

Bioavailability of two different formulations of coenzyme Q₁₀ in healthy subjects

ML Wahlqvist MD, FRACP, N Wattanapenpaiboon BPharm, PhD, GS Savige PhD, Grad Dip Diet and D Kannar BSc (Hons)

Monash University Department of Medicine, Monash Medical Centre, Clayton, Victoria, Australia

The bioavailability of coenzyme Q₁₀ (ubiquinone) formulated as an emulsion in a soft gelatin capsule (Ensoorb™, NDS Pty Ltd, Sydney, Australia) was compared with a hard gelatin powder-filled capsule. The study design was a randomized cross-over trial with a 3-week wash-out period. The study population comprised 23 apparently healthy adults (12 men and 11 women), aged 20-43 years. Each participant took two 50 mg capsules, and blood samples were taken over a period of 36 h. The plasma concentration of coenzyme Q₁₀ peaked between 3 and 4 h after administration of both preparations. The area under the curve (AUC) of Ensoorb™ was 927% higher than that observed with the powder-filled capsule ($P < 0.0001$), suggesting that this emulsion preparation has a higher bioavailability.

Key words: coenzyme Q₁₀, ubiquinone, bioavailability, emulsion dosage form.

Introduction

Coenzyme Q is a component of the mitochondrial electron transfer chain and in its reduced form can function as an antioxidant.¹ Several studies have shown that coenzyme Q₁₀ (CoQ), which is the most dominant form in humans, exerts a protective effect on *in vitro* LDL oxidation when reduced,²⁻⁴ and therefore the concentration of CoQ in blood may be protective against atherosclerosis. There is a decrease in CoQ tissue content with increasing age^{5,6} which may account, in part, for the age-related increase observed in oxidative damage to protein and DNA.^{7,8} Its concentration in tissues can alter under various pathological conditions¹ and 3-hydroxy-3-methyl-glutaryl-coenzyme A (HMG-CoA) reductase inhibitors appear to reduce CoQ levels.⁹⁻¹¹

The optimal dietary intake of CoQ is unknown. Recently it has been reported that the average CoQ intake of the Danish population was 3-5 mg/d.¹² It is proposed that increasing the intake of dietary sources rich in CoQ (meat and poultry) or the administration of CoQ capsules may be beneficial under certain conditions where lower plasma or tissue levels of CoQ are found, especially in elderly adults⁶ or those on cholesterol lowering medications.⁹⁻¹¹ Orally administered oxidized CoQ can increase the plasma concentration of reduced CoQ, thus providing potential antioxidative protection for the LDL from lipid peroxidation and prevention of free radical damage caused by neutrophils in inflammatory diseases.^{1,2,13}

It is important to obtain data on the bioavailability of dietary supplements for several reasons. From a safety point of view, excessive absorption of certain ingested products may be toxic. Furthermore, if supplements are to be prescribed for therapeutic or prophylactic use, bioavailability must be taken into account in order to evaluate the efficacy of these products.

There is evidence that fat-soluble vitamins are better absorbed from aqueous or emulsified vehicles than from oily

preparations. Certain formulations can affect the absorption of lipid soluble substances. Many commercial vitamin preparations are formulated as compressed tablets, hardshell gelatin capsules or soft gelatin capsules which contain a complex matrix of excipients, fillers and other adjuvants. In the present study the bioavailability of CoQ from two different preparations was compared in order to ascertain if a new preparation could enhance the bioavailability of CoQ over the currently available commercial preparation.

Subjects and methods

Two different gelatin capsules containing 50 mg of CoQ were used in this study. The commercial preparation was crystalline CoQ, with dicalcium phosphate as a filler and magnesium stearate as an excipient, filled in a hard gelatin capsule. The new formulation, Ensoorb™ (NDS Pty Ltd, Sydney, Australia), contained CoQ as a complex micelle in an emulsion encapsulated into a soft gelatin capsule.

