Complex I (NADH:ubiquinone oxidoreductase) from mitochondria is a complicated membrane-bound, energy-transducing enzyme, which contains 45 different subunits and 9 redox cofactors. The overall enzyme mechanism comprises NADH oxidation at a flavin mononucleotide, intramolecular electron transfer via a series of iron-sulphur clusters, quinone reduction, and proton translocation. In order to define the overall mechanism we aim to begin by defining the mechanism of NADH oxidation, and then to build the complete mechanism in a stepwise fashion. This talk will describe results from experiments using artificial electron acceptors, such as hexaammine ruthenium III, which are used to accept the electrons from NADH oxidation as rapidly as possible. Progress towards interpreting the data to construct a comprehensive picture of the reaction, using steady-state analysis and numerical simulation of candidate mechanisms, is described.