

Figure S1 A probability density graph of distribution of read alignment mapping qualities in a *C. intestinalis* (blue line) and *C. savignyi* (black line) dataset is shown at bottom. Mapping qualities range from 0 to 37, with higher numbers meaning better quality alignment (Li *et al.* 2009). Red dashed line indicates the MapQ value of 15, which was used as the cutoff for mapping analysis.

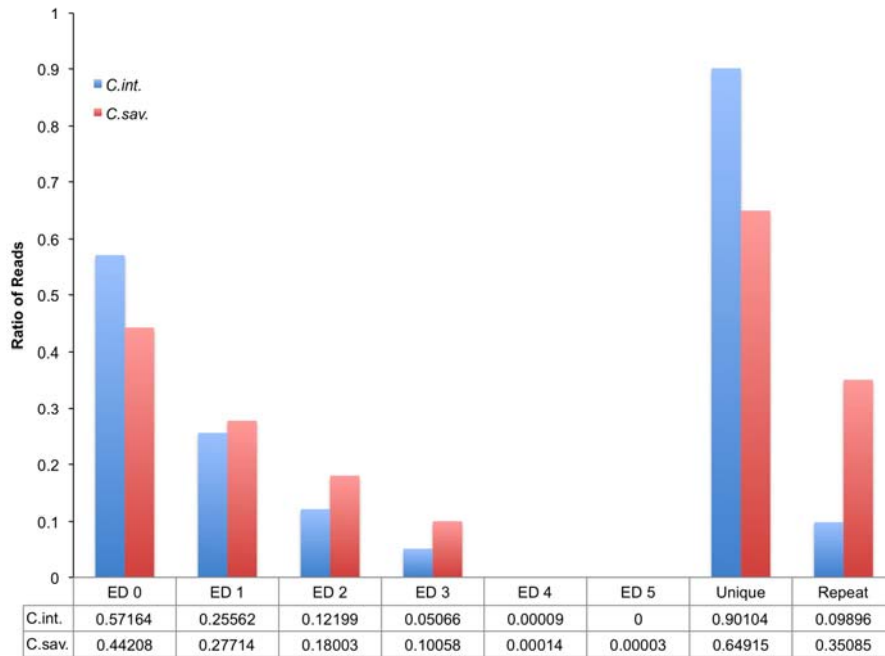


Figure S2 Comparison of Edit Distance (ED), and Unique versus Repeat character, of sequence reads between *C. intestinalis* (C.int.) and *C. savignyi* (C.sav.). The analysis was performed on a sample of 5 million randomly-picked reads. The values given for the two species represent the fraction of aligned reads with either the indicated ED assignment (0-5), or the Unique versus Repeat characteristic. Overall greater ED was observed in the *C. savignyi* sequence alignments in comparison to those from *C. intestinalis*. In addition, a greater fraction of *C. savignyi* reads were characterized as Repeats post-alignment. Both these factors, as well as others, contribute to the differential mapping quality of the two species (Li *et al.* 2009).

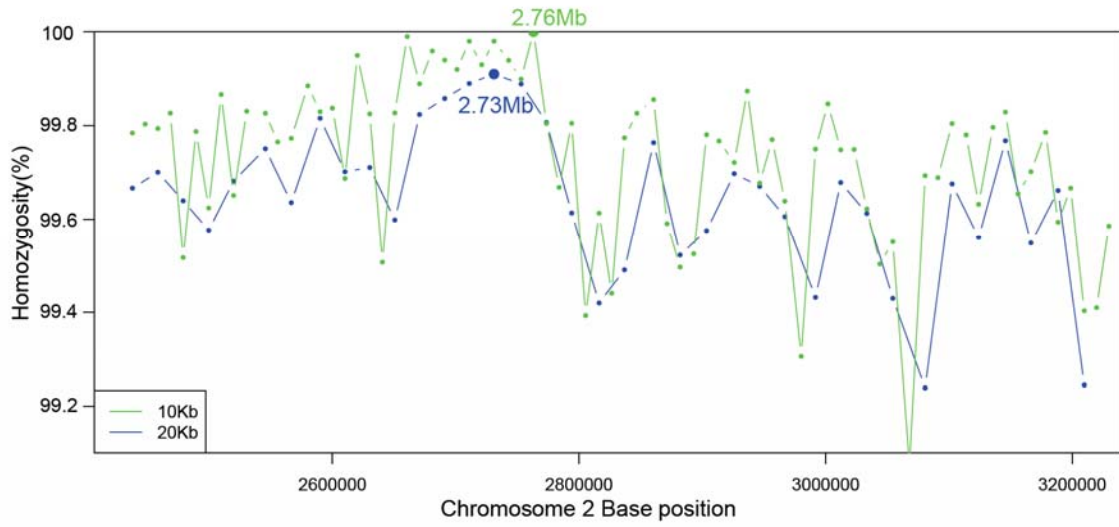


Figure S3 Window size differences for *C. intestinalis chongmague* genome-wide mapping. A zoomed in area of chromosome 2, near the peak calls for both a 10 Kb (green) and 20 Kb (blue) window analysis of homozygosity. Peaks of each analysis are shown as larger solid filled circles with positional values indicated next to each point.

