Figure S1

Phylogenetic relationships among 25 *S. noctiflora* samples and closely related species based on 13 mitochondrial markers. Supporting bootstrap values greater than 80 are shown for each node.

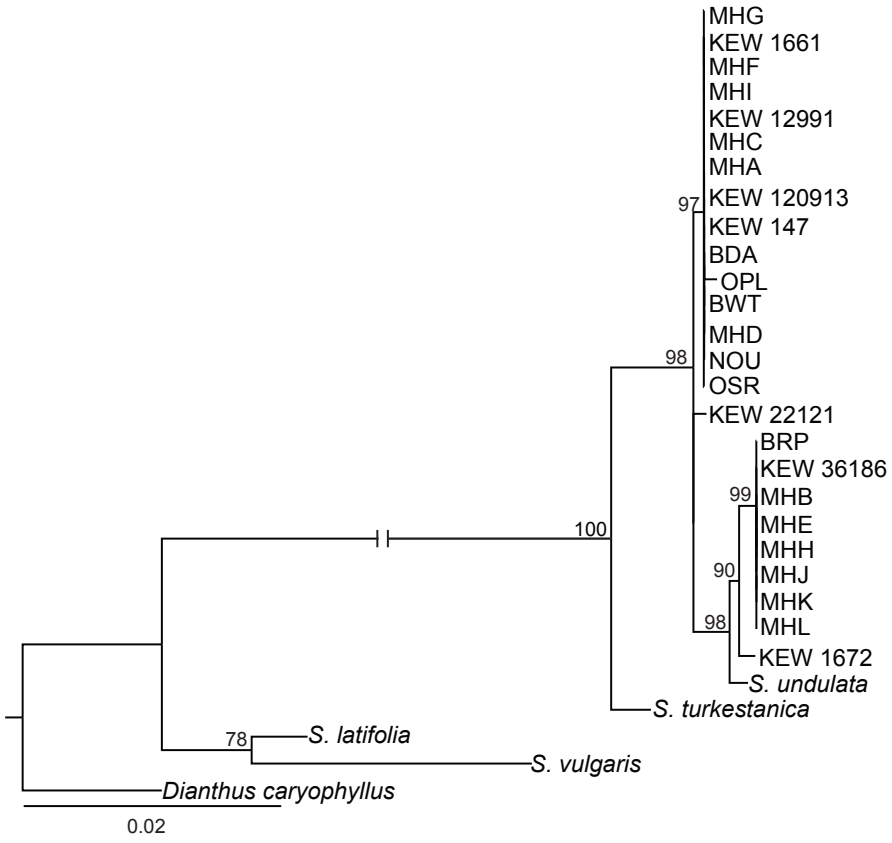


Figure S2

Reconstruction of ancestral States for the presence or absence of each of the 22 sampled chromosomes across 25 *S. noctiflora* samples and the close relative *S. undulata* under Dollo parsimony. Filled purple boxes indicate presence of the chromosome. Open purple boxes represent an ancestral absence of the chromosome. Open orange boxes represent a derived absence (i.e., loss) of the chromosome). See Figure 4 for details on chromosome sampling and constraint trees.

