Supplementary Figure 2.

. .

The Runt/Bgb heterodimer is required for reporter activation. Growth rates of cells containing different bait combinations were examined under positive selection. In these experiments the α -Runt bait was paired with α -Bgb, α -SH3 or α -HAF, where α -SH3 contains the *crk* SH3 domain and α -HAF contains a *crk* SH3 ligand (PPPALPPKRRR). Neither α -SH3 nor α -HAF are anticipated to facilitate or inhibit the interaction of Runt with its target sequence. These bait combinations were tested with a reporter vector containing either the consensus α -Runt/ α -Bgb binding site (+ = TTGCGGTTT) or with a control reporter (-) containing an unrelated sequence. Each population of cells was serially diluted 10-fold from left to right and plated on 2xYT (top panel) or on NM minimal media containing 2 mM 3-AT (bottom panel). Both plates contain carbinicillin, kanamycin and chloramphenicol to select for cells containing all three plasmids. The left-most spot of cells in each titration contains approximately 10⁶ cells. Robust growth is observed under selective conditions for the α -Runt/ α -Bgb combination with the appropriate prey (+), whereas no significant growth is observed for the other bait/prey combinations. These results are consistent with in vitro data for this complex: the Runt/Bab complex binds efficiently to its recognition sequence, whereas Runt in the absence of Bgb has a lower affinity for its sequence, and Bgb cannot bind DNA by itself¹.

Baits	Prey	
α -Runt/ α -Bgb	+	0000000000
α -Runt/ α -Bgb	-	000000
α -Runt/ α -SH3	+	000000000
α -Runt/ α -HAF	+	
α-Runt/α-Bgb	+	60000
α -Runt/ α -Bgb	-	·····
α -Runt/ α -SH3	+	
α -Runt/ α -HAF	+	2mM 3-AT

1. Golling, G., Li, L., Pepling, M., Stebbins, M. & Gergen, J.P. Drosophila homologs of the proto-oncogene product PEBP2/CBF beta regulate the DNA-binding properties of Runt. *Mol Cell Biol* **16**, 932-942 (1996).