

Distribution of the number of promoter regions bound per regulator (blue). For regulators profiled under multiple conditions, the union of promoter regions bound under all conditions is reported. An average of randomized distributions for the same set of *P* values randomly assigned among regulators and promoter regions is shown in pink.

Harbison et al. Supplementary Figure 2



Overview of motif discovery and assignment. Motifs were identified by applying a suite of motif discovery programs to the intergenic sequences identified by the binding data. The resulting specificity predictions were filtered for significance and then clustered to yield representative motifs. Conservation-based metrics were used to identify the highest-confidence subset of these motifs. For cases in which multiple significant binding motifs were found for a factor, we used statistical scores or information from specificity databases to choose a single motif for each regulator. A complete description of the method can be found in Supplementary Methods.



Comparison of Cin5 binding to two sequences. Recombinant Cin5 was purified from bacteria and incubated with a Cy5-labeled oligonucleotide containing the sequence (gcgacaTTACCTAAgggc) and challenged with one of two unlabeled competitors: the same sequence (lanes 2-8) or the previoulsy published binding site (gcgacaTTACTAAgggc; lanes 9-15). The concentration of each competitor was varied in 3-fold steps. The probe based on our discovered motif was approximately 27-fold better in competing away the shifted band compared to the probe based on the previously published specificity. Similar results were obtained for a probe containing a core sequence of TTACGTAA.



Pairwise comparison of the number of promoter regions bound under two different conditions for 25 regulators (based solely on genome-wide location data with $P \le 0.001$). Dark blue bars represent the number of promoter regions bound under growth in rich medium; light blue bars represent the number of promoter regions bound under growth in amino acid starvation medium.



Quality of Gcn4 binding sites among intergenic regions bound under different conditions. Each intergenic region was scored based on the quality of the best matching subsequence to the Gcn4 binding specificity (TGASTCA). In rich media conditions 68% of the intergenic regions contain high-quality matches to the Gcn4 specificity. Under starvation conditions the levels of Gcn4 protein rise, and the set of bound intergenic regions expands. Of the newly bound regions, only 27% contain high-quality matches. By contrast, only 3% of all intergenic regions contain matches of this quality.