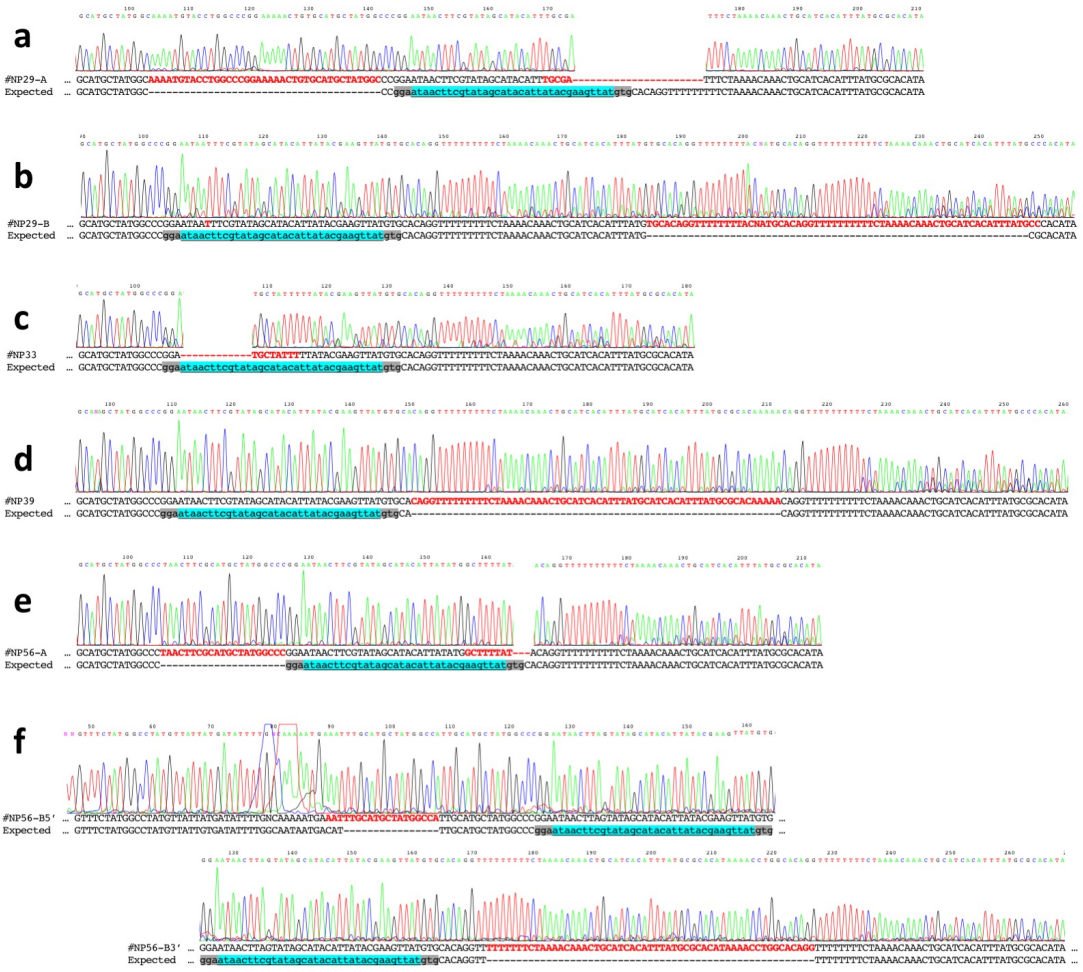

Supplementary Figure 2. Screening for integrations of the second loxP site into *tbx20*. **a, b.** Sequencing of individual embryos from outcross of candidate founder #NP29. One out of sixteen embryos contained an incomplete loxP site with a small deletion (**a**), and two out of sixteen embryos were heterozygous for an allele with a complete loxP site containing a single nucleotide substitution, with an insertion of additional 64 nucleotides (**b**). **c.** Sequencing of individual embryos from cross of founder #NP33. Nine out of sixteen individual embryos were heterozygous for this integration of an incomplete loxP site. **d.** Sequencing of individual embryo from cross of founder #NP39. Five out of sixteen individual embryos were heterozygous for integration of a complete loxP site with an insertion of additional 62 nucleotides. Floxed allele *tbx20*^{tpl145} (**Figure 2**) was established from this founder. **e, f.** Sequencing of individual embryos from outcross of founder #NP58. Four out of sixteen individual embryos were heterozygous for an integration of an incomplete loxP site with insertion of additional 21 nucleotides (**e**), and one out of sixteen was heterozygous for integration of complete loxP site containing a single nucleotide substitution, with insertions of 16 and 55 nucleotides on each side of the loxP site (**f**). Note that all large insertions appear to be partial target site duplications.