103. High--throughput genomic studies using CRISPRi

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We recently showed that dCas9 can be used as a transcriptional-control tool in a variety of species, ranging from bacteria to mammals. We have optimized this technology and adapted it to high throughput studies. Using next-generation sequencing and a library of oligos that targets >10000 annotated functional elements in the E. coli genome (coding sequences, promoters, transcription factor binding sites, small RNAs), we were able to investigate the effect of all 10000 features in a single tube, in a single experiment. Our results agree well with published results on coding sequence effects. Because our system is based on inducible phenotypes, however, we are able to investigate the effect of important genes in any condition. We demonstrate this by finding that while NrdA and NrdB are essential aerobically (and thus generally classified as essential by existing databases), they are dispensable anaerobically. Our data also provides a rich source of information about regulatory effects in E. coli which allows us, for example, to ascribe deleterious effects of transcription factor knock-downs to particular binding sites. Our technology can be easily applied to any organism in which dCas9 works, and thus should prove of general interest as a high-throughput discovery tool.