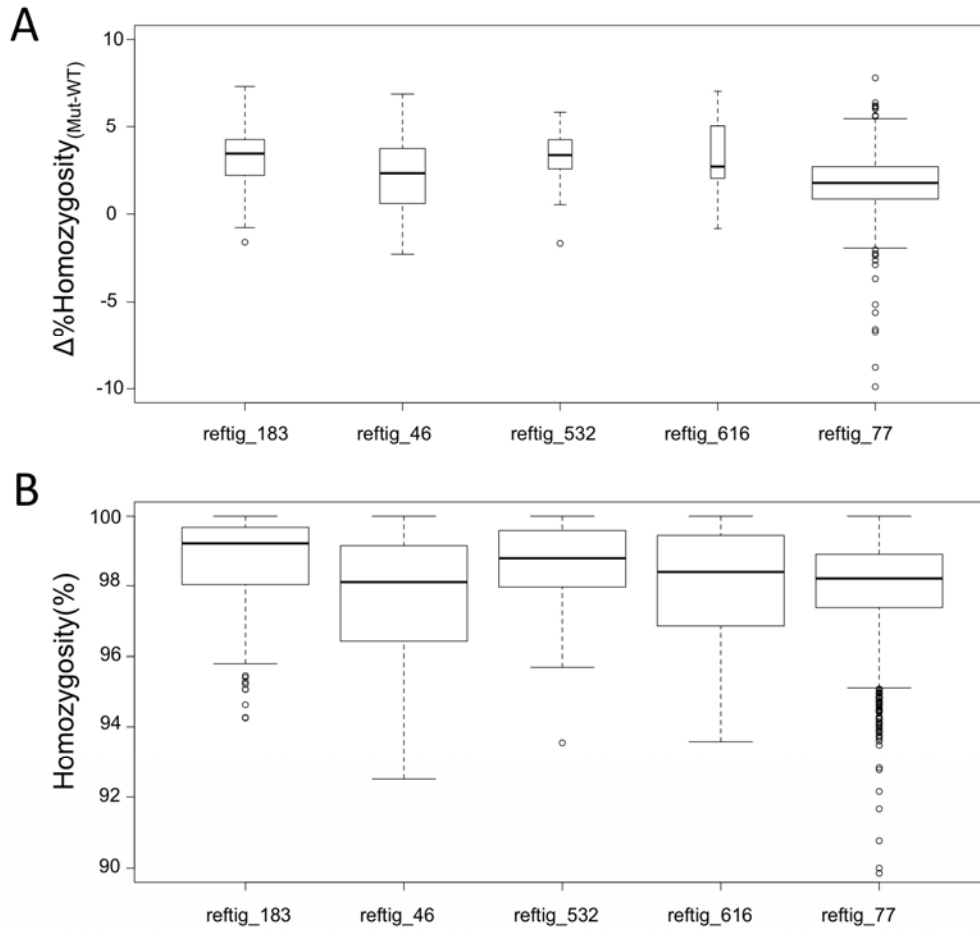


Figure S4 Homozygosity mapping analysis for *bugeye*. **A)** Box-whisker plot of Δ homozygosity values for each candidate high value reftig from Figure 2B. Δ homozygosity values were calculated for 1 Kb windows across each reftig. Width of boxplots depicts amount of data points for each reftig. Solid lines indicate median values, and whiskers indicate extreme values of reftig. Reftig 183 had the highest median Δ homozygosity value (3.46%). **B)** Box-whisker plot of homozygosity values for the candidate reftigs (mutant sample only). Plots were done as above for panel A.

Table S1 Difference in coverage between coding and non-coding areas. Coding sequence regions were collected from Ensemble database (release 74) of each species' genome. A 5 Mb genomic region on Chromosome 1 of *C. intestinalis* and ref1g 1 of *C. savignyi* were used for sampling differences in coverage in each sample.

| Species | CDS | | non-CDS | | Average |
|------------------------|----------|---------------------|----------|---------------------|----------|
| | Coverage | Fraction of Average | Coverage | Fraction of Average | Coverage |
| <i>C. savignyi</i> | 34.15 | 1.42 | 22.34 | 0.93 | 24.06 |
| <i>C. intestinalis</i> | 52.23 | 1.06 | 48.68 | 0.99 | 49.29 |

Table S2 Primers for qRT-PCR

| Gene | Forward Primer | Reverse Primer |
|-----------------------|----------------------|-----------------------|
| ci-Actin | CCAGCAGATTCCATACCAAG | CGTTTTCCCATCCATCGTAG |
| ci-alpha Laminin PP#1 | CGGTGACGAAAATGAGGAAC | AGACACCACCACCCTCGTAG |
| ci-alpha Laminin PP#2 | TCAAGTTGGTTCCGCATGTA | GTTCCACATTCCACCAATCC |
| cs-CAV3 PP#1 | GCGCATTTTGGTCATGCTAC | GGCTTGCCCACTTGATAATG |
| cs CAV3 PP#2 | ACCATTTTGTTCGCCTTTT | ATTGAAGATATTGGGGTCCA |
| cs RPS27A | CCACCTGATCAGCAGAGGTT | TTATTCGCCCTCTGGTTTGA |
| cs Rab21 | TTCGTGGTGGGAAATAAAGC | GTTTTCCGTTTTTCACGCAAT |
| cs FLRT2 | GTACACTGCTGCGAGGAACA | CCGTCTGATTGGTGGAAAGT |
| cs M.R. | CCGATGCTACGCCTATGACT | AGCCTCTACGTCGCCATCTA |