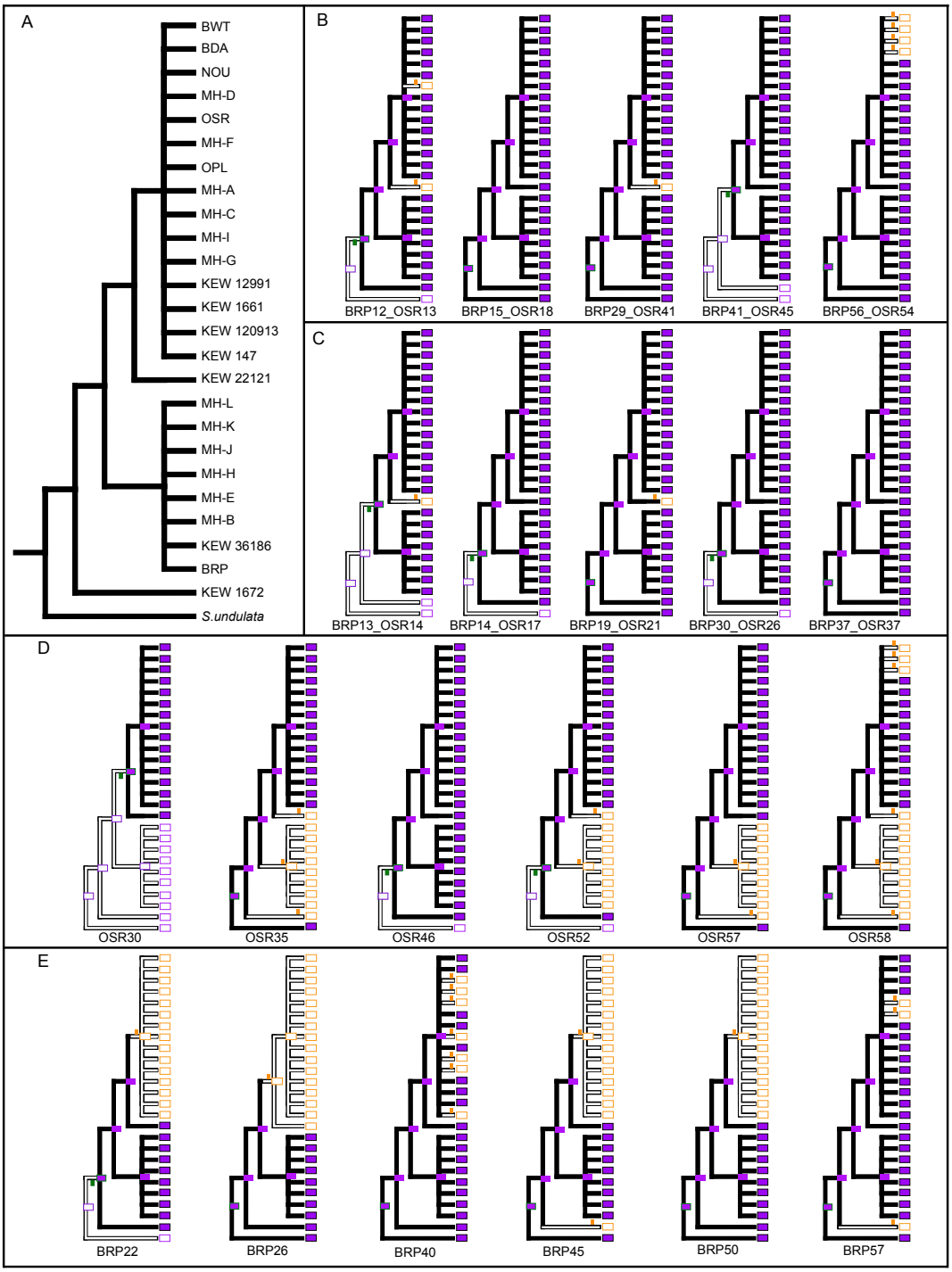
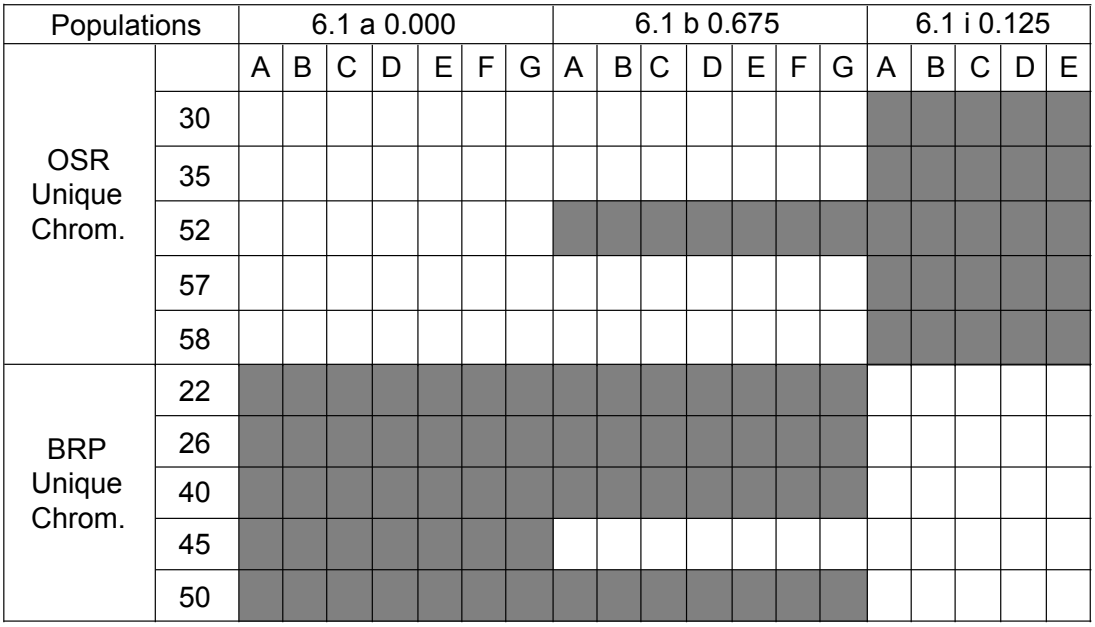


Figure S3.
Presence/absence survey of 10 mitochondrial chromosomes (five OSR-specific and five BRP-specific) for *S. noctiflora* individuals from three sites in a metapopulation from southwestern Virginia.



Figures S4-S12.

Read depth across the chromosomes of the *S. noctiflora* OSR (Figures S4, S6, S8, S10, and S12) or BRP (Figures S5, S7, S9 and S11) reference mitochondrial genome based on Illumina sequencing of total-cellular DNA. Coverage estimates are based on a sliding window with a window size of 1000 bp and a step size of 500 bp. Reference chromosomes are ordered in decreasing size on the same x-axis scale, so coverage maps end before the end of the x-axis. Sequencing datasets are derived *S. noctiflora* KEW 22121 (Figure S4), *S. noctiflora* OPL (Figures S5 and S6), *S. noctiflora* KEW 1672 (Figures S7 and S8), *S. undulata* (Figures S9 and S10), and *S. turkestanica* (Figures S11 and S12). The extensive heterogeneity in coverage observed for *S. turkestanica* dataset (Figures S11 and S12) likely reflects the bias introduced by whole genome amplification.

