**Supplementary Methods**

**Genome sequencing** Genome sequencing was done on an Illumina GA IIx sequencer. Five micrograms of genomic DNA was sheared to ~400 bp using a Covaris E210 with settings: duty cycle 20%, intensity 4, cycles 200. The DNA was size-selected for the range 300-600 bp on a 2% agarose gel and extracted using a Gel Extraction Kit (Qiagen). The library was constructed using NEB Next DNA Library Prep Master Mix Set for Illumina (New England Biolabs) and Illumina’s Paired-End DNA Sample Prep Oligo Only Kit. The resulting library was selected on a 2% agarose gel for 440-560 bp and extracted using a Gel Extraction Kit (Qiagen). To insure that the library was not over-amplified a test amplification was performed in which aliquots of a PCR reaction were removed every 2 cycles from 4-16. These aliquots were evaluated on a 2% agarose gel and an optimal cycle number was selected for subsequent large scale amplification in which unique barcodes were added to each library. Eight cycles were selected for these libraries. The amplification reaction was cleaned using two rounds of AMPure XP Beads (Agencourt). Each library was run in one lane of a GAIIx to generate paired-end, 101 base reads. Data was processed using RTA1.6.32.0 and GERALD 1.15. For *Tel* 66,654,840 reads were obtained and for *y w* 76,757,736 reads, representing genome coverage of 48X and 55X, respectively.

**Supplementary Results**

**Analysis of large indels** Large indels (>5 bp) identified by manually scanning the *Tel1* genome assembly in the 79 kb region (3R:15,151,000-15,230,000). To confirm the identity of these indels, we designed primers flanking these indels, which were then PCR amplified and sequenced by Sanger sequencing. Figure S4 shows the 145 bp (3R: 15,157,140 – 284) and 85 bp (3R: 15,170,445-524) indels initially identified in *Tel1* but not *y w*. Sequencing of these PCR fragments showed that; the 145 bp ‘deficiency’ is actually many small deletions (6 bp, 5 bp, 23 bp and 2 bp) present very close to each other; the 85 bp deficiency is actually a 82 bp deletion in Tel; the 20 bp insertion (3R: 15,163,360) is actually three closely linked insertions (1 bp, 11 bp and 7 bp). All these indels were also found in one or more of DGRP lines, so they are all natural polymorphisms.

The CLC generated assembly suggested a 38 bp deletion at 3R:15,196,876 in both *Tel* and *y w*. This region is part of exon11 of *Ino80* coding region and thus of potential functional importance. To confirm the presence/absence of this deletion we PCR amplified this region and re-sequenced by Sanger sequencing. This showed complete identity to the reference sequence in both the Tel1 and y w genomes at this region, suggesting the apparent deletion was an assembly artifact, possibly due to the high GC content in this region.