ENHANCED ORAL BIOAVAILABILITY OF UBIQUINONE (COENZYME Q10) FORMULATED IN EMULSOME™ DRUG DELIVERY SYSTEM

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ABSTRACT

Coenzyme Q10 (Ubiquinone), a fat-soluble natural antioxidant with potential use as adjuvant therapy in heart diseases and to protect neurons against age-related degeneration, is a very lipophilic compound. It was formulated using the Emulsome™ drug delivery system as a freze-dried solid dosage form in hard gelatin capsules. The Emulsome-encapsulated Coenzyme Q10 showed better in vitro drug release and enhanced oral bioavailability compared to a commercial free-drug formulation.

1. INTRODUCTION

Ubiquinone or Coenzyme Q10 (CoQ10) is a naturally occurring coenzyme involved in electron transport in the mitochondria. Coenzyme Q10 is classed as a fat-soluble quinone and is an essential component of the mitochondrial respiratory chain constituting a redox-link between flavoproteins and cytochromes and acting as an electron shuttle controlling the efficiency of oxidative phosphorylation. Although known for many years to protect biological membranes against oxidation, ubiquinone has recaptured interest as a natural, lipid-soluble antioxidant which acts as a membrane stabilizing agent, for avoiding lipid peroxidation and regulating lipid fluidity probably by removing free radicals [1]. Ubiquinone has been shown to protect cultured cerebellar neurons against age-related and excitotoxin-induced degeneration [2]. The efficacy of Coenzyme Q10 as adjuvant therapy in heart diseases in a multicenter study has been reported [3].

CoQ10 is a very lipophilic compound and practically insoluble in water due to its long side chain of 10 isoprenoid units. The oral bioavailability of CoQ10 is generally very low and was found to be related to the dissolution rate of the formulation. Emulsions and microemulsions have been shown to be advantageous as vehicles for the oral delivery of lipophilic drugs resulting in improved oral bioavailability of water-insoluble compounds [4,5].

The objective of the present work was to examine the oral bioavailability of CoQ10 formulated using the Emulsome™ technology, a recently developed drug delivery system [6,7,8]. Emulsomes are a new type of lipid particles considered as an intermediate or "hybrid" system between liposomes and oil-in-water emulsions. Emulsome particles have a new type of lipid assembly comprising a hydrophobic core, as in standard oil-in-water emulsions, but surrounded and stabilized by one or more phospholipid bilayers as in liposomes. The Emulsome technology represents a new entity as lipoidal drug vehicle and its successful development was achieved by the incorporation of a relatively high lecithin content (5-10%) compared to standard emulsions (0.5-2%), the use of fats or triglycerides which are solid at room temperature instead of oils, and the utilization of high pressure emulsification technique. The combination of the specific lipid composition and manufacturing technology results in the formation of stable lipid particles in the submicron range.

2. MATERIALS AND METHODS

2.1 Materials

Ubiquinone (Coenzyme Q10) was obtained from Global Marketing Associates, Inc. (San Francisco, CA). D-α tocopherol succinate was purchased from Merck (Germany). Lecithin was from Lipoid KG (Germany). Solid triglycerides were obtained from Hulls (Germany).

2.2 Methods

2.2.1 Determination of CoQ10

CoQ10 was determined in commercial product, Emulsome formulation, and in release medium of in vitro study by extraction with Dole reagent (isopropanol:heptane:water, 45:36:17) and measuring absorbance at 270nm using a calibration curve. CoQ10 samples (0.5ml) were added to 3.5ml of Dole reagent and mixed thoroughly and the two phases were allowed to separate for 10 min at room temperature. CoQ10 was extracted selectively in Dole heptane upper phase which was transferred to a quartz cuvette for absorbance measurement.

2.2.2 Analysis of CoQ10 in Plasma Samples

CoQ10 in plasma samples was identified and quantified by HPLC [9]. Blood samples were drawn into plastic test tubes containing EDTA. Plasma was separated by centrifugation in a non-cooled centrifuge and stored at -20° C till analyzed. CoQ10 was extracted from plasma with hexane. After evaporation to dryness, samples were dissolved in isopropanol for HPLC. The mobile phase consisted of methanol:isopropanol (4:1). Detection was carried out by a UV detector at 275nm wavelength.

2.2.3 Formulation of CoQ10 in Emulsomes

CoQ10 was dissolved together with the lipid ingredients (phospholipids, tocopherol succinate and solid triglycerides) in dichloromethane. The solvent was evaporated until complete dryness, and the dry drug-lipid mixture was then hydrated with the aqueous phase by mechanical shaking. The resultant lipid dispersion was consequently homogenized by high-pressure homogenization (800 bar) using an Emulsiflex™ C-30 high pressure homogenizer (Avestin Inc., Canada) to reduce the particle size to the submicron range. To the resultant Emulsome-CoQ10 preparation a cryoprotectant material was added and the formulation was then freeze-dried using a Christ lyophilizer (Germany). The final Emulsome-CoQ10 dry powder was filled into hard gelatin capsules.

