



Figure S3 Oligo design to sequence FLAG knock-in heterozygotes. Design of oligos to sequence into 5' end (A) and 3' end (B) of *nhr-23* 2x and 3x FLAG knock-ins. The non-coding strand of *nhr-23 (+)* is shown paired with the sequencing oligo. The stop codon (blue text), PAM #1 (red text), and a portion of the 2xFLAG knock-in sequence (orange text) are indicated. The oligo is designed to bind the genomic sequence at the insertion site with the last two bases binding bases in the insert. In cases where sequence is too poor to confirm correct insertion of an epitope, an additional round of PCR can be performed using one of the insert-specific oligos and an external primer that binds in the genomic sequence. Purification of this product followed by sequencing using the primer that binds in the genomic sequence provides the entire epitope sequence. In events where 2 bp of knock-in sequence is not sufficient to confer specificity, increasing the knock-in specific homology will correct the problem at the expense of sequencing coverage of the knock-in.