Table S3. Sequences of the oligonucleotides for off-target effect analysis.

### White-A		sgRNA name	embryos	total GO	fertile G0	survival rate*	fertile rate**
white-B 61 16 13 26.2% 81.3% white-C 60 15 8 25.0% 53.3% white-D 50 10 5 20.0% 50.0% 50.0% white-E 51 10 8 19.6% 80.0% white-E 51 10 8 19.6% 80.0% white-E 65 16 7 24.6% 68.8% 43.8% white-H 65 14 9 21.5% 64.3% white-H 50 6 5 16 11 24.6% 68.8% 33.6% white-H 50 10 5 20.0% 50.0% 50.0% white-H 50 10 5 20.0% 50.0% 50.0% white-H 65 14 9 21.5% 64.3% white-H 65 15 5 23.1% 33.3% 50.0% white-H 66 19 6 28.8% 31.6% white-H 65 15 5 23.1% 33.3% 50.0% white-D 65 15 5 5 23.1% 33.3% white-M 66 18 12 27.3% 66.7% white-D 65 18 5 5 27.7% 27.8% white-D 65 18 5 27.7% 27.8% white-D 65 18 5 27.7% 27.8% white-D 65 16 10 24.6% 62.5% white-D 60 15 9 9 50.0% 60.0% white-E 65 16 10 24.6% 62.5% 65 16 10 24.6% 62.5% white-D 65 16 10 24.6% 62.5% 65.0% white-E 65 16 10 24.6% 62.5% 65.0% white-E white-E 65 16 17 10 26.2% 58.8% white-D white-B 65 16 17 10 26.2% 58.8% white-D white-B 65 16 10 7 17.9% 70.0% white-E white-E 65 16 19 24.2% 56.3% 68.8% white-D white-B 66 16 9 24.2% 56.3% 68.8% white-D white-B 65 16 19 24.6% 25.5% 68.8% white-D white-B 65 16 9 24.2% 56.3% 68.8% white-E white-B 65 16 9 24.6% 50.0% 60.0% white-E white-B 65 16 9 24.2% 56.3% 68.8% white-E white-B 65 16 9 24.6% 50.0% 6	Cas9						
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White-K + white-S 65 17 9 26.2% 52.9% white-F + white-L 65 19 13 29.2% 68.4% white-K + white-P 55 10 7 18.2% 70.0% white-F + white-M 55 10 5 18.2% 50.0% white-S + white-N 65 18 17 27.7% 94.4% white-E + white-F 50 7 5 14.0% 71.4% white-D + white-R 68 18 11 26.5% 61.1% white-D + white-H 50 9 4 18.0% 44.4% white-Q + white-B 45 7 6 15.6% 85.7% white-D + white-C 55 13 9 23.6% 69.2%			60	13	12	21.7%	92.3%
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White-K + white-P 55 10 7 18.2% 70.0% White-F + white-M 55 10 5 18.2% 50.0% White-S + white-N 65 18 17 27.7% 94.4% White-E + white-F 50 7 5 14.0% 71.4% White-D + white-R 68 18 11 26.5% 61.1% White-D + white-H 50 9 4 18.0% 44.4% White-Q + white-B 45 7 6 15.6% 85.7% White-E + white-R 45 7 6 15.6% 85.7% White-D + white-C 55 13 9 23.6% 69.2%				19			68.4%
White-F + white-M 55 10 5 18.2% 50.0% White-S + white-N 65 18 17 27.7% 94.4% White-E + white-F 50 7 5 14.0% 71.4% White-D + white-R 68 18 11 26.5% 61.1% White-D + white-H 50 9 4 18.0% 44.4% White-Q + white-B 45 7 6 15.6% 85.7% White-E + white-R 45 7 6 15.6% 85.7% White-D + white-C 55 13 9 23.6% 69.2%				10			70.0%
Cas9 ^{H840A} White-S + white-N 65 18 17 27.7% 94.4% white-E + white-F 50 7 5 14.0% 71.4% white-D + white-R 68 18 11 26.5% 61.1% white-D + white-H 50 9 4 18.0% 44.4% white-Q + white-B 45 7 6 15.6% 85.7% white-E + white-R 45 7 6 15.6% 85.7% white-D + white-C 55 13 9 23.6% 69.2%					5		
Cas9 ^{H840A} White-E + white-F white-D + white-R white-D + white-H white-D + white-B white-Q + white-B white-E + white-R white-E + white-R white-D + white-R white-D + white-B white-D + white-C 55 13 9 23.6% 71.4% 7				18			
Cas9 ^{H840A} white-D + white-R white-D + white-H bo 9 4 18.0% 44.4% white-Q + white-B white-E + white-R white-D + white-C 55 13 9 23.6% 61.1%			50	7			71.4%
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white-E + white-R 45 7 6 15.6% 85.7% white-D + white-C 55 13 9 23.6% 69.2%							
white-D + white-C 55 13 9 23.6% 69.2%							
		white-E + white-H	50	10	10	20.0%	100.0%
white-E + white-I 65 16 11 24.6% 68.8%							

^{*}The survival rate is calculated as the percentage of total G0 adults divided by embryos injected.

**The fertile rate is calculated as the percentage of fertile G0 flies divided by total G0 adults.

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