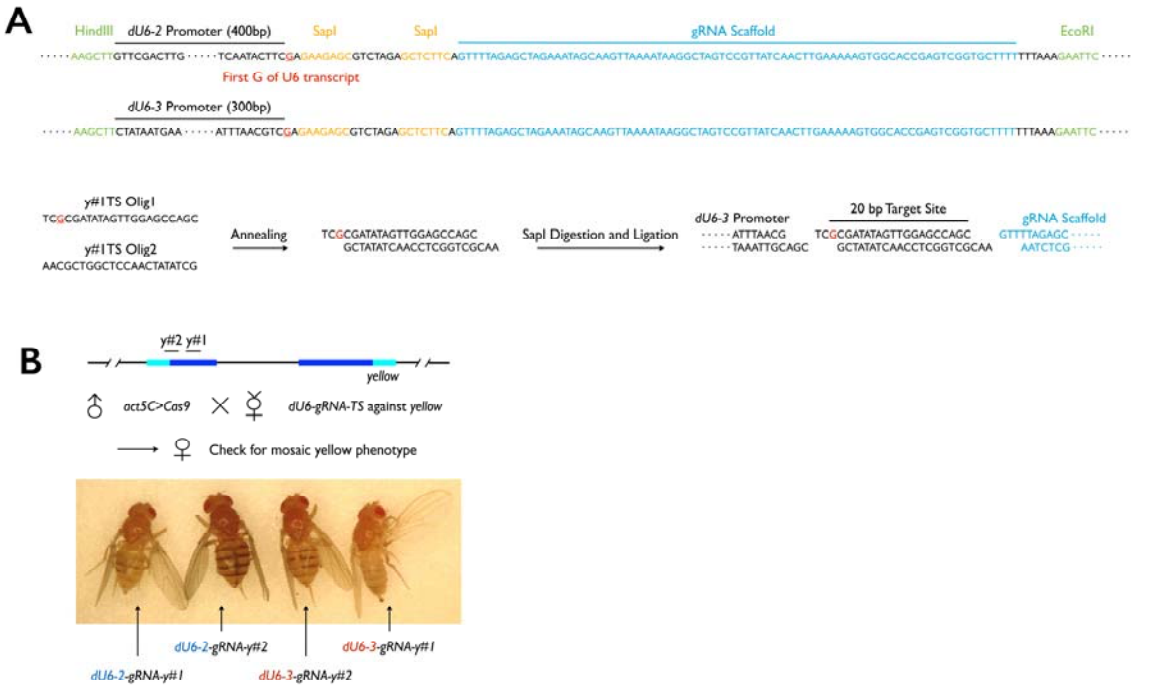

Figure S2 Generation and validation of dU6-gRNA transgenes

Sequence arrangement of the dU6-gRNA scaffolds are shown on top. dU6-gRNA backbone was cloned into pJFRC28 using *Hin*dIII and *Eco*RI sites. Two *Sap*I sites were put in-between *dU6* promoter and the gRNA scaffold for easy target site cloning. gRNA scaffold is the same as the published one (Cong *et al*. 2013; Mali *et al*. 2013). After annealing two corresponding target site primers, the target site can be directly ligated with the SapI-digested empty dU6-gRNA with its TCG and AAG 5' overhangs to constitute a functional dU6-gRNA. (B) Females carrying various *U6-gRNAs* against *yellow* were crossed to males with *act5C>Cas9*. Their female progeny showed allele-dependent yellow body color mosaicism, with *dU6-3-gRNA-y#1* causing a yellow phenotype throughout almost the whole body whereas *dU6-2-gRNA-y#2* affected few, if any, cells.