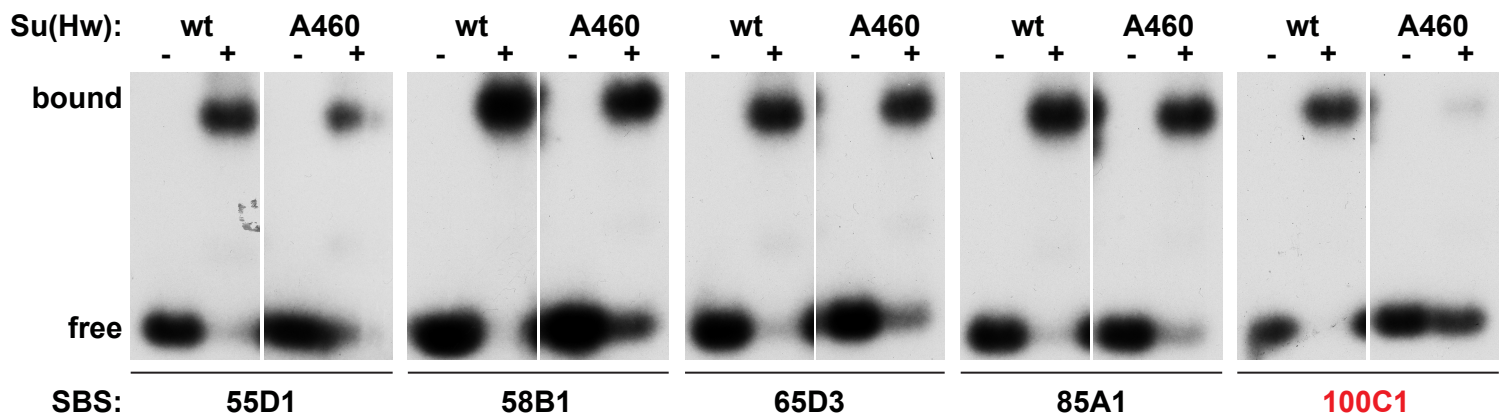
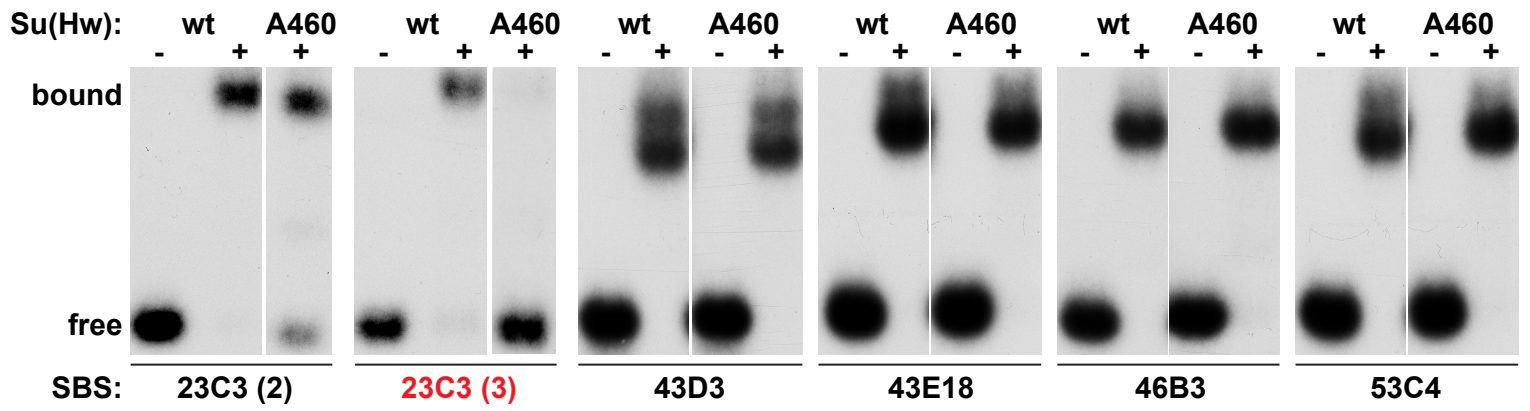
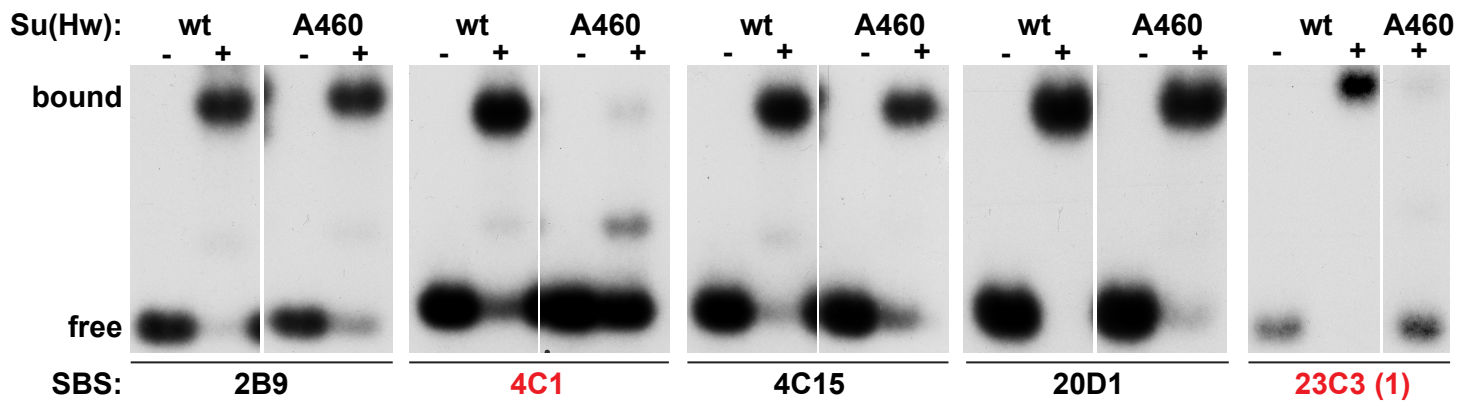