Figure S4. Kew 22121 (OSR Reference)

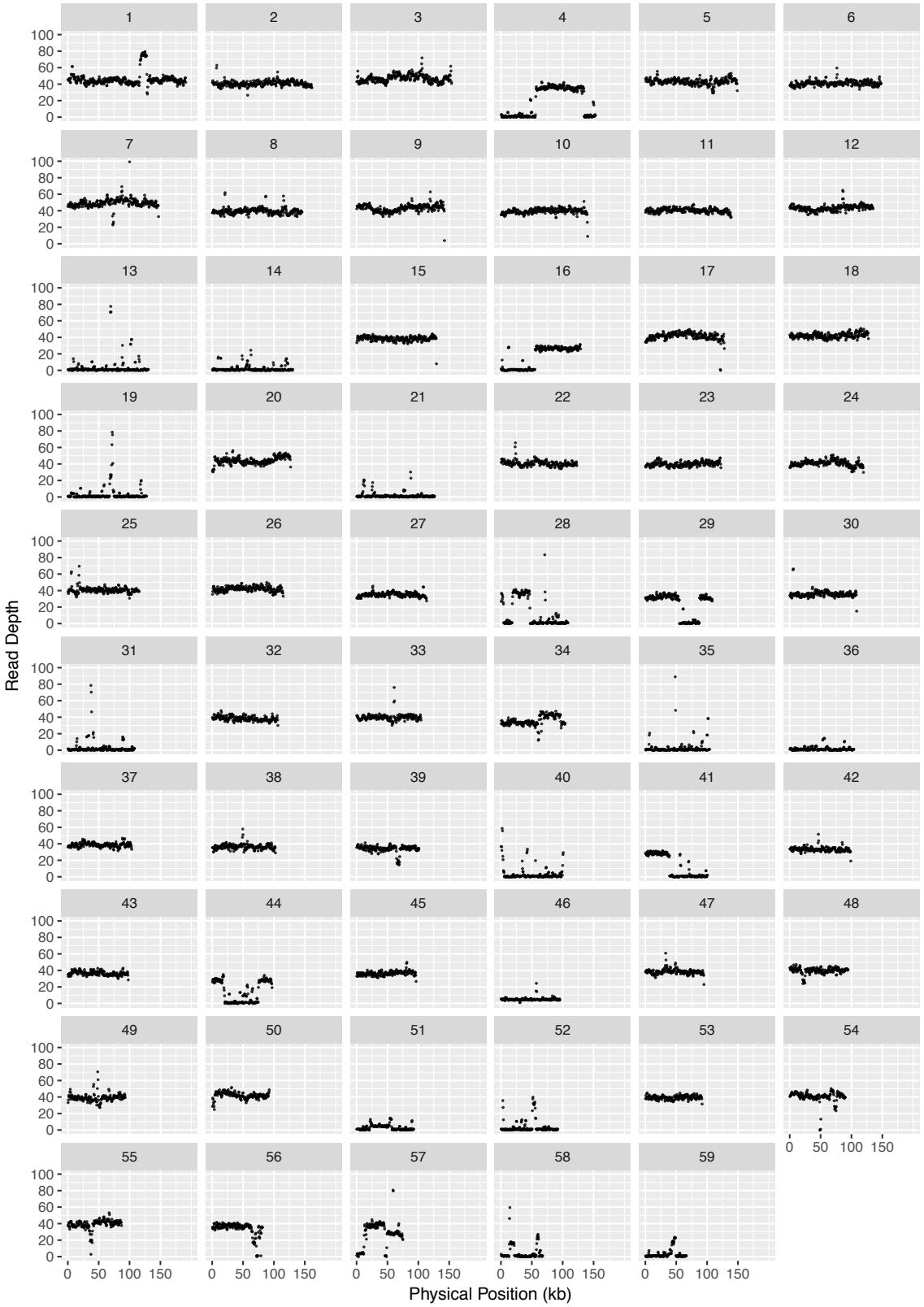


Figure S5. OPL (BRP Reference)

