGGTATAAATTTTGcCTcTACTAAcTtTccATTTcATTtTctcTaaaaacattgtATgTCTGgTAgTcctTTCaTataTtGAAcTcctCTCAAAcAGATGTCTGCTcGcCAATTtAqAA : 1063
nrl1_WEW110776 : GATTAAAGAGGTATAAATTTTGcCTcTACTAAcTtTccATTTcATTtTctcTaaaaacattgtATgTCTGgTAgTcctTTCaTataTtGAAcTcctCTCAAAcAGATGTCTGCTcGcCA

TdNLR1 : ACTTGAGAA* 1940 * 1960 * 1980 * 2000 * 2020 * 2040 : 2016
nrl1_Zavitan : ACTTGAGAA* 1940 * 1960 * 1980 * 2000 * 2020 * 2040 : 1754
nrl1_WEW110596 : ACTTGAGAA* 1940 * 1960 * 1980 * 2000 * 2020 * 2040 : 1754
nrl1_WEW110776 : ACTTGAGAA* 1940 * 1960 * 1980 * 2000 * 2020 * 2040 : 1754
nrl1_WEW110869 : ACTTGAGAA* 1940 * 1960 * 1980 * 2000 * 2020 * 2040 : 1754

ACTTGAGAA* 1940 * 1960 * 1980 * 2000 * 2020 * 2040 : 1754
 ACTTGAGAA* 1940 * 1960 * 1980 * 2000 * 2020 * 2040 : 1754
 ACTTGAGAA* 1940 * 1960 * 1980 * 2000 * 2020 * 2040 : 1754
 ACTTGAGAA* 1940 * 1960 * 1980 * 2000 * 2020 * 2040 : 1754

TdNLR1 : AGGGTTCATTATTGAGAAGATGGcAGAC* 2060 * 2080 * 2100 * 2120 * 2140 * 2160 : 2136
nrl1_Zavitan : AGGGTTCATTATTGAGAAGATGGcAGAC* 2060 * 2080 * 2100 * 2120 * 2140 * 2160 : 1874
nrl1_WEW110596 : AGGGTTCATTATTGAGAAGATGGcAGAC* 2060 * 2080 * 2100 * 2120 * 2140 * 2160 : 1874
nrl1_WEW110776 : AGGGTTCATTATTGAGAAGATGGcAGAC* 2060 * 2080 * 2100 * 2120 * 2140 * 2160 : 1874
nrl1_WEW110869 : AGGGTTCATTATTGAGAAGATGGcAGAC* 2060 * 2080 * 2100 * 2120 * 2140 * 2160 : 1874

AGGGTTCATTATTGAGAAGATGGcAGAC* 2060 * 2080 * 2100 * 2120 * 2140 * 2160 : 1874
 AGGGTTCATTATTGAGAAGATGGcAGAC* 2060 * 2080 * 2100 * 2120 * 2140 * 2160 : 1874
 AGGGTTCATTATTGAGAAGATGGcAGAC* 2060 * 2080 * 2100 * 2120 * 2140 * 2160 : 1874
 AGGGTTCATTATTGAGAAGATGGcAGAC* 2060 * 2080 * 2100 * 2120 * 2140 * 2160 : 1874

TdNLR1 : GAACTATCA* 2180 * 2200 * 2220 * 2240 * 2260 * 2280 : 2256
nrl1_Zavitan : GAACTATCA* 2180 * 2200 * 2220 * 2240 * 2260 * 2280 : 1994
nrl1_WEW110596 : GAACTATCA* 2180 * 2200 * 2220 * 2240 * 2260 * 2280 : 1994
nrl1_WEW110776 : GAACTATCA* 2180 * 2200 * 2220 * 2240 * 2260 * 2280 : 1994
nrl1_WEW110869 : GAACTATCA* 2180 * 2200 * 2220 * 2240 * 2260 * 2280 : 1994

GAACTATCA* 2180 * 2200 * 2220 * 2240 * 2260 * 2280 : 1994
 GAACTATCA* 2180 * 2200 * 2220 * 2240 * 2260 * 2280 : 1994
 GAACTATCA* 2180 * 2200 * 2220 * 2240 * 2260 * 2280 : 1994
 GAACTATCA* 2180 * 2200 * 2220 * 2240 * 2260 * 2280 : 1994

TdNLR1 : GTTCTCTGTC* 2300 * 2320 * 2340 * 2360 * 2380 * 2400 : 2376
nrl1_Zavitan : GTTCTCTGTC* 2300 * 2320 * 2340 * 2360 * 2380 * 2400 : 2114
nrl1_WEW110596 : GTTCTCTGTC* 2300 * 2320 * 2340 * 2360 * 2380 * 2400 : 2114
nrl1_WEW110776 : GTTCTCTGTC* 2300 * 2320 * 2340 * 2360 * 2380 * 2400 : 2114
nrl1_WEW110869 : GTTCTCTGTC* 2300 * 2320 * 2340 * 2360 * 2380 * 2400 : 2114

GTTCTCTGTC* 2300 * 2320 * 2340 * 2360 * 2380 * 2400 : 2114
 GTTCTCTGTC* 2300 * 2320 * 2340 * 2360 * 2380 * 2400 : 2114
 GTTCTCTGTC* 2300 * 2320 * 2340 * 2360 * 2380 * 2400 : 2114
 GTTCTCTGTC* 2300 * 2320 * 2340 * 2360 * 2380 * 2400 : 2114

TdNLR1 : GATATTCA* 2420 * 2440 * 2460 * 2480 * 2500 * 2520 : 2496
nrl1_Zavitan : GATATTCA* 2420 * 2440 * 2460 * 2480 * 2500 * 2520 : 2234
nrl1_WEW110596 : GATATTCA* 2420 * 2440 * 2460 * 2480 * 2500 * 2520 : 2234
nrl1_WEW110776 : GATATTCA* 2420 * 2440 * 2460 * 2480 * 2500 * 2520 : 2234
nrl1_WEW110869 : GATATTCA* 2420 * 2440 * 2460 * 2480 * 2500 * 2520 : 2234

GATATTCA* 2420 * 2440 * 2460 * 2480 * 2500 * 2520 : 2234
 GATATTCA* 2420 * 2440 * 2460 * 2480 * 2500 * 2520 : 2234
 GATATTCA* 2420 * 2440 * 2460 * 2480 * 2500 * 2520 : 2234
 GATATTCA* 2420 * 2440 * 2460 * 2480 * 2500 * 2520 : 2234

TdNLR1 : ACCATCCCA* 2540 * 2560 * 2580 * 2600 * 2620 * 2640 : 2616
nrl1_Zavitan : ACCATCCCA* 2540 * 2560 * 2580 * 2600 * 2620 * 2640 : 2354
nrl1_WEW110596 : ACCATCCCA* 2540 * 2560 * 2580 * 2600 * 2620 * 2640 : 2352
nrl1_WEW110776 : ACCATCCCA* 2540 * 2560 * 2580 * 2600 * 2620 * 2640 : 2352
nrl1_WEW110869 : ACCATCCCA* 2540 * 2560 * 2580 * 2600 * 2620 * 2640 : 2354

ACCATCCCA* 2540 * 2560 * 2580 * 2600 * 2620 * 2640 : 2354
 ACCATCCCA* 2540 * 2560 * 2580 * 2600 * 2620 * 2640 : 2352
 ACCATCCCA* 2540 * 2560 * 2580 * 2600 * 2620 * 2640 : 2352
 ACCATCCCA* 2540 * 2560 * 2580 * 2600 * 2620 * 2640 : 2354

TdNLR1 : TAAAAGCA* 2660 * 2680 * 2700 * 2720 * 2740 * 2760 : 2736
nrl1_Zavitan : TAAAAGCA* 2660 * 2680 * 2700 * 2720 * 2740 * 2760 : 2468
nrl1_WEW110596 : TAAAAGCA* 2660 * 2680 * 2700 * 2720 * 2740 * 2760 : 2462
nrl1_WEW110776 : TAAAAGCA* 2660 * 2680 * 2700 * 2720 * 2740 * 2760 : 2468
nrl1_WEW110869 : TAAAAGCA* 2660 * 2680 * 2700 * 2720 * 2740 * 2760 : 2474