Supplemental Table 1. Amino acid sequence of the twelve Su(Hw) zinc fingers.

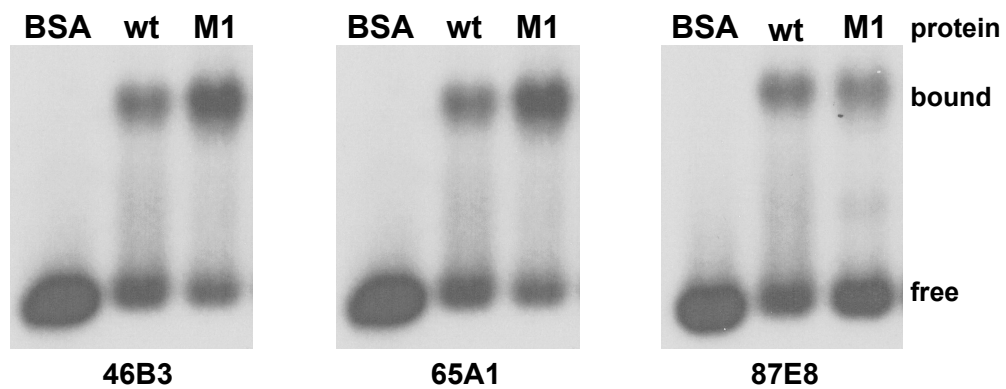
ZF	Zinc Finger Sequence	Spacer (aa)
1	HV <u>C</u> <u>G</u> K <u>C</u> YKTFRRVQSLKK <u>H</u> LEF-- <u>C</u> RY	45
2	IN <u>C</u> PD <u>C</u> PKSFKTQTSYER <u>H</u> IFIT- <u>H</u> SE	3
3	FP <u>C</u> SI <u>C</u> NANLRSEALLAL <u>H</u> EEQ-- <u>H</u> KS	4
4	YA <u>C</u> KI <u>C</u> GKDFTRSUHLKR <u>H</u> QKYSS <u>C</u> SS	5
5	MS <u>C</u> KV <u>C</u> DRVFYRLDNLRS <u>H</u> LKQ-- <u>H</u> LG	8
6	YM <u>C</u> HT <u>C</u> KNCFYSLPTLNI <u>H</u> IRT-- <u>H</u> TG	3
7	FD <u>C</u> DL <u>C</u> DKKFSALVALKK <u>H</u> RRY-- <u>H</u> TG	3
8	YS <u>C</u> TV <u>C</u> NQAFAVKEVLNR <u>H</u> MKR-- <u>H</u> TG	3
9	HK <u>C</u> DE <u>C</u> GKSFIRATQLRT <u>H</u> SKT-- <u>H</u> IR	1
10	FP <u>C</u> EQ <u>C</u> DEKFKTEKQLER <u>H</u> VKT-- <u>H</u> SR	5
11	FS <u>C</u> AE <u>C</u> KRNFRTPALLKE <u>H</u> MDEGK <u>H</u> SP	16
12	TD <u>C</u> AI <u>C</u> DKNFDSSDTLRR <u>H</u> IRTV- <u>H</u> EC	

For each ZF, the cysteine and histidine amino acids involved in zinc binding are bold and underlined. Amino acids involved in DNA binding that correspond to the -1, 2, 3 and 6 positions (left to right) of the recognition helix are highlighted in gray. The spacer column represents the number of additional amino acids (aa) separating each ZF. Dashes have been inserted for ZF sequence alignment.