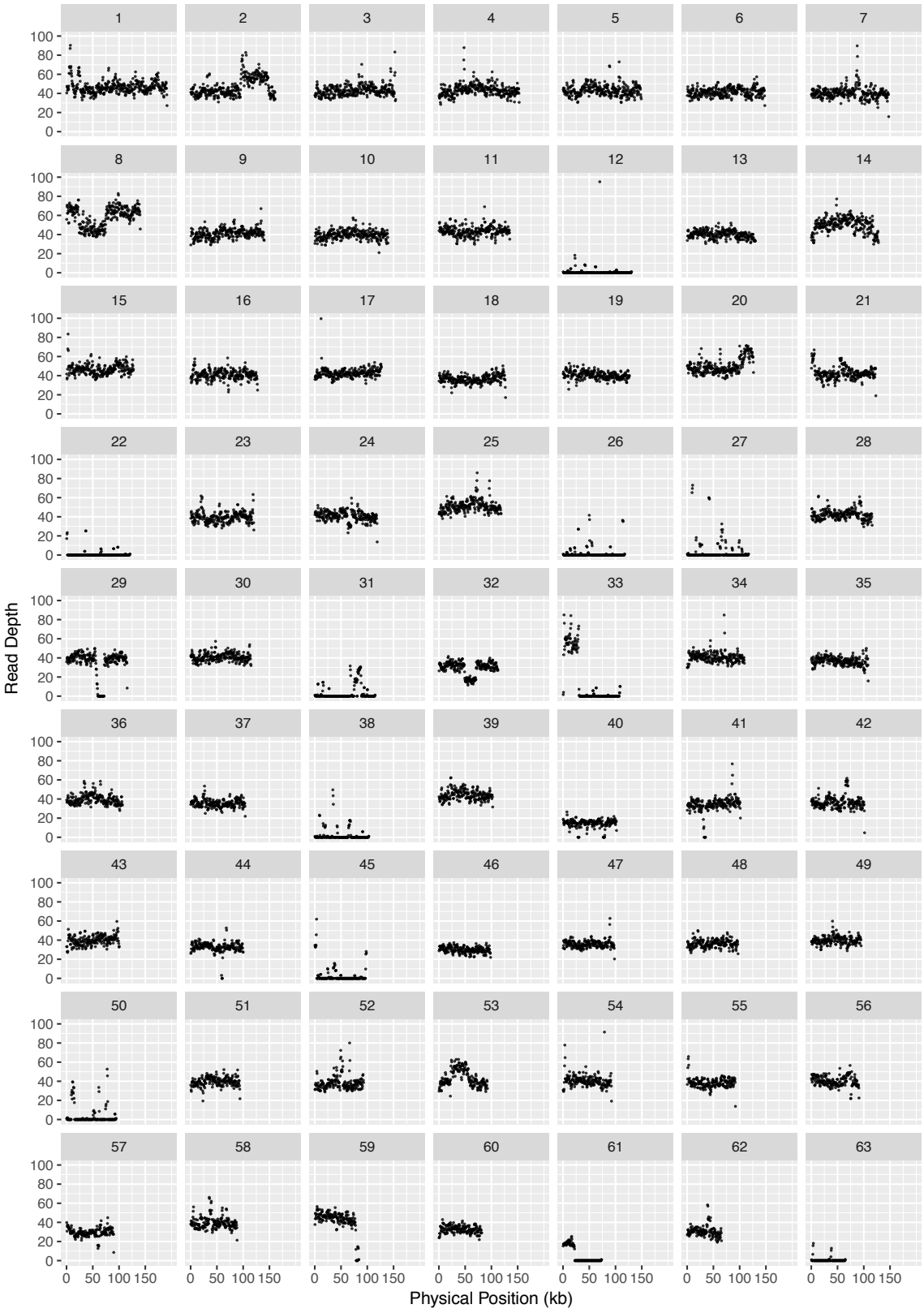


Figure S6. OPL (OSR Reference)

