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Antioxidant Function of Coenzyme Q
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I. INTRODUCTION

Coenzyme Q (CoQ) or ubiquinone is distributed ubiquitously in the natural kingdoms. CoQ homologs (CoQn) containing 9 and 10 isoprene units occur in mammals, which coexist in different amounts depending on the species. CoQ9 is the predominant form in mouse and rat, and CoQ10 is predominant in rabbit, guinea-pig, dog, pig and man [1-3].

CoQ plays an important role in ATP synthesis as an electron-carrying component of the mitochondrial respiratory chain. In recent years, increasing attention has focused on CoQn as an antioxidant both in vitro and in vivo, and the antioxidant activity of CoQn is exhibited by its reduced form, CoQnH2 [4, 5]. There are some excellent recent reviews on the function of CoQnH2 in this area [4-6].

In view of all the facts reported relatively little is known about the mechanism of the antioxidant function of CoQnH2 at cellular and biomembrane level. It is not yet known whether CoQ9H2 and CoQ10H2 coexisting in the cell play the same or different antioxidative roles and further, whether CoQnH2 acts alone or in concert with other cellular antioxidants, such as α-tocopherol.

To solve these problems, we have carried out a series of in vivo experiments [7-11], in which antioxidative action of endogenous CoQnH2 was determined, together with the protective effect of CoQ10 accumulated and reduced to CoQ10H2 in the tissue after injection on peroxidative reperfusion injury.

Then, we have investigated antiperoxidative activity of endogenous CoQ9H2 and CoQ10H2 in rat and guinea-pig hepatocytes exposed to a water-soluble radical initiator, 2,2'-azobis(2-amidinopropane)dihydrochloride (AAPH). The results obtained indicate a difference in the antioxidant activity between CoQ9H2 and CoQ10H2, together with the action of CoQnH2 independent of α-tocopherol.

II. EVIDENCE FOR in vivo ANTIOXIDATIVE ACTIVITY OF REDUCED CoQ

Good evidence for in vivo antiperoxidative activity of CoQnH2 was obtained from studies with experimental endotoxemia in mice [7, 8]. Endotoxin-induced lipid peroxidation in the liver was found to be associated with drastic decreases in hepatic levels of CoQ9H2, α-tocopherol, and reduced glutathione (GSH). Administered CoQ10 (oxidized form), after accumulation in the liver and reduction to CoQ10H2, prevented the decreases in endogenous antioxidants, such as CoQ9H2, α-tocopherol and GSH, completely suppressed lipid peroxidation, and markedly increased the survival rates of endotoxemic mice.

Other findings indicating in vivo antioxidant activity of CoQnH2 include decreased tissue levels of CoQ9H2, CoQ10H2, α-tocopherol and GSH, which was accompanied by the increases in tissue and mitochondrial lipid peroxides in rat liver following ischemia and reoxygenation [9, 10]. Administered CoQ10 has been shown to prevent the decreases in these endogenous antioxidants after reduction...
to CoQ$_{10}$H$_2$ in the liver, and to protect against acute postischemic hepatic injury in a similar mechanism to that found in the experimental endotoxemia. In another model experiment on canine heart preservation and the following reperfusion, the antioxidative activity of administered CoQ$_{10}$, was also demonstrated [11]. The accumulated CoQ$_{10}$H$_2$ protected the organ against reperfusion injury and maintained left ventricular functions to almost the normal level by suppressing the stimulated lipid peroxidation and by preventing the decreases in endogenous CoQ$_9$H$_2$ and α-tocopherol.