TAAAAGCA* 2660 * 2680 * 2700 * 2720 * 2740 * 2760 : 2468
 TAAAAGCA* 2660 * 2680 * 2700 * 2720 * 2740 * 2760 : 2462
 TAAAAGCA* 2660 * 2680 * 2700 * 2720 * 2740 * 2760 : 2468
 TAAAAGCA* 2660 * 2680 * 2700 * 2720 * 2740 * 2760 : 2474

TdNLR1 : ATTCAAG* 2780 * 2800 * 2820 * 2840 * 2860 * 2880 : 2856
nrl1_Zavitan : ATTCAAG* 2780 * 2800 * 2820 * 2840 * 2860 * 2880 : 2588
nrl1_WEW110596 : ATTCAAG* 2780 * 2800 * 2820 * 2840 * 2860 * 2880 : 2582
nrl1_WEW110776 : ATTCAAG* 2780 * 2800 * 2820 * 2840 * 2860 * 2880 : 2588
nrl1_WEW110869 : ATTCAAG* 2780 * 2800 * 2820 * 2840 * 2860 * 2880 : 2594

ATTCAAG* 2780 * 2800 * 2820 * 2840 * 2860 * 2880 : 2588
 ATTCAAG* 2780 * 2800 * 2820 * 2840 * 2860 * 2880 : 2582
 ATTCAAG* 2780 * 2800 * 2820 * 2840 * 2860 * 2880 : 2588
 ATTCAAG* 2780 * 2800 * 2820 * 2840 * 2860 * 2880 : 2594

TdNLR1 : ATTGCTCT* 2900 * 2920 * 2940 * 2960 * 2980 * 3000 : 2976
nrl1_Zavitan : ATTGCTCT* 2900 * 2920 * 2940 * 2960 * 2980 * 3000 : 2708
nrl1_WEW110596 : ATTGCTCT* 2900 * 2920 * 2940 * 2960 * 2980 * 3000 : 2702
nrl1_WEW110776 : ATTGCTCT* 2900 * 2920 * 2940 * 2960 * 2980 * 3000 : 2708
nrl1_WEW110869 : ATTGCTCT* 2900 * 2920 * 2940 * 2960 * 2980 * 3000 : 2714

ATTGCTCT* 2900 * 2920 * 2940 * 2960 * 2980 * 3000 : 2708
 ATTGCTCT* 2900 * 2920 * 2940 * 2960 * 2980 * 3000 : 2702
 ATTGCTCT* 2900 * 2920 * 2940 * 2960 * 2980 * 3000 : 2708
 ATTGCTCT* 2900 * 2920 * 2940 * 2960 * 2980 * 3000 : 2714

TdNLR1 : GCACAC* 3020 * 3040 * 3060 * 3080 * 3100 * 3120 : 3096
nrl1_Zavitan : GCACAC* 3020 * 3040 * 3060 * 3080 * 3100 * 3120 : 2828
nrl1_WEW110596 : GCACAC* 3020 * 3040 * 3060 * 3080 * 3100 * 3120 : 2822
nrl1_WEW110776 : GCACAC* 3020 * 3040 * 3060 * 3080 * 3100 * 3120 : 2828
nrl1_WEW110869 : GCACAC* 3020 * 3040 * 3060 * 3080 * 3100 * 3120 : 2834

GCACAC* 3020 * 3040 * 3060 * 3080 * 3100 * 3120 : 2828
 GCACAC* 3020 * 3040 * 3060 * 3080 * 3100 * 3120 : 2822
 GCACAC* 3020 * 3040 * 3060 * 3080 * 3100 * 3120 : 2828
 GCACAC* 3020 * 3040 * 3060 * 3080 * 3100 * 3120 : 2834

TdNLR1 : TGC* 3140 * 3160 * 3180 * 3200 * 3220 * 3240 : 3216
nrl1_Zavitan : TGC* 3140 * 3160 * 3180 * 3200 * 3220 * 3240 : 2948
nrl1_WEW110596 : TGC* 3140 * 3160 * 3180 * 3200 * 3220 * 3240 : 2942
nrl1_WEW110776 : TGC* 3140 * 3160 * 3180 * 3200 * 3220 * 3240 : 2948
nrl1_WEW110869 : TGC* 3140 * 3160 * 3180 * 3200 * 3220 * 3240 : 2948

TGC* 3140 * 3160 * 3180 * 3200 * 3220 * 3240 : 2948
 TGC* 3140 * 3160 * 3180 * 3200 * 3220 * 3240 : 2942
 TGC* 3140 * 3160 * 3180 * 3200 * 3220 * 3240 : 2948
 TGC* 3140 * 3160 * 3180 * 3200 * 3220 * 3240 : 2948

TdNLR1 : GC* 3260 * 3280 * 3300 * 3320 * 3340 * 3360 : 3336
nrl1_Zavitan : GC* 3260 * 3280 * 3300 * 3320 * 3340 * 3360 : 3067
nrl1_WEW110596 : GC* 3260 * 3280 * 3300 * 3320 * 3340 * 3360 : 3061
nrl1_WEW110776 : GC* 3260 * 3280 * 3300 * 3320 * 3340 * 3360 : 3067
nrl1_WEW110869 : GC* 3260 * 3280 * 3300 * 3320 * 3340 * 3360 : 3068

GC* 3260 * 3280 * 3300 * 3320 * 3340 * 3360 : 3067
 GC* 3260 * 3280 * 3300 * 3320 * 3340 * 3360 : 3061
 GC* 3260 * 3280 * 3300 * 3320 * 3340 * 3360 : 3067
 GC* 3260 * 3280 * 3300 * 3320 * 3340 * 3360 : 3068

TdNLR1 : TTC* 3380 * 3400 * 3420 * 3440 * 3460 * 3480 : 3456
nrl1_Zavitan : TTC* 3380 * 3400 * 3420 * 3440 * 3460 * 3480 : 3187
nrl1_WEW110596 : TTC* 3380 * 3400 * 3420 * 3440 * 3460 * 3480 : 3181
nrl1_WEW110776 : TTC* 3380 * 3400 * 3420 * 3440 * 3460 * 3480 : 3187
nrl1_WEW110869 : TTC* 3380 * 3400 * 3420 * 3440 * 3460 * 3480 : 3188

TTC* 3380 * 3400 * 3420 * 3440 * 3460 * 3480 : 3187
 TTC* 3380 * 3400 * 3420 * 3440 * 3460 * 3480 : 3181
 TTC* 3380 * 3400 * 3420 * 3440 * 3460 * 3480 : 3187
 TTC* 3380 * 3400 * 3420 * 3440 * 3460 * 3480 : 3188

TdNLR1 : CTC* 3500 * 3520 * 3540 * 3560 * 3580 * 3600 : 3511
nrl1_Zavitan : CTC* 3500 * 3520 * 3540 * 3560 * 3580 * 3600 : 3259
nrl1_WEW110596 : CTC* 3500 * 3520 * 3540 * 3560 * 3580 * 3600 : 3253
nrl1_WEW110776 : CTC* 3500 * 3520 * 3540 * 3560 * 3580 * 3600 : 3259
nrl1_WEW110869 : CTC* 3500 * 3520 * 3540 * 3560 * 3580 * 3600 : 3260

CTC* 3500 * 3520 * 3540 * 3560 * 3580 * 3600 : 3259
 CTC* 3500 * 3520 * 3540 * 3560 * 3580 * 3600 : 3253
 CTC* 3500 * 3520 * 3540 * 3560 * 3580 * 3600 : 3259
 CTC* 3500 * 3520 * 3540 * 3560 * 3580 * 3600 : 3260

cTCCCTcTGTgATCGT GATGgTACaAGaAtcTcgGcGgAGtaagtagagta ttgatgtga

Supplementary Fig. 1 Alignments of *nrl1* allelic sequences.

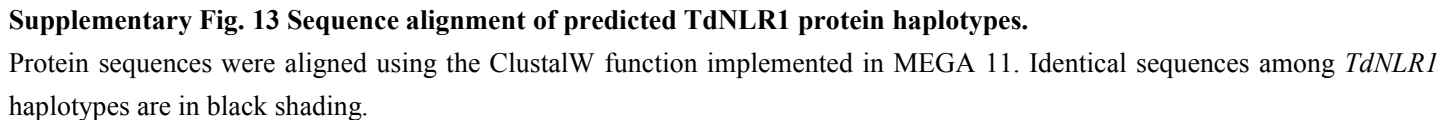
Identical sequences are highlighted in shading.