The bioavailability study was conducted as a randomized two-period crossover trial. Twenty-three apparently healthy Caucasian volunteers (12 men and 11 women) with a body mass index between 19 and 26 kg/m², aged 20-43 years, were recruited for this study. Subjects with chronic disease, those on medication or vitamin and/or mineral therapy, and smokers were excluded from the study. All subjects were requested to abstain from nutritional supplements for at least 2 weeks prior to the study and from alcohol for 48 h before the study.

Subjects were asked to fast for 10 h prior to the dose administration (i.e. from 22.00 h the night before an 08.00 h

Correspondence address: Professor Mark L Wahlqvist, Department of Medicine Monash Medical Centre, 246 Clayton Road, Clayton, Victoria, Australia. Tel: +61 39550 5525, Fax: +61 39550 5437 E-mail: mark.wahlqvist@med.monash.edu.au

dose administration). Venous blood (about 4 mL) was drawn into an EDTA tube immediately before dosing as the baseline sample (Time 0). A single dose of CoQ (two \times 50 mg capsules) was administered with 200 mL of water at 08.00 h. Blood samples were drawn at 2, 4, 6, 8, 10, 24 and 36 h after administration. All blood samples were centrifuged within 30 min after collection and the plasma frozen at -70°C until analysed for CoQ. The same meals were served to all subjects in the first 10 h of the study. Breakfast was withheld until 2 h after dosing. Lunch and afternoon tea was eaten after dosing at 5 and 9 h, respectively. This protocol was repeated with the other preparation after a 3-week wash-out period.

Informed consent was obtained from each subject and the study protocol was approved by the Monash University Standing Ethics Committee for Human Research.

High pressure liquid chromatography analysis of coenzyme Q

Coenzyme Q is present in the blood in both oxidized and reduced form. Plasma CoQ concentrations were determined by modifying the high pressure liquid chromatography (HPLC) method of Edlund.¹⁴ Before the determination of CoQ, the samples were oxidized with ferric chloride in hydrochloric acid. The result of the determination was therefore the total amount of CoQ.

In brief, plasma was mixed with ferric chloride solution (0.1%) in the presence of diluted hydrochloric acid to allow oxidation of reduced CoQ. The mixture was then shaken with *n*-propanol, let stand at ambient temperature for 10 min, and then centrifuged at 2100 *g* for 5 min. One hundred microlitres of the supernatant were injected to the HPLC system. Reverse-phase chromatography was carried out on an Allsphere ODS-2, 5 μm analytical column in conjunction with a guard column. Mobile phase was *n*-propanol: methanol (75:25, by volume) containing 33 mmol/L perchloric acid and 57 mmol/L sodium perchlorate, run at a flow rate of 1.2 mL/min. Detection was performed by UV absorption at 275 nm. Quantification was done by comparing results with the standard curve by linear-regression analysis.

Data analysis

The area under the curve from time 0 to 36 h ($\text{AUC}_{[0-36]}$) after administration was calculated as the integral of the trapezium formed by the area between the time axis and the CoQ concentration using the linear trapezoid method, and did not include the basic AUC. The basic AUC was calculated as the integral from time 0 to 36 h of the trapezium formed by the area between the time axis and the CoQ concentration at baseline.

The Statistical Analysis System (SAS Institute Inc, Cary, NC, USA) software package was used for data analysis. Descriptive statistics including means and standard deviations were calculated for each continuous variable. A paired *t*-test was used to evaluate differences between the two study formulations in plasma CoQ concentration and AUC. Gender differences in these variables were determined by a non-parametric Wilcoxon rank sum test. The significance level was set at 5%.

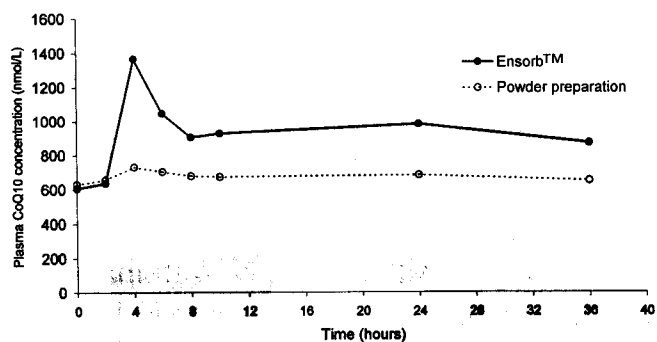


Figure 1. Mean plasma coenzyme Q₁₀ concentration as a function of time following the oral administration of either two capsules of Ensorb™ (●) or the powder preparation (○), each containing 50 mg coenzyme Q₁₀.

