The landscape of genetic complexity across 5,700 gene expression traits in yeast

Supporting Text

Simulations. For each simulated transcript, we initially assigned all segregants to have an expected transcript level of 0 and added normally distributed noise whose variance $s_e^2$ we set, without loss of generality, to be 1. Then, at each locus, we randomly assigned segregant genotypes as either "high" or "low." For each "high" allele inherited by a segregant, we increased the transcript level by the quantitative trait locus (QTL) effect size $x$. For each simulated transcript, we assigned $x$ to reflect the heritability of one transcript from the real data, as follows. We calculated the heritability of each real transcript as $h^2 = (s_s^2 - s_p^2)/s_s^2$, where $s_s^2$ and $s_p^2$ are the variance among phenotype values in the segregants and the pooled variance (1) among parental measurements, respectively. Because the variance in parental measurements comes only from measurement noise, in the simulation we set $s_p^2 = s_e^2 = 1$. Following ref. 2, to model $n$ loci of equal additive effect we wrote $s_e^2 = nx^2/4 + s_s^2$, where $x$ is the QTL effect size, and solved for the unknown $x$ in terms of the known $h^2$.

Alternatively, for a model in which a single main QTL explained a fraction $y < 1$ of the genetic variance, we wrote $s_e^2 = x^2/4 + as_s^2$, where $a$ is the factor by which additional unspecified genetic variation inflates the remaining variance above measurement error; we solved for the unknowns $x$ and $a$ in terms of $y$ and $h^2$. In all simulations, the variance of measurement noise set the scale of the simulation, i.e., the QTL effect size relative to this variance ensured a heritability consistent with the real data.

Optimal choice of test for transgressive segregation. To choose the test with the highest power to detect transgression, we developed a general power simulation of transgressive traits controlled by two loci. In this simulation, for each transcript, we initially assigned all segregants to have an expected transcript level of 0 and added normally distributed noise whose variance $s_e^2$ we set, without loss of generality, to be 1. Then, at each locus, we randomly assigned segregant genotypes as either "high" or "low." For each "high" allele inherited by a segregant, we added to the transcript level the QTL effect size $x$. For the first locus $x = a$, and for the second locus $x = -b$. When $a = b$, effects of the QTLs canceled in each parent; then, both parents had similar phenotypes, which made transgressive segregation easier to detect by our tests. Transgression was also easier to detect for larger values of $a$ relative to the noise variance. Thus, the two parameters $a$ and $a/b$ determined the transgressive behavior of the model. We ran 1,000 simulations for various values of $\{a, b\}$. Then we applied the transgression tests derived from the real data to have equal FDR of 0.05 (see main text: $d = 1.0$, $d = 58$; $d = 1.5$, $j = 35$; $d = 2.0$, $j = 21$; $d = 2.5$, $j = 13$; and $d = 3.0$, $j = 8$) and asked how many simulated transcripts passed each test. Fig. 5 shows that the test requiring 21 segregants to be at least two standard deviations outside the means of the parents gave approximately maximal power across a range of parameters. We saw similar results in simulated traits controlled by three or four QTLs of opposite sign and when we varied $a$ and $b$ (data not shown).

Epistasis. Given $n_s$ segregant trait values, $n_R$ measurements in RM11-1a (RM), and $n_B$ measurements in BY4716 (BY), we computed the mean trait value in each group of data, $m_S$, $m_R$, and $m_B$, and the corresponding variances, $s_s^2$, $s_R^2$, and $s_B^2$. Next we computed $D$, the difference between the segregant and midparent means, as $m_S - (m_R + m_B)/2$. We computed the squared standard error of the mean, SSEM, for this quantity as $s^2_S/n_S + (s^2_R/n_R + s^2_B/n_B)/4$. We then formed the $t$ statistic $D$/sqrt(SSEM), which allowed a test (with $n_S + n_R + n_B - 4$ degrees of freedom) for the significance, $P$, of a $D$ value relative to its expected sampling variance. For the permutation test, we computed the significance of the $t$ statistic in each such permuted trait; the total number of such null traits with $P$ less than or equal to a given threshold $P_0$ represented the genome-wide false positive count at $P_0$. The false discovery rate was computed as the ratio between the estimated false positive count at $P_0$ and the number of real transcripts with $P > P_0$. In simulations, genotypes, parental phenotypes, the variance of measurement noise $s_e^2$, and QTL effect sizes $x$ were assigned to mimic heritabilities from the real data, analogous to the additive simulations (see above and Materials and Methods). We first modeled the nonadditive case in
which segregants with either parental allele combination all had one expression level and those with nonparental combinations all had another. This models, for example, coevolution of multiple protein subunits in each parent, which no longer fit together into a functional complex when mixed. For each transcript all segregants with parental combinations of alleles had a mean expression level of 0; all segregants with nonparental combinations of alleles had a mean expression level of $x$. Under this model, for two loci, the variance across all segregants $s^2 = x^2/4 + s_e^2$, and for three loci, $s^2 = 3x^2/16 + s_e^2$. We also simulated traits under a model in which segregants with one parental allele combination had one expression level, and all other segregants had another. This models, for example, mutations in multiple protein subunits in one parent. Here, segregants with one parent’s combination of alleles had a mean expression level corresponding to the QTL effect size, and all other segregants had a mean expression level of 0. Under this model, for two loci $s^2 = 3x^2/16 + s_e^2$, and for three loci, $s^2 = 7x^2/64 + s_e^2$. For each simulated transcript, we applied the epistasis test using the FDR = 0.05 significance cutoff from the real data. Results in the main text are the average of 10 independent simulations.