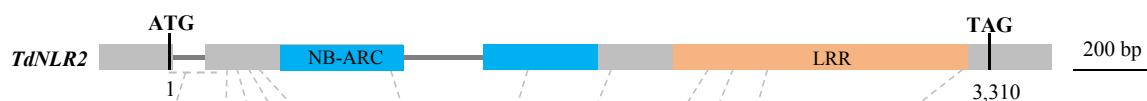


Position Haplotype	87-107	228	248	279	600	656	914	933	1030	1038	1257	2184	2264	2941	3192	3221	3262	3290	CDS_haplotypes	Protein types	GenBank numbers	Numbers
<i>TdNLR1_h1</i>	CATCCAGGGCGTCCATGACGA	C	T	G	T	G	C	A	T	C	G	T	G	T	T	A	A	G	TdNLR1-CDS_h1	TdNLR1_P1	OP948046	4
<i>TdNLR1_h2</i>	CATCCAGGGCGTCCATGACGA	C	T	G	T	G	C	C	T	C	G	C	G	T	T	G	A	G	TdNLR1-CDS_h2	TdNLR1_P2	OR472555	1
<i>TdNLR1_h3</i>	CATCCAGGGCGTCCATGACGA	T	T	G	T	G	C	C	T	C	G	C	G	T	T	A	A	G	TdNLR1-CDS_h3	TdNLR1_P3	OP948047	9
<i>TdNLR1_h4</i>	Deletion	C	C	G	T	G	C	C	C	C	G	C	G	A	C	A	A	G	TdNLR1-CDS_h4	TdNLR1_P4	OP948057	18
<i>TdNLR1_h5</i>	Deletion	C	C	G	T	G	C	C	C	C	A	C	G	A	C	A	A	G	TdNLR1-CDS_h4	TdNLR1_P4	OP948058	4
<i>TdNLR1_h6</i>	CATCCAGGGCGTCCATGACGA	C	T	G	C	C	C	A	T	C	G	C	A	T	T	A	A	G	TdNLR1-CDS_h5	TdNLR1_P5	OP948048	3
<i>TdNLR1_h7</i>	CATCCAGGGCGTCCATGACGA	C	T	A	T	G	C	C	T	C	G	C	G	T	T	A	A	G	TdNLR1-CDS_h6	TdNLR1_P3	OP948049	2
<i>TdNLR1_h8</i>	CATCCAGGGCGTCCATGACGA	C	T	G	T	G	A	C	C	C	G	C	G	A	C	A	A	G	TdNLR1-CDS_h7	TdNLR1_P6	OP948050	5
<i>TdNLR1_h9</i>	CATCCAGGGCGTCCATGACGA	C	T	G	T	G	C	C	T	C	G	T	G	T	T	A	A	A	TdNLR1-CDS_h8	TdNLR1_P7	OP948051	4
<i>TdNLR1_h10</i>	CATCCAGGGCGTCCATGACGA	C	T	G	T	G	C	C	T	C	G	C	G	T	T	A	A	A	TdNLR1-CDS_h9	TdNLR1_P8	OP948052	2
<i>TdNLR1_h11</i>	CATCCAGGGCGTCCATGACGA	C	T	G	T	G	C	C	T	C	G	T	G	T	T	A	C	A	TdNLR1-CDS_h10	TdNLR1_P7	OP948053	1
<i>TdNLR1_h12</i>	CATCCAGGGCGTCCATGACGA	C	T	G	C	G	C	A	T	C	G	C	A	T	T	A	A	G	TdNLR1-CDS_h11	TdNLR1_P9	OP948054	2
<i>TdNLR1_h13</i>	CATCCAGGGCGTCCATGACGA	C	T	G	T	G	C	C	T	T	G	C	G	T	T	A	A	G	TdNLR1-CDS_h12	TdNLR1_P3	OP948055	1
<i>TdNLR1_h14</i>	CATCCAGGGCGTCCATGACGA	C	T	G	T	G	A	C	C	C	G	C	G	T	C	A	A	G	TdNLR1-CDS_h13	TdNLR1_P10	OP948056	1
<i>TdNLR1_h15</i>	Deletion	C	C	G	T	G	C	C	C	C	G	T	G	T	C	A	A	G	TdNLR1-CDS_h14	TdNLR1_P11	OP948059	1
<i>TdNLR1_h16</i>	Deletion	C	C	G	T	G	C	C	T	C	G	T	G	T	C	A	A	G	TdNLR1-CDS_h14	TdNLR1_P11	OP948060	1

Supplementary Fig. 12 Haplotype analysis of *TdNLR1* from 59 wild emmer accessions.

Rectangular boxes represent exons and gray lines show introns of *TdNLR1*. Capital letters in red indicate nucleotide variations. Nucleotide positions located in exons are marked in gray shading.