2.2.4 In vitro Release of CoQ10

In vitro drug release of CoQ10 from Emulsome-CoQ10 formulation and commercial product containing equivalent amounts of CoQ10 were determined by placing a hard gelatin capsule in 50ml of simulated gastric fluid (150mM NaCl, pH 1.2, 37° C) containing 1% Tween 80 as sink. Gentle stirring was provided by a magnetic bar. Samples were drawn from the release medium at prefixed time intervals, filtered through a 2.7 μm Whatman GF filter and analyzed for CoQ10 concentration using the UV method above described.

2.2.5 Human oral bioavailability

Hard gelatin capsules containing CoQ10 either as the free compound (EnergyCOQ10, Herbamed Ltd., Israel) or Emulsome-encapsulated formulation were administered orally to

human volunteers. Plasma samples for CoQ10 analysis were drawn before and 1h post administration.

3. RESULTS AND DISCUSSION

Figure 1 shows the in vitro release patterns of CoQ10 from the Emulsome formulation and the commercial product EnergyCOQ10 (non-encapsulated drug) in simulated gastric fluid.

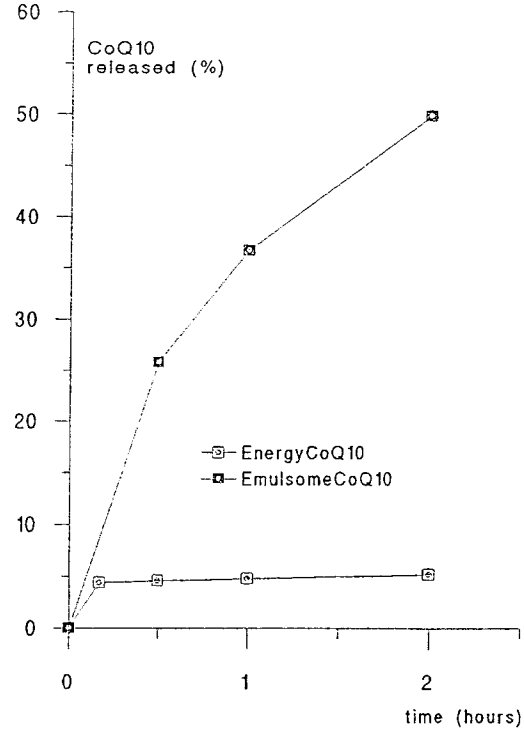

Figure 1 In vitro release of CoQ10 in simulated gastric fluid

The % release of CoQ10 from the marketed product was very low compared to a very significant release (50% after 2 hours) from the Emulsome formulation. Each Energy COQ10 hard gelatin capsule contains 50mg of CoQ10 mixed with rice powder. After capsule disruption in the simulated gastric fluid, big aggregates or clusters of CoQ10 and swelled rice powder were observed which may explain the low CoQ10 dissolution into the release medium. Since particle size is a determinant factor in the rate and extent of drug absorption from gastrointestinal tract, this result indicates low oral bioavailability of CoQ10 from the commercial product compared to the Emulsome formulation.

The results of a human oral bioavailability study involving oral ingestion of a 50mg hard gelatin capsule containing the free CoQ10 product and Emulsome-encapsulated formulation are presented in Figure 2. Enhanced CoQ10 plasma levels 1h post administration were observed with the Emulsome-CoQ10 formulation compared to very low plasma concentration for the EnergyCOQ10 product supporting the in vitro release results.

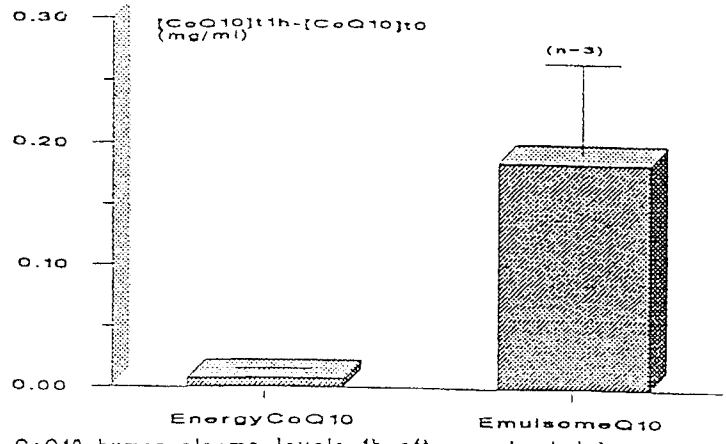

Figure 2 CoQ10 human plasma levels 1h after oral administration of a 50mg CoQ10 hard gelatin capsule

4. CONCLUSIONS

The Emulsome lipoidal vehicle has shown high drug-trapping efficiency and improved oral delivery of Coenzyme Q10. Preliminary results have shown good drug loadings for other lipophilic drugs and biopharmaceuticals. The fact that Emulsome formulations can be freeze-dried to get solid-dosage forms in the form of hard gelatin capsules or tablets makes this lipoidal drug delivery system very atractive for the development and oral delivery of other water-insoluble compounds.

ACKNOWLEDGEMENTS
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