Maintenance of the monomeric structure of glucokinase under reacting conditions

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Glucokinase is a monomeric protein under native and denaturating conditions yet presents a sigmoidal saturation function for glucose. These peculiarities suggested the possibility that polymerization occurs under assay conditions. Thus the apparent molecular weight of glucokinase was determined by gel filtration at 4 °C and at 30 °C in the presence of substrates and products, singly and in combination, creating during the filtration similar conditions as used in the assay. Gel filtration was performed also in the presence of N-acetylglucosamine, which is a competitive inhibitor and shifts to an hyperbole the saturation function for glucose. The same elution behavior, that is, a single symmetrical peak, was observed in every system used. This persistent monomeric form of glucokinase excludes the possibility that the sigmoidal function is the result of the interaction of different subunits. The possibility of an association-dissociation equilibrium in which the kinetic properties of the