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ance to a wide range of modifications within the recognition sequence. MvaI shares ~20% sequence identity and structural similarity with BcnI, a REase recognizing the related pseudopalindromic sequence CC/SGG (S stands for G or C).

Here, we show that MvaI has two specificities: in addition to cutting its well-known recognition site (CC↓AGG/CC↑TGG), it can nick the related CC↓GGG/CCCGG sequence (BcnI site) if the underlined cytosines are C5-methylated (CC↓GGG/CCCGG). The single-strand scission occurs in the G-strand as indicated. At sequences, where two oppositely oriented methylated BcnI sites partially overlap (SmaI sites), double-nicking leads to double-strand cleavage ($CC^{m5}C$↓GGG/$CC^{m5}C$↑GGG), generating fragments with blunt ends. The double-strand cleavage rate at these sites is ~five to tenfold lower than at the canonical target sites.

MvaI is the first restriction enzyme, for which activity on an unmethylated as well as on a methylated substrate site has been shown. The new, methylation-dependent activity represents nicking and double-stranded cleavage specificities ($C^{m5}C$↓GGG/$CC^{m5}C$GG and $CC^{m5}C$↓GGG/$CC^{m5}C$↑GGG, respectively) not known before.

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Automated refinement of a genome-scale metabolic model of yeast based on high-throughput genetic interaction data

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Genome-scale stoichiometric models are *in silico* representations of the metabolism of living organisms. Genetic and conditional changes can be introduced to these models and the effect of these modifications can be investigated. Certain phenotypes, like single gene deletion, can be predicted with high accuracy; however, it has remained untested whether the metabolic models can also successfully capture genetic interactions (*i.e.* non-independence between mutation effects). Genetic interactions have two major forms: positive (or alleviating) and negative (or aggravating) epistasis. Positive interactions occur when deletion of two genes simultaneously has a higher fitness than would be expected based on the fitness effect of the single mutations (e.g. if two genes are in the same linear pathway the second mutation has no additional fitness effect). Similarly, genes show negative interaction when the double mutant has significantly lower fitness than the combined effect of the single deletions (e.g. synthetic lethality, when the single mutants are viable but the joint deletion of the genes is lethal).

To determine the in silico model's accuracy to predict genetic interactions we systematically compared computational predictions with a unique genetic interaction dataset generated, as part of a collaboration, by the Boone lab[1] and which comprises ~185,000 metabolic gene pairs. In the *in silico* analyses we used a *Saccharomyces cerevisiae* metabolic reconstruction containing 904 genes (iMM904)[2]. The metabolites and reactions are represented by their stoichiometric coefficients and the model contains information on reaction reversibility, however, it does not incorporate kinetic details. We applied the widely used flux balance analysis (FBA) modelling tool to compute mutant fitness (*i.e.* biomass production efficiency).

We found that several properties of the *in vivo* genetic network were successfully captured by the model (e. g. single mutants with severe fitness defects tend to show many genetic interactions); however, it recovered only a minority of experimentally observed interactions.

Because our knowledge of metabolism is certainly imperfect, we sought to improve the prediction performance of the metabolic model and developed an optimization-based algorithm to automatically refine the network based on empirical genetic interaction data. Our method suggested several modifications and we experimentally verified some of them. For example, the essentiality of the kynurenine pathway genes (*BNA1*, *BNA2*, *BNA4*, and *BNA5*) in the absence of nicotinic acid was undetected by the original model due to the erroneous presence of a NAD biosynthesis route. Finally, based on our algorithm and on literature data, we substantially revised the NAD biosynthesis pathway of the genome-scale metabolic reconstruction of yeast.

Our work has recently been accepted for publication in *Nature Genetics*.

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