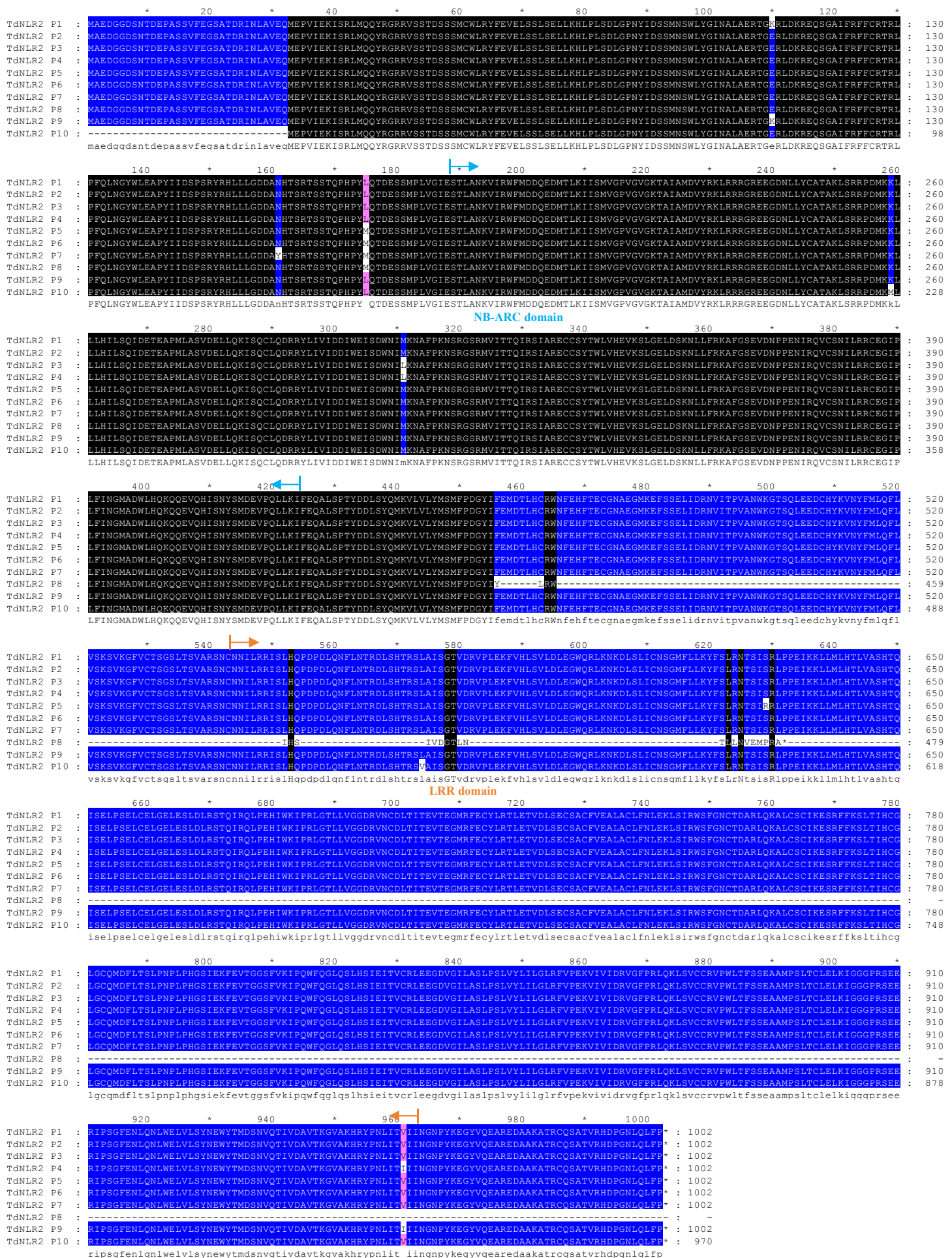




Position Haplotype	1-184	335	441	594	636	889	1232	1661	2021	2188	2374	3182	CDS_haplotypes	Protein types	GenBank numbers	Numbers
<i>TdNLR2_h1</i>	ATG...GGA	G	A	A	T	A	A	Deletion	C	C	A	G	TdNLR2-CDS_h1	TdNLR2_P1	PP874936	4
<i>TdNLR2_h2</i>	ATG...GGA	G	G	A	T	A	A	Deletion	C	C	A	G	TdNLR2-CDS_h2	TdNLR2_P2	PP874945	13
<i>TdNLR2_h3</i>	ATG...GGA	G	G	A	T	A	T	Deletion	C	C	A	G	TdNLR2-CDS_h3	TdNLR2_P3	PP874937	2
<i>TdNLR2_h4</i>	ATG...GGA	G	G	A	T	A	T	Deletion	C	C	A	A	TdNLR2-CDS_h4	TdNLR2_P4	PP874938	10
<i>TdNLR2_h5</i>	ATG...GGA	G	G	A	A	A	A	Deletion	C	A	A	G	TdNLR2-CDS_h5	TdNLR2_P5	PP874939	1
<i>TdNLR2_h6</i>	ATG...GGA	G	G	A	A	A	A	Deletion	C	C	A	G	TdNLR2-CDS_h6	TdNLR2_P6	PP874940	25
<i>TdNLR2_h7</i>	ATG...GGA	G	G	T	A	A	A	Deletion	C	C	A	G	TdNLR2-CDS_h7	TdNLR2_P7	PP874941	1
<i>TdNLR2_h8</i>	ATG...GGA	G	G	A	A	A	A	TA	C	C	A	G	TdNLR2-CDS_h8	TdNLR2_P8 *	PP874944	1
<i>TdNLR2_h9</i>	ATG...GGA	G	A	A	T	A	A	Deletion	C	C	A	A	TdNLR2-CDS_h9	TdNLR2_P9	PP874942	1
<i>TdNLR2_h10</i>	ATG...GGA	C	G	A	T	A	A	Deletion	C	C	A	G	TdNLR2-CDS_h10	TdNLR2_P2	PP874943	1
<i>TdNLR2_h11</i>	Deletion	G	G	A	T	T	A	Deletion	G	C	G	G	TdNLR2-CDS_h11	TdNLR2_P10	PQ412542	1

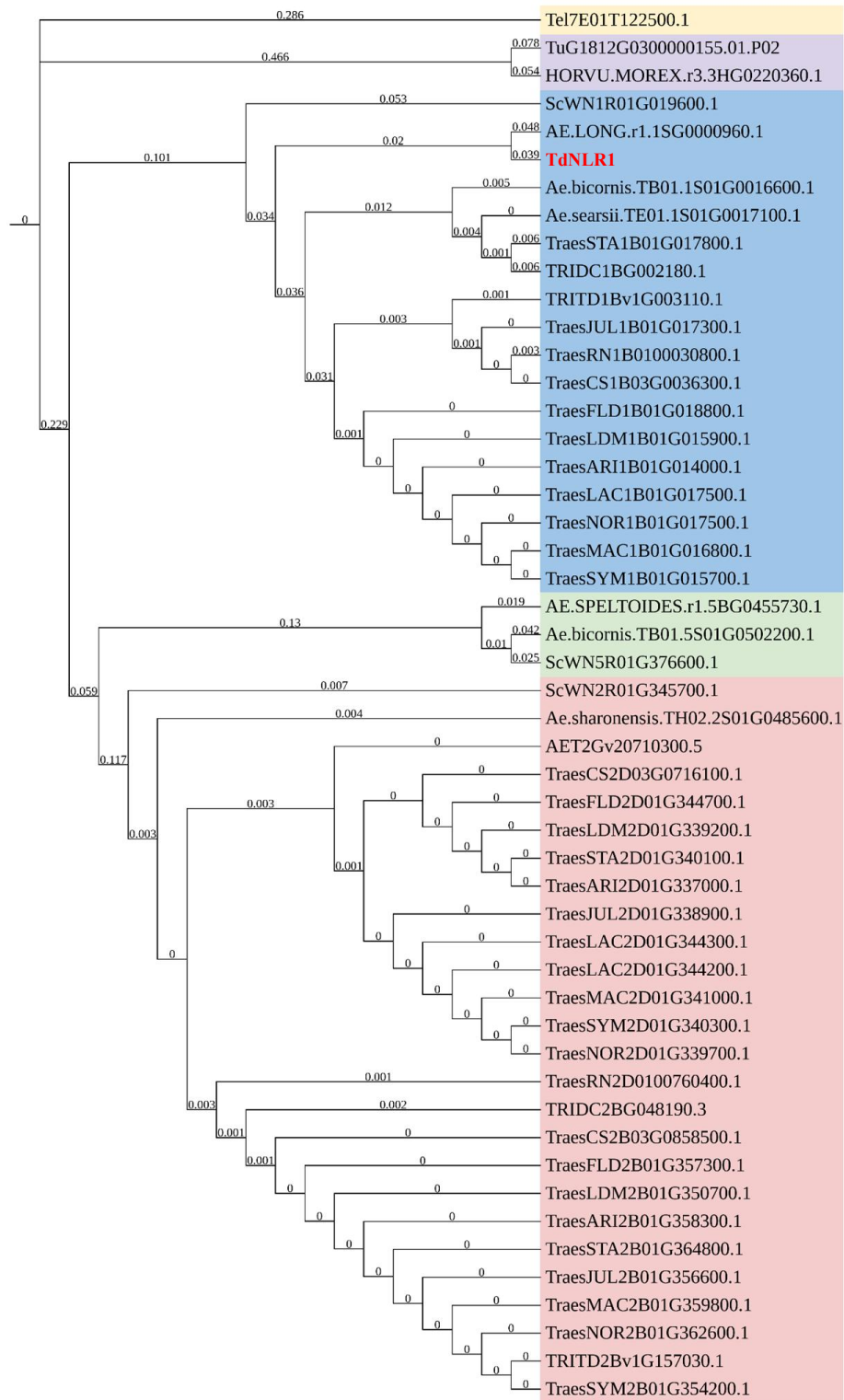
Supplementary Fig. 14 Haplotype analysis of *TdNLR2* from 60 wild emmer accessions.

Rectangular boxes represent exons and gray lines show introns of *TdNLR2*. Uppercase letters in red indicate nucleotide variations. Nucleotide positions located on exons are marked in gray shading. ATG...GGA represents a 184-nucleotide stretch of *TdNLR2* starting from ATG. Deletion of these 184 nucleotides in *TdNLR2_h11* leads to putative truncation of the protein. *, Premature termination of translation.



