Oxidation of NADH by a rotenone and antimycin-sensitive pathway in the mitochondrion of procyclic Trypanosoma brucei brucei

Abstract:

The pathway of NADH oxidation in the procyclic Trypanosoma brucei brucei was investigated in a crude mitochondrial membrane fraction and in whole cells permeabilized with digitonin. NADH:cytochrome c reductase activity was 75% inhibited by concentrations of antimycin that inhibited 95% succinate:cytochrome c reductase activity suggesting that the major pathway for NADH oxidation in the mitochondria involved the cytochrome bc1 complex of the electron transfer chain. Both NADH:cytochrome c and NADH:ubiquinone reductase activities were inhibited 80-90% by rotenone indicating the presence of a complex I-like NADH dehydrogenase in the mitochondrion of trypanosomes. In whole cells permeabilized with low concentrations of digitonin, the oxidation of malate, proline and glucose (in the presence of salicylhydroxamic acid, the inhibitor of the alternate oxidase) was inhibited 30-50% by rotenone. The presence of an alternative pathway for NADH oxidation involving fumarate reductase was indicated by the observation that malonate, the specific inhibitor of succinate dehydrogenase, inhibited 30-35% the rate of oxygen uptake with malate and glucose as substrates in the digitonin-permeabilized cells. We conclude that in the mitochondrion of the procyclic form of T. brucei, NADH is preferentially oxidized by a rotenone-sensitive NADH:ubiquinone oxidoreductase; however, NADH can also be oxidized to some extent by the enzyme fumarate reductase present in the mitochondrion of T. brucei.