of protons and atoms as shown in equation 1. Thus, a synchronous shifting of protons should occur with the breaking of the P–S and formation of the P–O bonds. A system which combines the acid and base in a single unit capable of such synchronous action may well be the reason for the efficiency of enzyme catalysis.

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Isolation of a quinone from beef heart mitochondria*

From lipid extracts of beef heart mitochondria we have isolated a new compound capable of undergoing reversible oxidation and reduction. The absorption spectrum of the oxidized and reduced forms are shown in Fig. 1. The oxidation-reduction behaviour as well as the infrared spectrum indicate that the compound is a quinone. For convenience it will be referred to as Q-275. A compound with similar spectral properties has been observed also in beef liver mitochondria. This yellow-orange, crystalline material has been recrystallized from several solvents to a constant melting point (48–49° C) and to a constant extinction coefficient. The purity of the compound is being investigated by chromatographic procedures. Q-275 is insoluble in water, but is soluble in most lipid solvents.

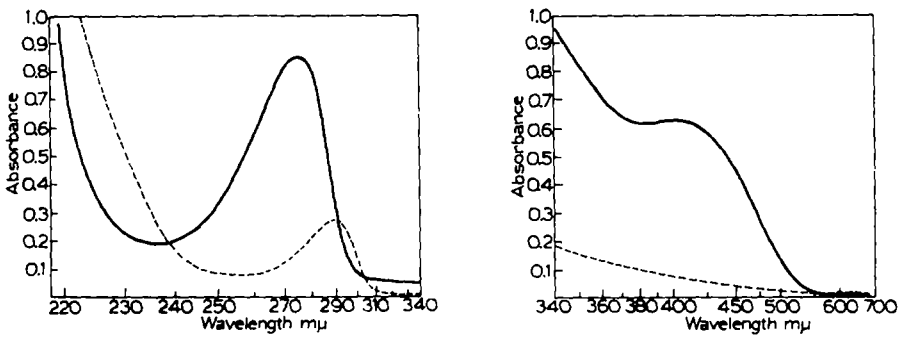

Fig. 1. Absorption spectrum of Q-275 in absolute ethanol. Solid line represents oxidized form, and the dotted line represents the spectrum obtained after shaking with a few grains of KBH_4. Concentrations used for 1 cm path in mg/ml: Ultraviolet range, 0.0425; visible range, 0.75.

In addition to beef heart mitochondria, Q-275 has been found in various electron transporting particles derived from these mitochondria. The concentration of Q-275 (mg/g protein) was found to be as follows: Mitochondria[1], 2.5; ETP[1], 2.7; SDC[2], 6.0; green fraction[3], 0.5. The presence of Q-275 appears to be correlated with succinate oxidizing capacity.

That Q-275 is involved in the electron transport activities of the aforementioned particles is indicated by several lines of evidence.

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I. After exposure of mitochondria to air, the extracted compound is observed to be in the oxidized form. After a short incubation in the presence of succinate and cyanide, the compound then appears in the reduced form. This reduction is inhibited by Antimycin A. The same effect cannot be duplicated by similar incubation of the particles with DPNH.

II. It has been possible to show that the oxidation and reduction of externally added Q-275 is catalyzed by mitochondria and derivative particles. Reduction of added Q-275 by succinate occurs in the presence of mitochondria, ETP and SDC; by DPNH in presence of mitochondria and ETP; and by pyruvate plus malate in presence of mitochondria. The reduction occurs only in the presence of cyanide or under anaerobic conditions. The oxidation of the chemically reduced compound is catalyzed by mitochondria or ETP. This oxidation is inhibited by cyanide. The reduced compound is oxidized by ferricytochrome *c* in the presence of ETP or SDC.

III. When mitochondria or ETP are extracted with heptane or 2,2,4-trimethylpentane, Q-275 appears in the organic solvent. This extraction decreases the succinoxidase activity; addition of Q-275 restores the initial activity. Although no detectable cytochrome *c* is released from the particles by this treatment, addition of cytochrome *c** also restores activity (Table I). The restored succinoxidase system is completely inhibited by antimycin A at 0.002 mg/1.68 mg protein. Other compounds which were tested as substitutes for Q-275 either had no effect or inhibited the low blank rate. The compounds** tested which had no effect were *p*-xyloquinone; menadione; 2-hydroxy-1,4-naphthoquinone; 3-methyl-1,2-naphthoquinone; bovine serum albumin; *d*-α-tocopherol; vitamin K_1; β-carotene; 2,3-dimethyl-1,4-naphthoquinone, 2-undecyl-1,4-naphthoquinone; and tuna fish oil. The blank rate was inhibited by lapachol, norlapachol and norlapachol acetate. All these compounds were added at concentrations comparable to that of Q-275.

TABLE I

EFFECT OF Q-275 ON SUCCINOXIDASE

Enzyme treatment	*Additions*	*Activity* μatoms oxygen.5 min mg
None	—	2.5
None	Q-275, 0.088 mg	2.5
None	cytochrome *c*, 1 mg	2.6
Extracted	—	0.4
Extracted	Q-275, 0.088 mg	2.7
Extracted	cytochrome *c*, 1 mg	2.9

Assay for succinoxidase at 38° as previously reported[1] except air used as gas phase. 2.5 mg of ETP, and 1.58 mg of extracted ETP used in a total volume of 1.5 ml. Q-275 added in 0.03 ml ethanol.

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