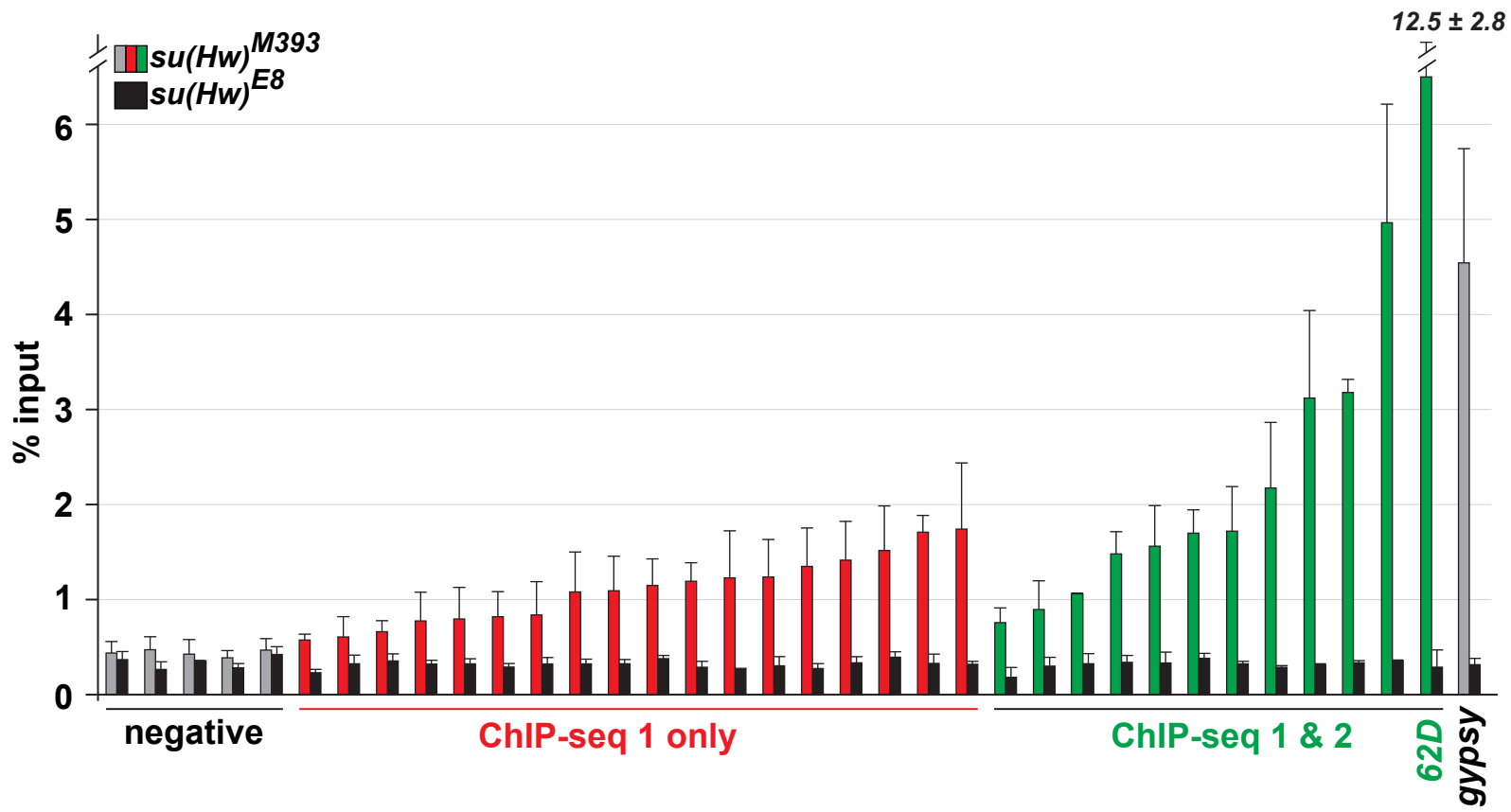
Supp Figure 2.