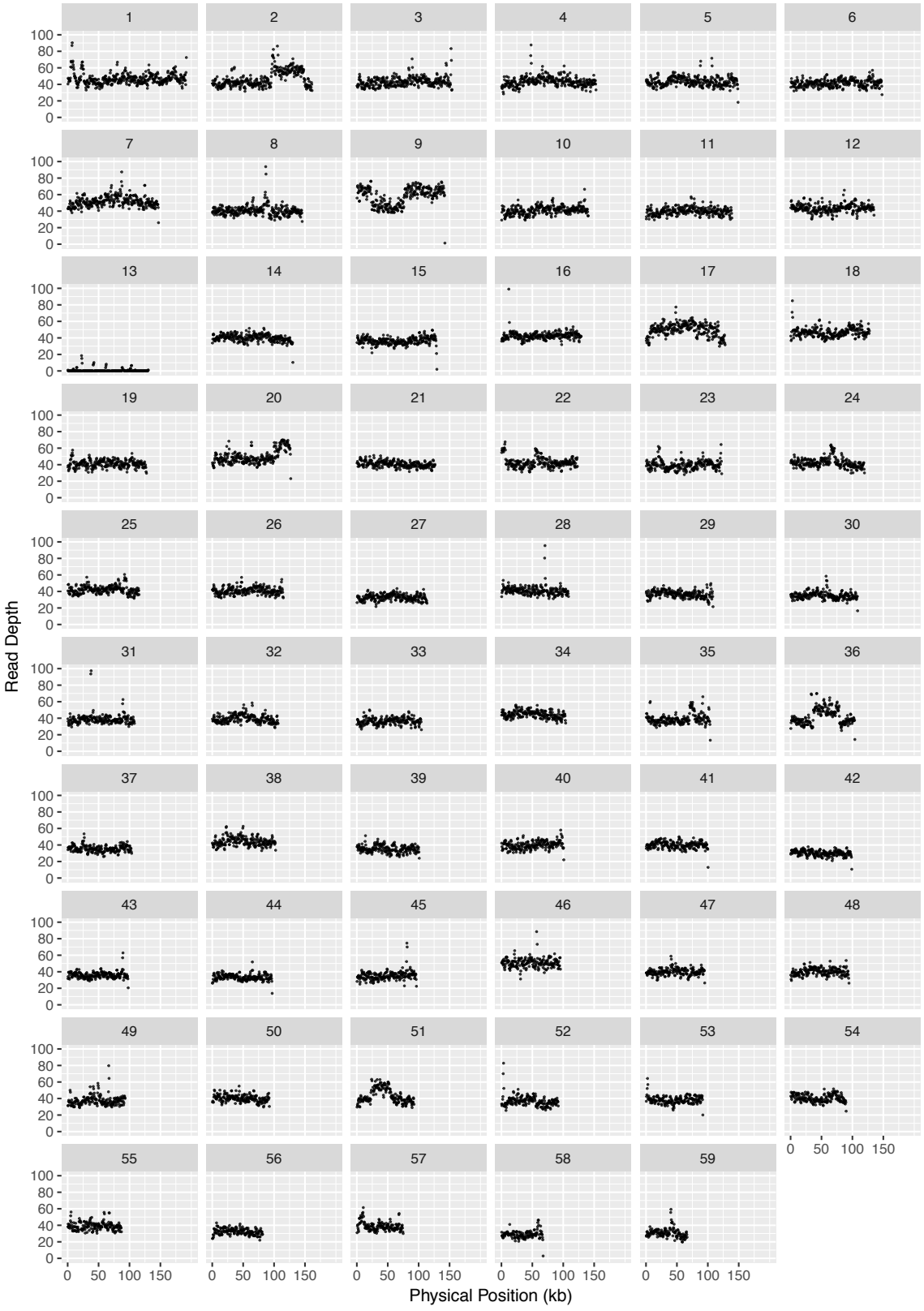


Figure S7. KEW 1672 (BRP Reference)

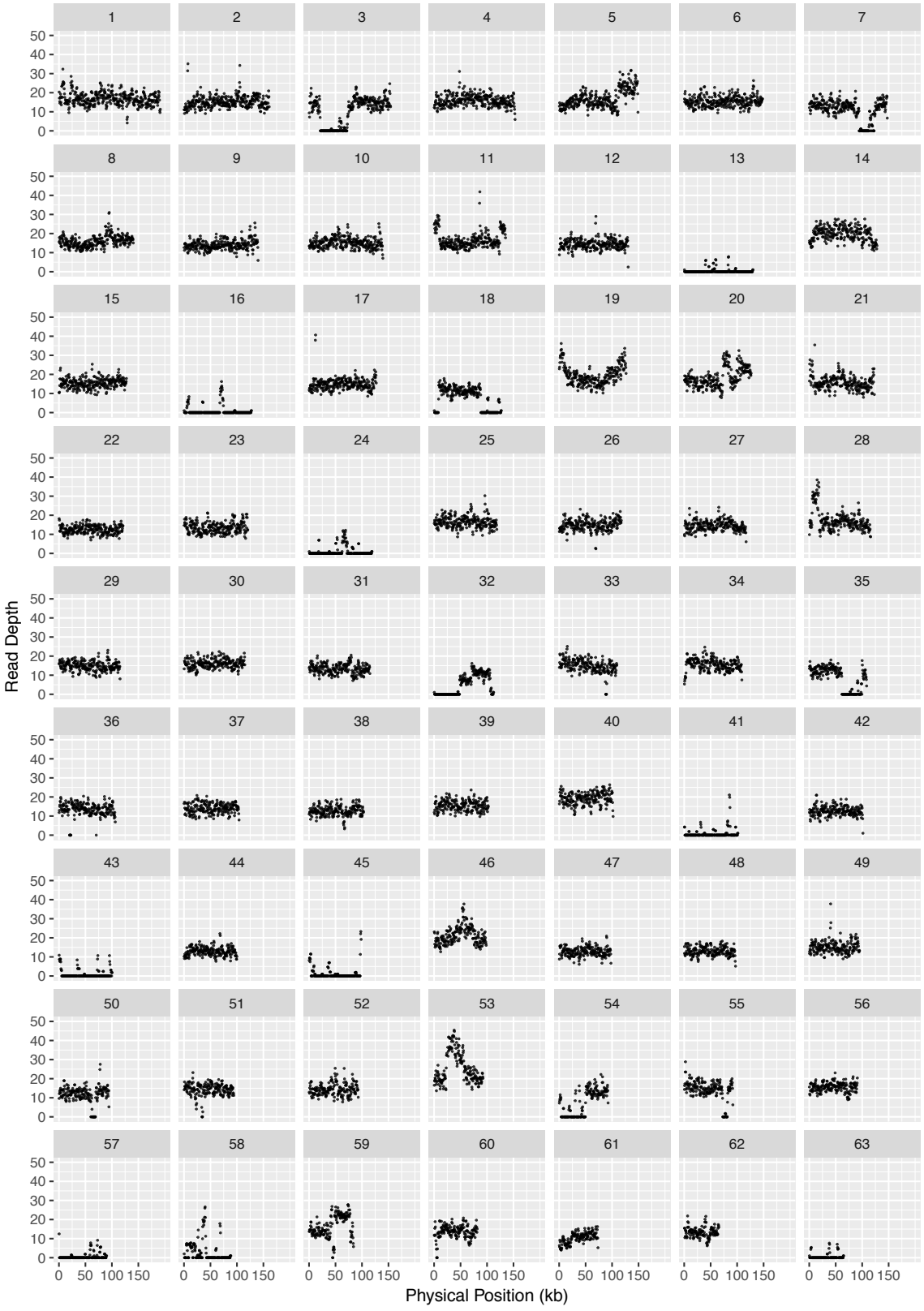


Figure S8. KEW 1672 (OSR Reference)