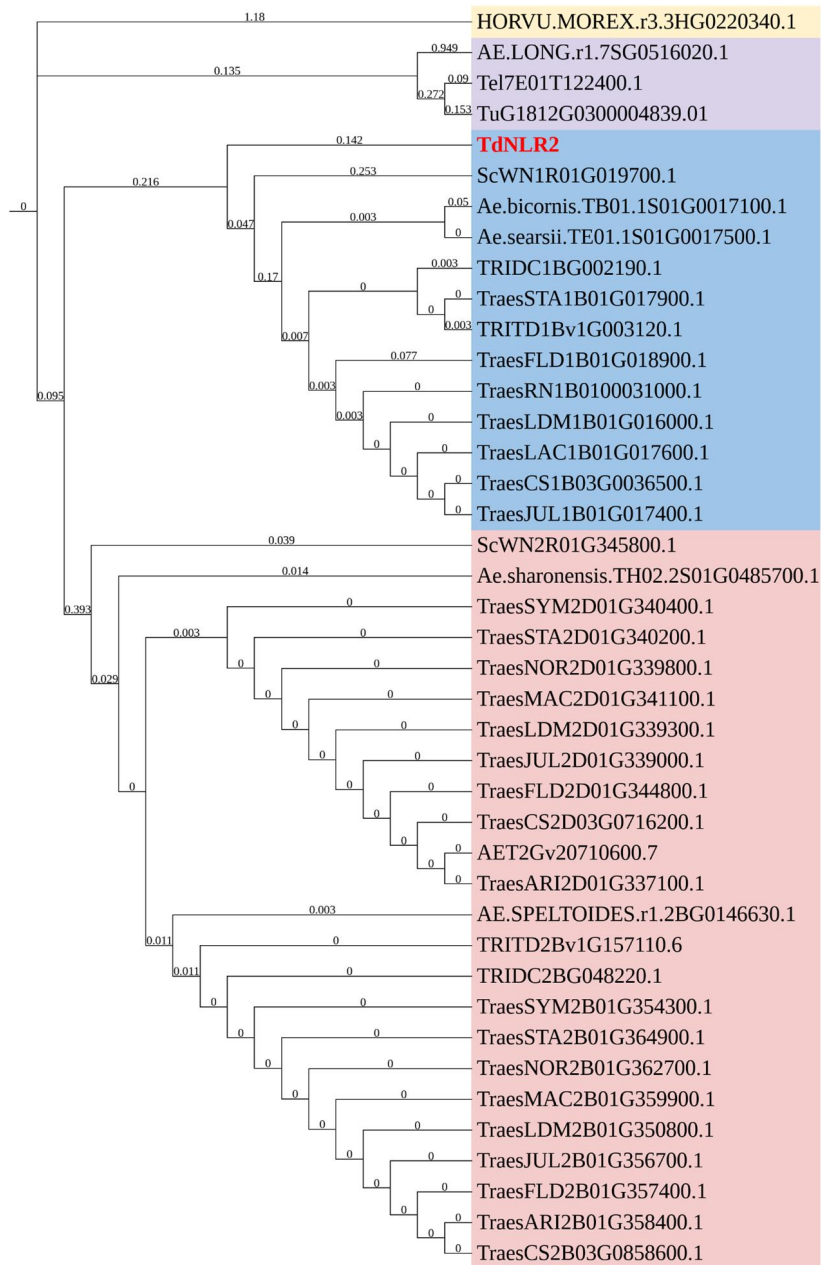
Supplementary Fig. 15 Sequence alignment of predicted TdNLR2 protein haplotypes.

Protein sequences aligned using the ClustalW function implemented in MEGA 11. Identical sequences among TdNLR2 haplotypes are highlighted in shading.



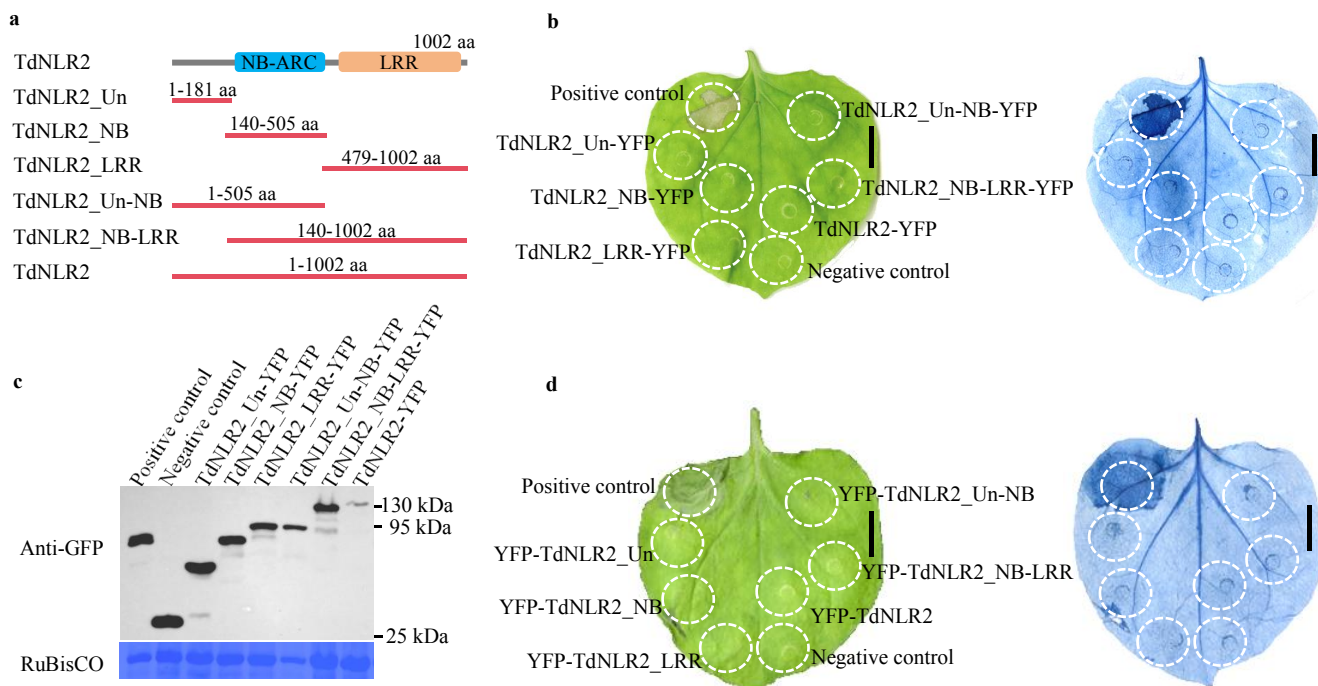
Supplementary Fig. 16 Phylogenetic analysis of TdNLR1 homologs from cereal species.

A rooted maximum-likelihood phylogenetic tree constructed using TdNLR1 and homologs from *T. dicoccoides* (Zavitan), *T. durum* (Svevo), *T. aestivum* (Chinese Spring, Fielder, CDC_Landmark, ArinaLrFor, LongReach_Lancer, Norin 61, SY_Mattis, Mace, Julius, Renan, and CDC_St Stanley), *Ae. searsii* (accession TE01), *Ae. longissima* (AEG-6782-2), *Ae. bicornis* (TB01), *Ae. sharonensis* (TH02), *Ae. tauschii* (AL8/78), *Ae. speltoides* (AEG-9674-1), *Th. elongatum* (D-3458), *H. vulgare* (Morex), *S. cereale* (Weining), and *T. urartu* (G1812) (Supplementary Table 9). Branches are labeled with protein IDs from the corresponding reference genome. Light blue, homologs on chromosome 1. Pink, homologs on chromosome 2. Light green, homologs on chromosome 5. Numbers on the branches represent the branch lengths.



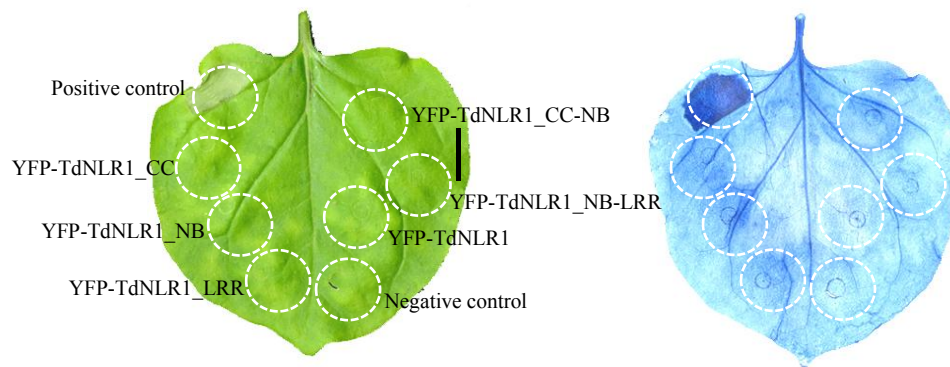
Supplementary Fig. 17 Phylogenetic analysis of TdNLR2 homologs from cereal species.

A rooted maximum-likelihood phylogenetic tree was constructed using TdNLR2 and homologs (see caption to Supplementary Fig. 16; Supplementary Table 10).