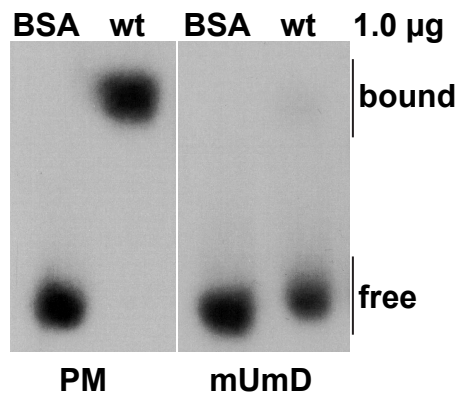
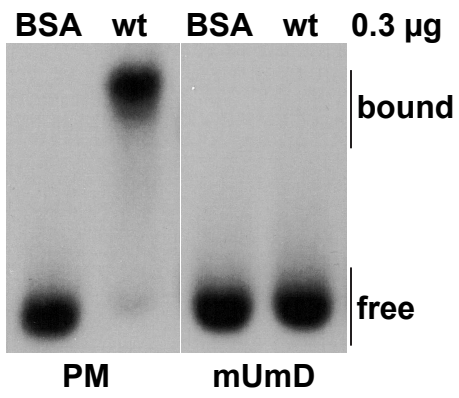
Supp Figure 3.



Supp Figure 4.



Supp Figure 5 .



Supplemental Figures legends

Supplemental Figure 1. Expression of Su(Hw) produced *in vivo* or *in vitro*.

A. Bacterially expressed Su(Hw)^{WT} and Su(Hw)^{ZF} mutants were run on a SDS-polyacrylamide gel and silver stained (left) or transferred to nitrocellulose for western analysis with Su(Hw) antibodies (right). The migration position of the full-length Su(Hw) is indicated by the black triangle on the right. **B.** Western blot analysis of ovary protein extracts obtained from *su(Hw)^{+/+}*, *su(Hw)^{2/v}* and *P[su(Hw)^{MZF}]*, *su(Hw)^{v/2}* females. Blots were probed with antibodies against Su(Hw) and alpha-Tubulin as a loading control.

Supplemental Figure 2. Analyses of *in vitro* binding properties of Su(Hw)^{A460}

Shown are representative EMSA results that compare *in vitro* binding of bacterially purified Su(Hw)^{wt} and Su(Hw)^{A460}. Labeled DNA probes carry endogenous SBSs isolated from genomic regions named for their cytological position. Binding was assayed using 1.0 µg of protein in lanes marked (+). The migration of free and bound DNA is indicated on the left. Four SBSs failed to bind to Su(Hw)^{A460}, indicated in red (4C1, 23C1, 23C3, 100C1)

Supplemental Figure 3. Analyses of *in vitro* binding proteins of Su(Hw)^{M1}.

Shown are representative EMSA results that compare *in vitro* binding of bacterially purified Su(Hw)^{wt} and Su(Hw)^{M1}, with BSA shown as a negative control. Labeled DNA probes carry endogenous SBSs isolated from the indicated cytological positions in the genome. These SBSs were chosen because they represent sites that are lost *in vivo* in

ChIP-qPCR studies of Su(Hw)M1 occupancy (Fig. 5). Binding was assayed using 1.0 μg of wild type and mutant protein. These studies show that Su(Hw)^{wt} and Su(Hw)M1 bind comparably to these SBSs. Further, levels of Su(Hw) protein match those used in other EMSA assays, implying that the observed incomplete shift reflects that these Su(Hw)M1 lost sites are lower affinity SBSs.

Supplemental Figure 4. Comparison of Su(Hw)M4^{M393} ChIP-seq experiments.

Percent input for Su(Hw)M4^{M393} (gray/red/green bars) and Su(Hw)M7^{E8} (black bars, negative control) is shown. SBSs are arranged in increasing order by percent input in two groups: 1) SBSs recovered in ChIP-seq1 only or 2) SBSs recovered in both ChIP-seq1 and ChIP-seq2. Negative controls represent five genomic regions that lack a SBS. Positive control regions (*gypsy*, *62D*) represent SBSs known to associate with Su(Hw)M4. Shown is the average percent input from two biologically independent samples. Error bars indicate standard deviation.

Supplemental Figure 5. EMSA of an SBS lacking the up- and down- stream modules.

Comparison of bacterially expressed Su(Hw)^{WT} binding to the PM or mUmD DNA probes. Binding was assayed using two protein amounts (0.3 μg and 1.0 μg). The migration of free and bound DNA is indicated on the right. BSA serves as a negative control.