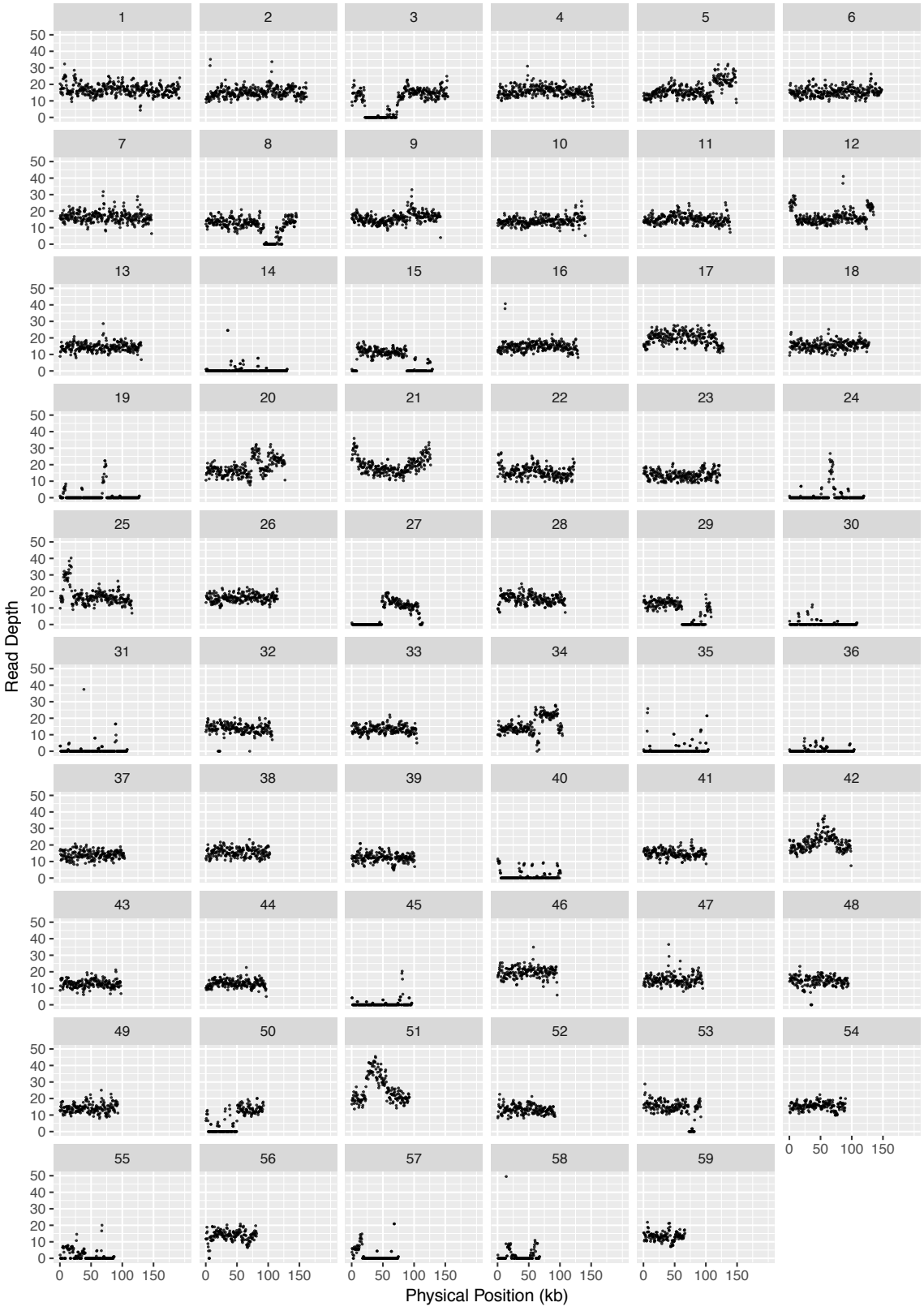


Figure S9. *S. undulata* (BRP Reference)