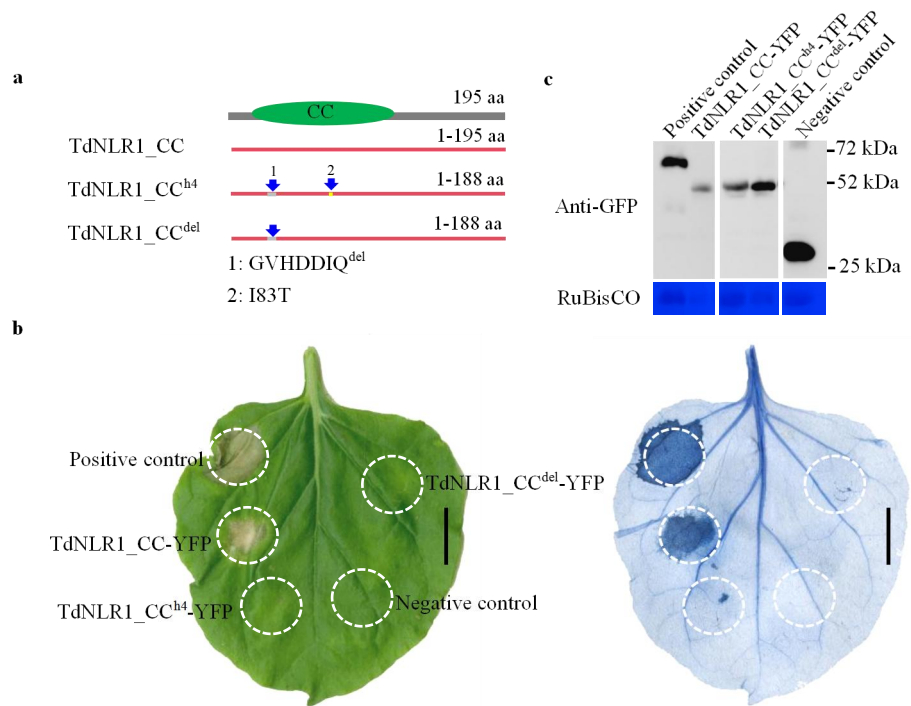
Supplementary Fig. 18 Assays of cell death in *N. benthamiana* leaves infiltrated with *Agrobacterium* carrying *TdNLR2* constructs.

a Schematic representing different domains of TdNLR2 (Un, NB, LRR, Un-NB, NB-LRR) and the full-length protein used in cell death assays in infiltrated *N. benthamiana* leaves. **b** Transient expression of different TdNLR2 domains in *N. benthamiana*. A YFP tag was fused to the C terminus of each domain. The infiltrated leaves were (left panel) stained with trypan blue (right panel). Cell death was observed at 72 hours post infiltration (hpi). **c** Western blotting analysis to detect the expression of constructs in *N. benthamiana* leaves. Coomassie brilliant blue staining of RuBisCO is shown below as loading control. **d** Transient expression of different TdNLR2 domains in *N. benthamiana*. A YFP tag was fused to the N terminus of each domain. Infiltrated leaves (left panel) were stained with trypan blue (right panel) to observe cell death at 72 hpi. Cell death was observed only in the positive control. Positive control, wheat TaZFP5B protein. Negative control, YFP. All experiments were repeated three times with similar results. Scale bar, 1cm.



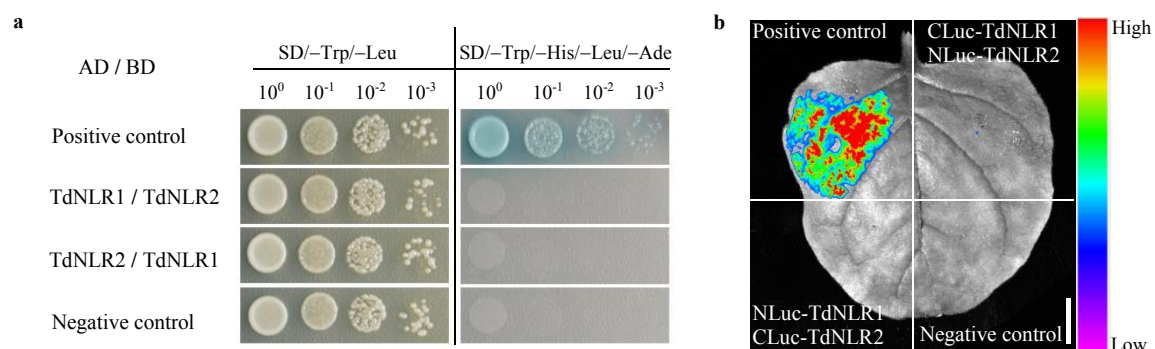
Supplementary Fig. 19 Cell death in *N. benthamiana* leaves infiltrated with *Agrobacterium* carrying TdNLR1 constructs.

Infiltrated leaves (left panel) were stained with trypan blue (right panel) to observe cell death at 72 hpi. A YFP tag was fused to the N terminus of the different TdNLR1 domains. Cell death was observed only in the positive control. Positive control, wheat TaZFP5B protein. Negative control, YFP. All experiments were repeated three times with similar results. Scale bar, 1cm.



Supplementary Fig. 20 Transient expression of the CC variants of TdNLR1 in *N. benthamiana* leaves.

a Schematic showing different CC variants of TdNLR1. TdNLR1_CC, CC domain of TdNLR1_h1; TdNLR1_CC^{h4}, CC domain of TdNLR1_h4; TdNLR1_CC^{del}, artificial chimeric CC variant of TdNLR1_h1 with the seven amino acid (GVHDDIQ) deletion. **b** A YFP tag was fused to the C terminus of TdNLR1_CC, TdNLR1_CC^{h4}, and TdNLR1_CC^{del}. Infiltrated leaves (left panel) stained with trypan blue (right panel) to observe cell death at 72 hpi. **c** Western blotting to detect the expression of constructs in *N. benthamiana* leaves. Coomassie brilliant blue staining of RuBisCO is shown below as loading control. Positive control, wheat TaZFP5B protein. Negative control, YFP. All experiments were repeated three times with similar results. Scale bar, 1 cm.



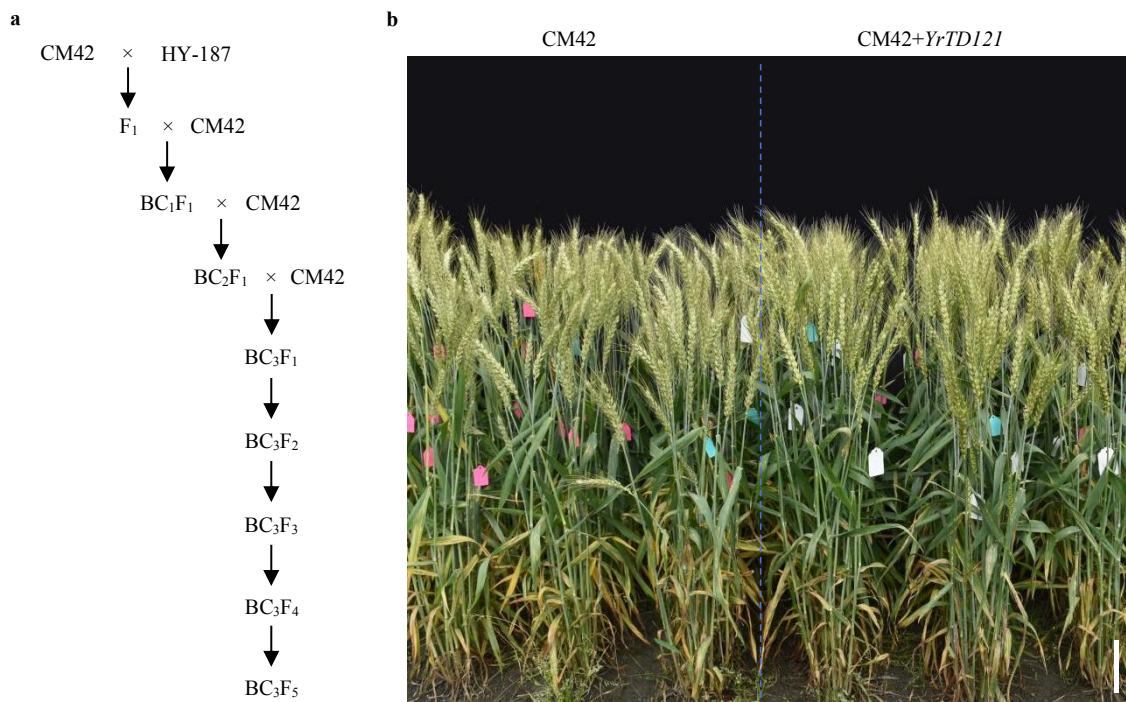
Supplementary Fig. 21 Detection of the interaction between TdNLR1 and TdNLR2 using yeast two-hybrid and luciferase complementation imaging assays.