III. ANTIOXIDANT ACTIVITY OF CoQ HOMOLOGS IN RADICAL-INDUCED HEPATOCYTE INJURY

As described above, CoQ$_9$H$_2$ and CoQ$_{10}$H$_2$ coexisting in animal cells, especially hepatocytes, are not known whether they have the same antioxidant activity or not. An approach to studying possible differences in the antioxidant role of CoQ homologs in animal cells is to know differences in subcellular distributions. Subcellular fractionation of rabbit liver revealed that most CoQ$_9$ existed in the cytosolic fraction, and most CoQ$_{10}$ existed in the mitochondrial fraction [3]. In addition, in rabbit liver %CoQ$_9$H$_2$ in total (oxidized plus reduced) CoQ$_9$ increased with growth at a rate much slower than that of %CoQ$_{10}$H$_2$, since CoQ$_{10}$ in mitochondria is expected to be more easily reduced by the respiratory chain than CoQ$_9$ in cytosol. These results suggest possible differences in the role of CoQ$_9$ and CoQ$_{10}$ in mammalian cells, together with the mechanism of antioxidant activity of CoQ homologs.

A water-soluble radical initiator, AAPH, induces peroxidative cellular damage from the outside of the cell [12, 13]. The concentration of CoQ$_9$H$_2$ in rat hepatocytes containing total CoQ$_9$ and CoQ$_{10}$ at a ratio of 6:1 decreased linearly after the addition of AAPH with a reciprocal increase in CoQ$_9$ (oxidized form). Although CoQ$_{10}$H$_2$ tended to decrease slightly during the incubation with AAPH, the change was not significant. No loss of cell viability or an increase in lipid peroxidation was observed until most of CoQ$_9$H$_2$ was consumed [14].

In guinea-pig hepatocytes containing total CoQ$_9$ and CoQ$_{10}$ at a ratio of 1:5, the consumption of CoQ$_{10}$H$_2$ was accompanied by an increase of TBARS when incubated with AAPH, indicating the effective antioxidant activity of CoQ$_{10}$H$_2$ in these cells. At the same time, complete consumption of cellular CoQ$_9$H$_2$ was also observed. The viability of AAPH-treated guinea-pig hepatocytes remained comparable to that of the control until consumption of CoQ$_{10}$H$_2$ and CoQ$_9$H$_2$ reduced their levels to about 20% of the original. If the insignificant consumption of CoQ$_{10}$H$_2$ in AAPH-treated rat hepatocytes is compared with the rapid consumption of CoQ$_9$H$_2$ in the guinea-pig cells, CoQ$_9$H$_2$ appears to be a potent antioxidant, with activity independent of its concentration [14].

IV. ANTIOXIDANT ACTIVITY OF REDUCED COENZYME Q HOMOLOGS IN RELATION TO THAT OF α-TOCOPHEROL

α-Tocopherol is a good chain breaking antioxidant in biomembranes. Recent reports have proposed some explanations for the mechanism of CoQ$_{H2}$ antioxidative activity in relation to the action of α-tocopherol; (a) CoQ$_{H2}$ directly scavenges lipid peroxyl radicals as effective as α-tocopherol [15], and (b) CoQ$_{H2}$ acts through recycling of α-tocopherol [16,17].

In hepatocytes treated with AAPH, α-tocopherol was consumed with time at the rate comparable to that of either CoQ$_9$H$_2$ in rat hepatocytes or CoQ$_{10}$H$_2$ and CoQ$_9$H$_2$ in guinea-pig hepatocytes [14] as shown in Fig. 1. This suggests that
both antioxidants act independently in the hepatocytes when AAPH induces cellular injury.

Fig. 1. Effect of AAPH on the concentrations of endogenous CoQ₉H₂, and CoQ₁₀H₂ and α-tocopherol in rat and guinea-pig hepatocytes. The values are expressed as percentage of the concentrations obtained at 0-time incubation [14].

SUMMARY

Evidence for in vivo antioxidative activity of reduced CoQ homologs has been presented. This came from studies with experimental endotoxemia in mice, reoxygenation of rat liver following ischemia, and reoxygenation of canine heart following 24-hour cold preservation. In radical-induced injury of hepatocytes, it has been first shown that reduced CoQ₉ acts as a potential antioxidant regardless of its cellular concentration, whereas reduced CoQ₁₀ acts in cells containing CoQ₁₀ as the predominant homolog. The antioxidant activity of reduced CoQ homologs appears to be independent of that of α-tocopherol under the conditions employed.
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