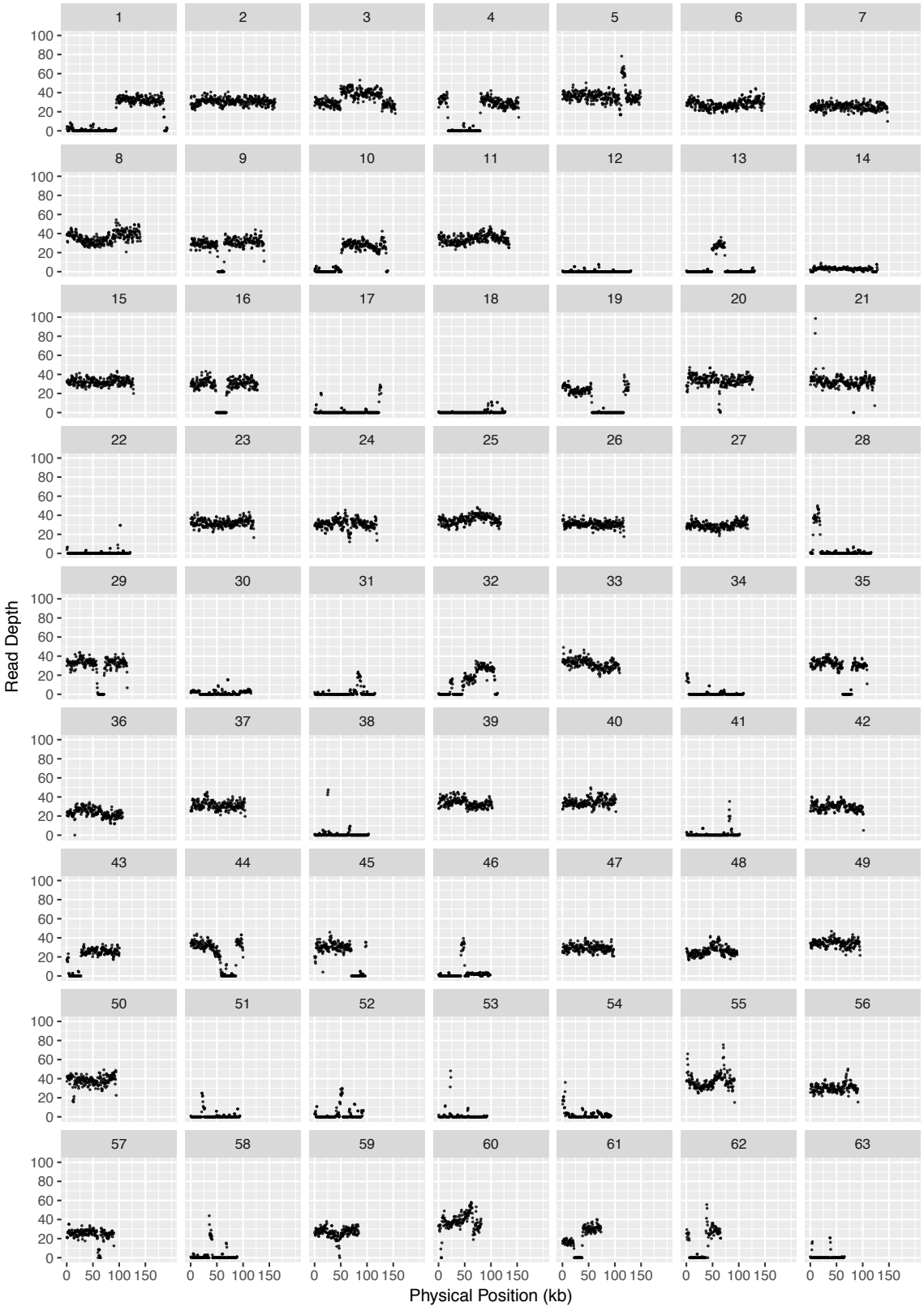


Figure S10. *S. undulata* (OSR Reference)