a Interaction between TdNLR1 and TdNLR2 was investigated using yeast two-hybrid assays. Different bait and prey recombinant constructs were co-transformed into yeast and tested on SD/ - Trp/ - Leu and SD/ - Trp/ - His/ - Leu/ - Ade media with X- α -gal. Positive control, pGADT7-T/pGBKT7-53. Negative control, pGADT7-T/pGBKT7-lam. **b** *N. benthamiana* leaves co-infiltrated with *Agrobacterium tumefaciens* strain GV3101(pSoup-19) containing the indicated constructs. Signals were detected at 48 hpi. At least 10 *N. benthamiana* leaves were analyzed in each of the three experiments. Positive control, wheat NLuc-TaZFP5B/CLuc-TaCML21. Negative control, NLuc/CLuc. Scale bar, 1cm.

AD / BD	SD/-Trp/-Leu				SD/-Trp/-His/-Leu/-Ade			
	10 ⁰	10 ⁻¹	10 ⁻²	10 ⁻³	10 ⁰	10 ⁻¹	10 ⁻²	10 ⁻³
Positive control								
TdNLR1_CC / TdNLR1								
TdNLR1_NB / TdNLR1								
TdNLR1_LRR / TdNLR1								
TdNLR1_CC-NB / TdNLR1								
TdNLR1_NB-LRR / TdNLR1								
TdNLR1 / TdNLR1								
Negative control								

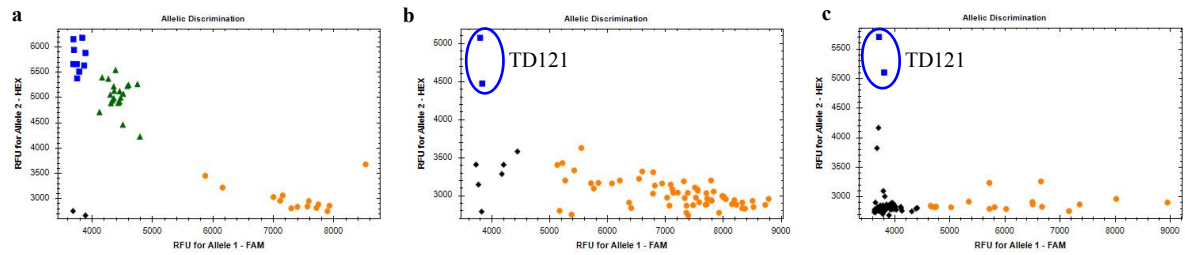
Supplementary Fig. 22 Self-interaction analysis of TdNLR1 and its different domains using a yeast two-hybrid assay.

Self-interactions among TdNLR1 and its different domains, using yeast two-hybrid assays. Different bait and prey constructs were co-transformed into yeast and tested on SD/-Trp/-Leu, and SD/-Trp/-His/-Leu/-Ade media with X- α -gal. CC, coiled-coil. NB, nucleotide-binding site. LRR, leucine-rich repeat. CC-NB, coil-coil nucleotide-binding site. NB-LRR, nucleotide-binding site and leucine-rich repeat. Positive control, pGADT7-T/pGBKT7-53. Negative control, pGADT7-T/pGBKT7-lam. All experiments were repeated three times with similar results.



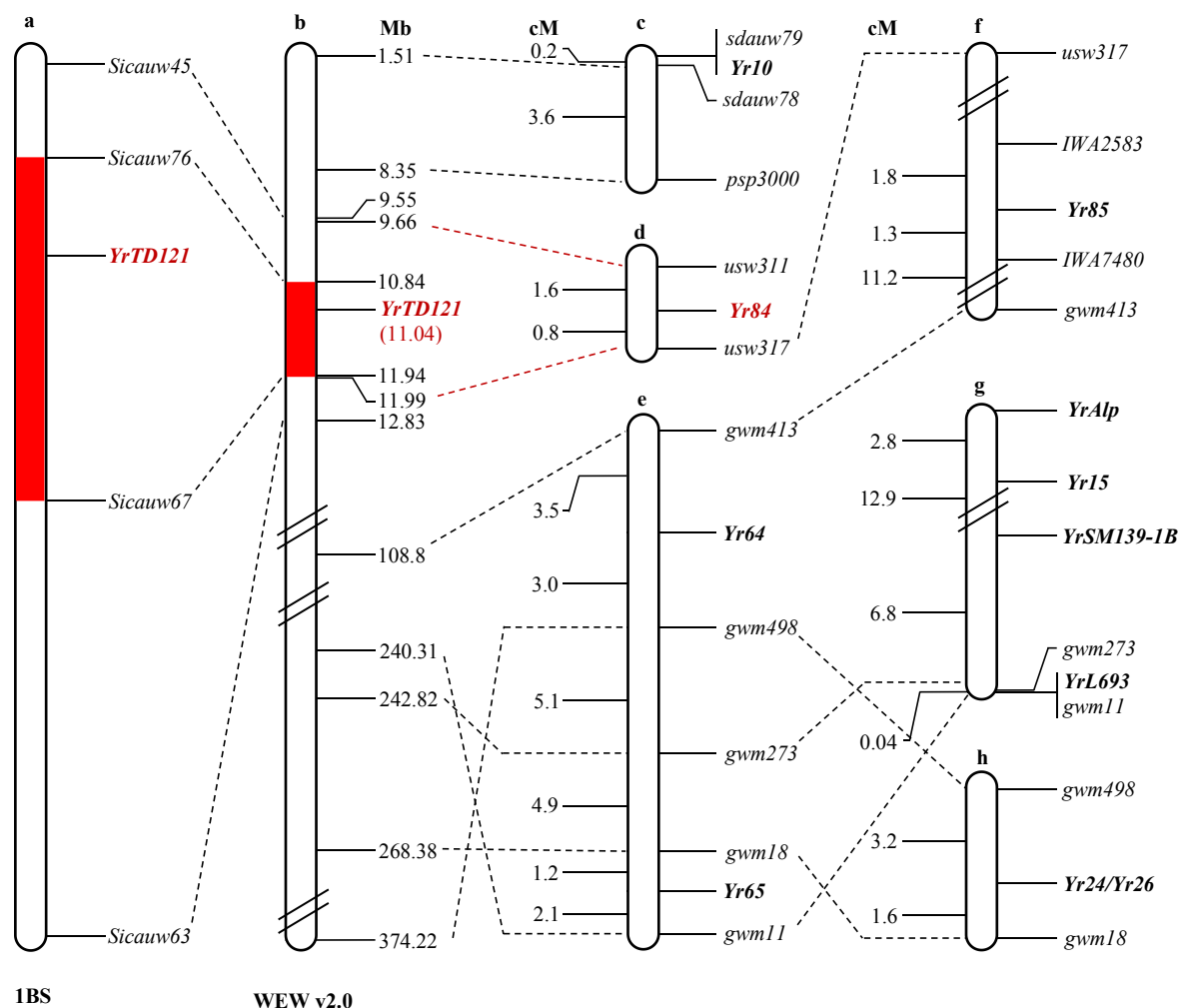
Supplementary Fig. 23 Introgression of *YrTD121* into wheat cultivar Chuanmai 42 (CM42).

a Schematic workflow of the marker-assisted backcross strategy for introgressing *YrTD121* into CM42. **b** CM42 (left) and CM42+*YrTD121* (right) plants showing similar phenotypes under disease-controlled conditions. Scale bar, 10 cm.



Supplementary Fig. 24 Validation of a functional marker for *YrTD121*.

