Modeling isotopomer distributions in biochemical networks using isotopomer mapping matrices

Within the last decades NMR spectroscopy has undergone tremendous development and has become a powerful analytical tool for the investigation of intracellular flux distributions in biochemical networks using C-13-labeled substrates. Not only are the experiments much easier to conduct than experiments employing radioactive tracer elements, but NMR spectroscopy also provides additional information on the labeling pattern of the metabolites. Whereas the maximum amount of information obtainable with C-14-labeled substrates is the fractional enrichment in the individual carbon atom positions, NMR spectroscopy can also provide information on the degree of labeling at neighboring carbon atom positions by analyzing multiplet patterns in NMR spectra or using 2-dimensional NMR spectra. It is possible to quantify the mole fractions of molecules that show a specific labeling pattern, i.e., information of the isotopomer distribution in metabolite pools can be obtained. The isotopomer distribution is the maximum amount of information that in theory can be obtained from C-13-tracer studies. The wealth of information contained in NMR spectra frequently leads to overdetermined algebraic systems. Consequently, fluxes must be estimated by nonlinear least squares analysis, in which experimental labeling data is compared with simulated steady state isotopomer distributions. Hence, mathematical models are required to compute the steady state isotopomer distribution as a function of a given set of steady state fluxes. Because $2^n$ possible labeling patterns exist in a molecule of n carbon atoms, and each pattern corresponds to a separate state in the isotopomer model, these models are inherently complex. Model complexity, so far, has restricted usage of isotopomer information to relatively small metabolic networks. A general methodology for the formulation of isotopomer models is described. The model complexity of isotopomer models is reduced to that of classical metabolic models by expressing the $2^n$ isotopomer mass balances of a metabolite pool in a single matrix equation. Using this approach an isotopomer model has been implemented that describes label distribution in primary carbon metabolism, i.e., in a metabolic network including the Embden-Meyerhof-Parnas and pentose phosphate pathway, the tricarboxylic acid cycle, and selected anaplerotic reaction sequences. The model calculates the steady state label distribution in all metabolite pools as a function of the steady state fluxes and is applied to demonstrate the effect of selected anaplerotic fluxes on the labeling pattern of the pathway intermediates. (C) 1997 John Wiley & Sons, Inc.