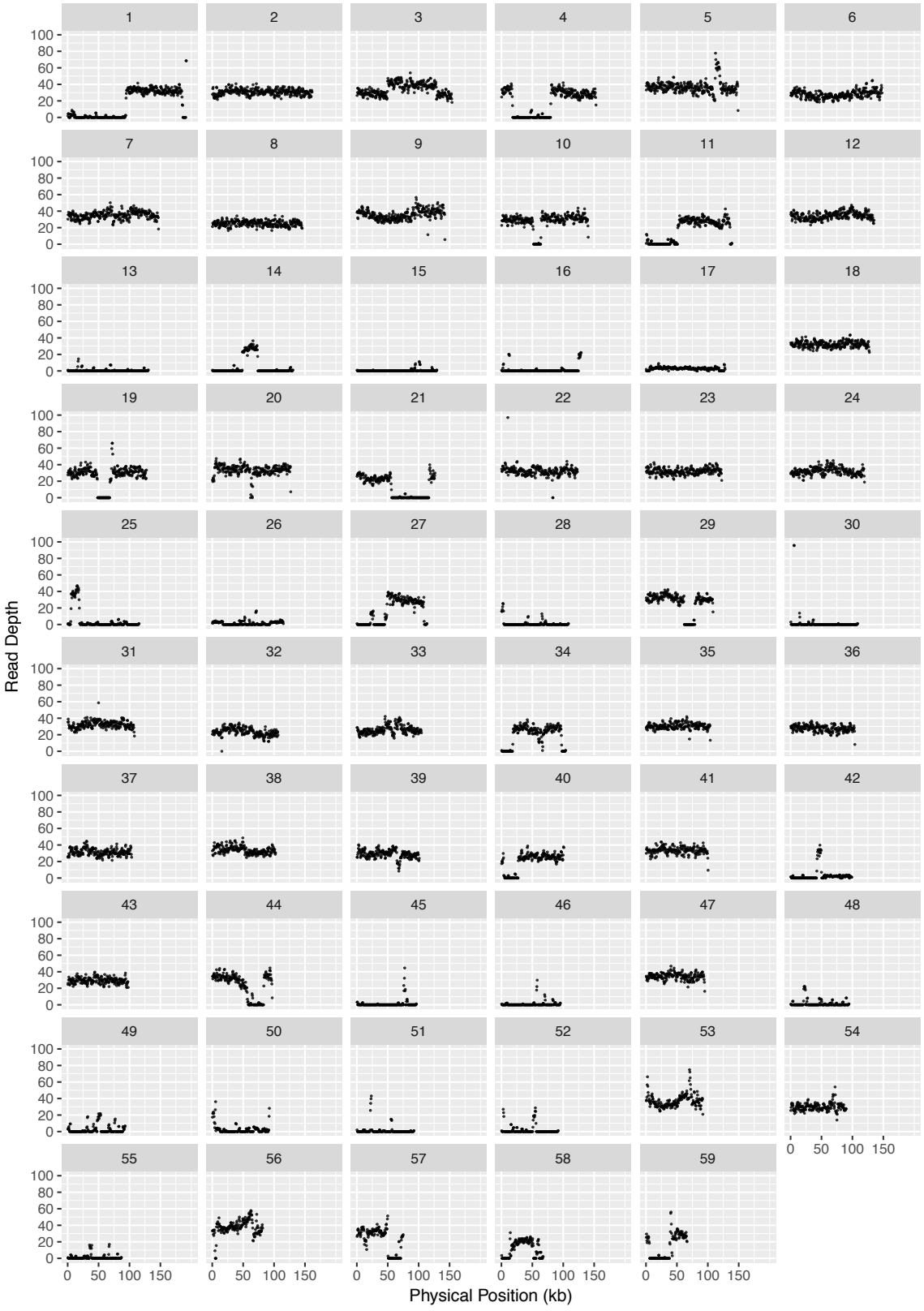


Figure S11. *S. turkestanica* (BRP Reference)

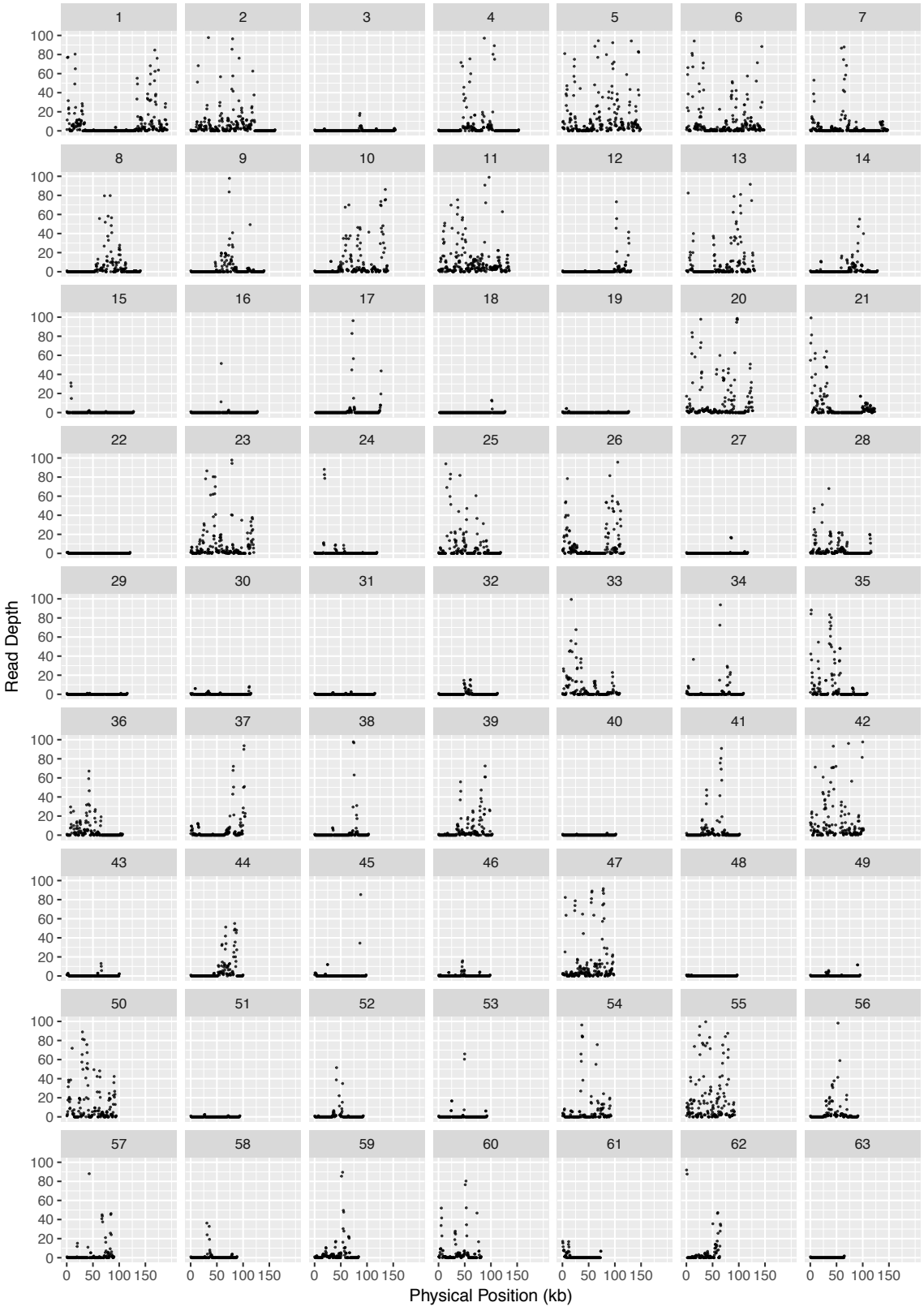


Figure S12. *S. turkestanica* (OSR Reference)

File S1
Concatenated alignment of 13 mitochondrial sequence markers used for phylogenetic analysis (Nexus format).

File S2
Alignment of plastid genomes used for phylogenetic analysis (Fasta format).

File S3
Concatenated alignment of mitochondrial genomes used for phylogenetic analysis (Fasta format).