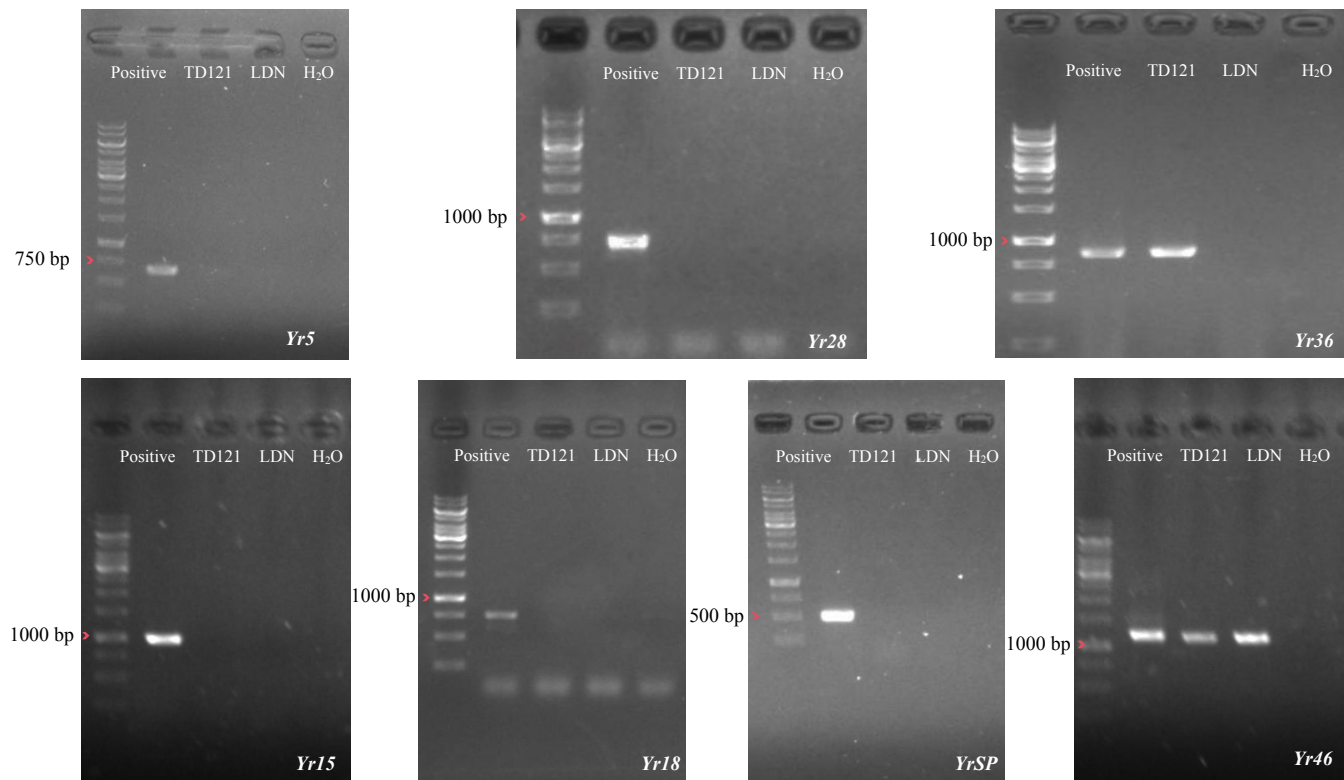
a Genotype clustering plots for the *KASP-FM* marker in an F₂ population from a cross between LDN and HY-187. Genotypes clustered as blue squares at the upper left are homozygous for the functional *TdNLR1*. Genotypes clustered at the lower right as orange circles are homozygous for the non-functional *nlr1* allele. Genotypes clustered in the middle as green triangles are heterozygous. Black diamonds are non-template controls (NTCs). **b, c** Genotyping results for *KASP-FM* in 57 wild emmer wheat (b) and 72 common wheat (c) accessions. Black diamonds, NTCs or genotypes with zero-amplification.



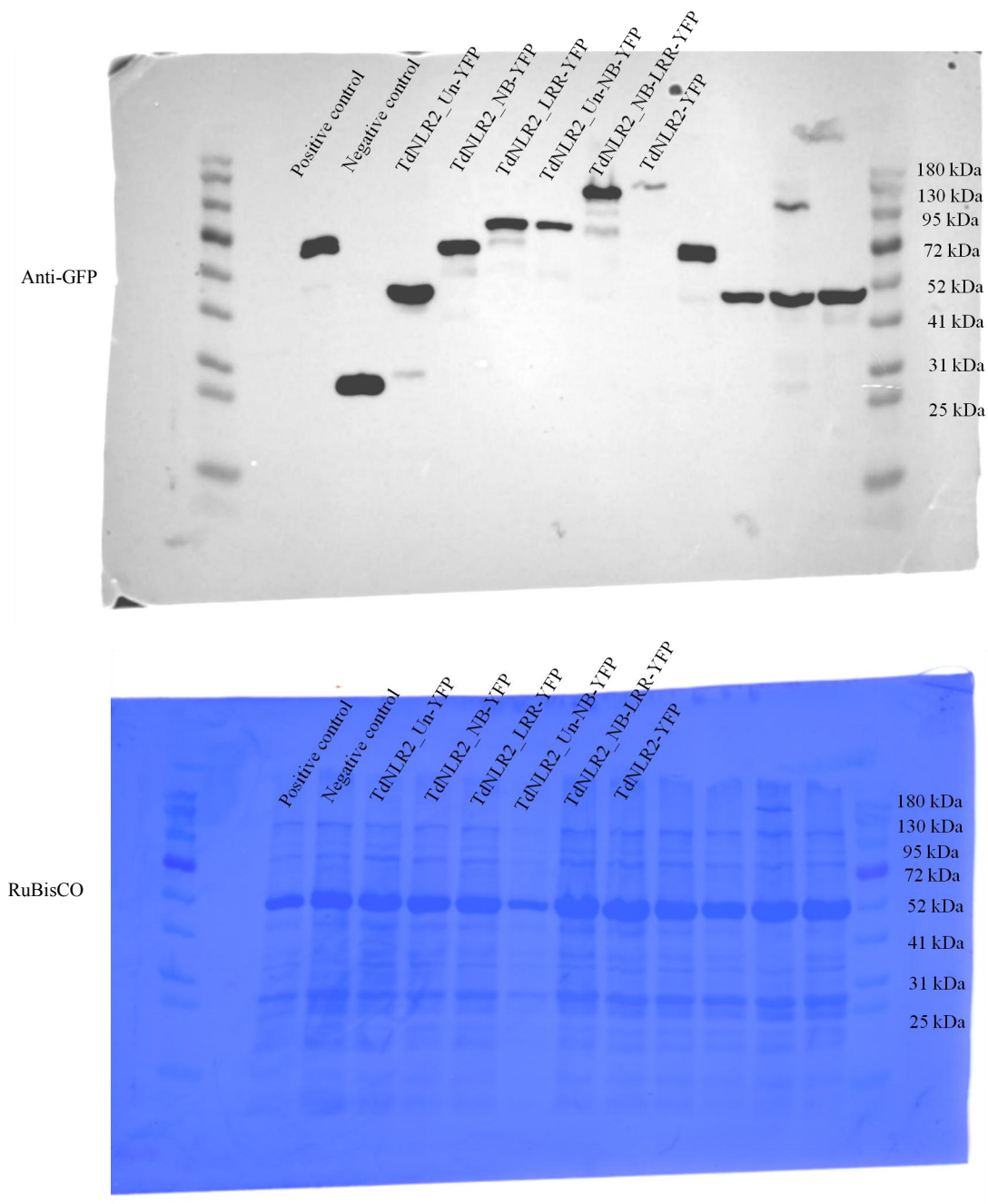
Supplementary Fig. 25 Comparison of the current partial chr 1B linkage map of *YrTD121* (a) with reported locations of *Yr10* (c), *Yr84* (d), *Yr64* and *Yr65* (e), *Yr85* (f), *YrAlp*, *Yr15*, *YrSM139-1B*, and *YrL693* (g), and *Yr24/Yr26* (h). b Physical map of the *YrTD121* region based on the WEW accession Zavitan reference sequence (v2.0). Physical locations in Mb are shown at the right. Red rectangle represents the *YrTD121* candidate region.

Uncropped images of Supplementary Figs. 1, 18, and 20.

Supplementary Fig. 1b



Supplementary Fig. 18c



Supplementary Fig. 20c

