

Functional dissection of protein complexes involved in yeast chromosome biology using a genetic interaction map

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Abstract

Defining the functional relationships between proteins is critical for understanding virtually all aspects of cell biology. Large-scale identification of protein complexes has provided one important step towards this goal; however, even knowledge of the stoichiometry, affinity and lifetime of every protein-protein interaction would not reveal the functional relationships between and within such complexes. Genetic interactions can provide functional information that is largely invisible to protein-protein interaction data sets. Here we present an epistatic miniarray profile (E-MAP) consisting of quantitative pairwise measurements of the genetic interactions between 743 *Saccharomyces cerevisiae* genes involved in various aspects of chromosome biology (including DNA replication/repair, chromatid segregation and transcriptional regulation). This E-MAP reveals that physical interactions fall into two well-represented classes distinguished by whether or not the individual proteins act coherently to carry out a common function (Fig. 1). Thus, genetic interaction data make it possible to dissect functionally multi-protein complexes, including Mediator, and to organize distinct protein complexes into pathways. In one pathway defined here (Fig. 2), we show that Rtt109 is the founding member of a novel class of histone acetyltransferases responsible for Asf1-dependent acetylation of histone H3 on lysine 56. This modification, in turn, enables a ubiquitin ligase complex containing the cullin Rtt101 to ensure genomic integrity during DNA replication.

Figure 1: **a**, ROC curves comparing the power of the genetic interaction patterns—using a score (the COP score) that accounts for both direct genetic interactions and patterns of interactions (red)—and large-scale affinity purification data (blue)—using a recent re-analysis of raw purification data—to predict co-membership of pairs of proteins in the same physical complex. The slope of the initial portion of each curve serves as a measure of the score's maximal accuracy. **b**, Distribution of direct genetic interaction scores for pairs of genes encoding physically interacting proteins (green) and non-interacting proteins (black). **c**, Distribution of the Pearson's correlation coefficients between the interaction patterns for the same sets of gene pairs as in **b**.

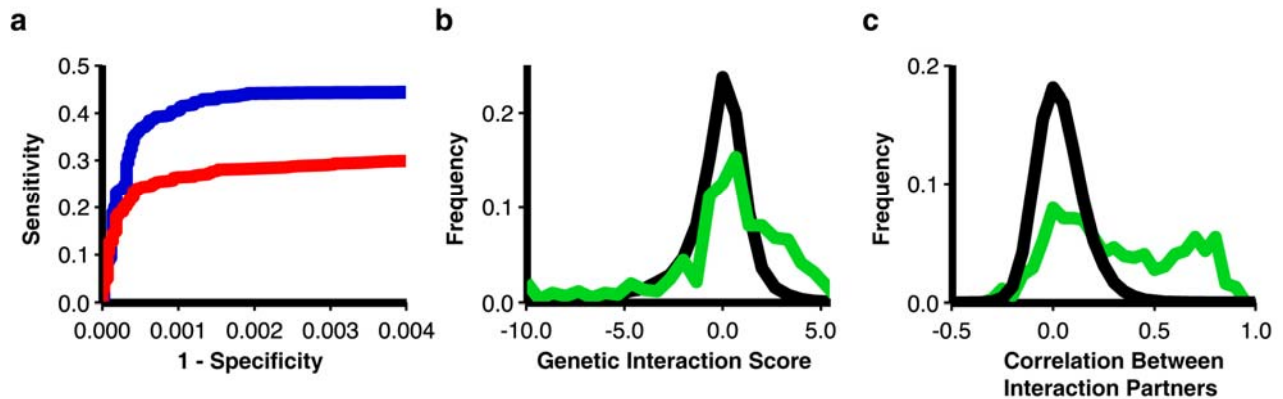


Figure 2: **a**, A subset of the genetic interaction patterns for the proposed members of a histone H3 K56 acetylation pathway: *RTT101*, *MMS22*, *MMS1*, *RTT109* and *ASF1*. Blue indicates negative (or synthetic) genetic interactions where double mutants grow more slowly than would be expected based on the single mutant phenotypes. Yellow indicates positive (or alleviating) genetic interactions where the double mutant grows more rapidly than expected. Note the positive interactions between pairs within the pathway (rightmost). Also note that that *ASF1*, but not the others, has negative interactions with histones and the chromatin assembly complex CAF-I, suggesting that *Asf1*'s role in chromatin assembly is independent of the other four genes. On the other hand, interactions with DNA damage repair components are common to all five genes. **b**, Model for the histone H3 K56 acetylation signaling pathway.