Results

Figure 1 shows the mean plasma concentration–time profile for both preparations. The peak plasma CoQ concentration observed at 4 h after dosing was more pronounced with Ensorb™ where an average increase of 130% over the baseline concentration was observed compared with a 20.3% rise with the powder-filled capsule (Table 1).

Total absorption of CoQ as determined from the AUC was also greater from the Ensorb™ formulation (Table 1), representing 927% higher than that observed with the powder-filled capsule. Some subjects, however, appeared to be better able to absorb CoQ than others and a large inter-individual variation in the AUC was evident with both formulations.

No gender differences were observed in respect of plasma CoQ concentration at baseline or AUC. It was, however, observed that the peak plasma CoQ concentration observed at 4 h after Ensorb™ administration, was higher in men compared with women ($P < 0.05$).

Table 1. Comparison of plasma coenzyme Q₁₀ concentration and the area under the curve following oral administration of two different formulations

	Total (<i>n</i> = 23)	Men (<i>n</i> = 12)	Women (<i>n</i> = 11)
Emulsion in soft gelatin capsules (Ensobr™)			
CoQ concentration at Time 0 (nmol/L) ^a	605 ± 121	614 ± 132	596 ± 113
CoQ concentration at 4 h (nmol/L)	1366 ± 361	1534 ± 384	1183 ± 231*
% Increase in CoQ concentration	130 ± 61	154 ± 62	104 ± 49
AUC _[0-36] (nmol/L*h)	12064 ± 3961	13203 ± 3893	10821 ± 3819
Powder blend in hard gelatin capsules			
CoQ concentration at Time 0 (nmol/L)	630 ± 165	672 ± 175	584 ± 147
CoQ concentration at 4 h (nmol/L)	730 ± 164	736 ± 156	724 ± 179
% Increase in CoQ concentration	20 ± 35	12 ± 14	30 ± 47
AUC _[0-36] (nmol/L*h)	1586 ± 3345	415 ± 2318	2864 ± 3907

^a To convert to mg/mL, multiply values with 863.4×10^{-6} . AUC_[0-36]: area under the curve from time 0 to 36 h. * Significantly different from men, $P < 0.05$ (Wilcoxon rank-sum test).

Discussion

The intestinal absorption of drugs can be markedly influenced by the oral dosage form as well as the formulation factors.¹⁵ Compounds formulated in soft gelatin capsules representing liquid fills tend to be better absorbed than hard gelatin capsules, which encapsulate a dry powder blend.^{16,17}

It is known that surfactants, as emulsifying agents, at concentrations below their critical micelle concentration can increase the solubility and dissolution rate of drugs from dosage forms; thus the drug becomes available for absorption.^{18,19} In addition, surfactants can penetrate and disrupt the normal structure of biological membranes resulting in increased membrane permeability. This may explain the results of the present study where the absorption of CoQ formulated as an emulsion (Ensorb™) was significantly enhanced over a powder-filled hard gelatin capsule. However, surfactants above their critical micelle concentrations solubilize and retain the drug, thereby causing an overall reduction in the amount of drug released and absorbed.¹⁸

Gender differences in peak coenzyme Q concentration

Gender differences in bioavailability of CoQ have been previously reported by Weis *et al.*²⁰ The difference in the AUC

values with respect to gender was observed in their study, while the difference in the peak concentration was observed in the present study. Nevertheless, the results of the present study still support their suggestion that men could have better absorption and/or a lower clearance than women.

Two-peak pattern

Plasma CoQ concentrations peaking at 4 and 24 h demonstrated a 'two-peak pattern' in the concentration-time profile which has been previously reported.²⁰ Coenzyme Q is administered as the oxidized form and, while circulating in the blood, it undergoes reduction, resulting in a less lipophilic form and impaired ability of CoQ to permeate cell membranes. This process could explain the two-peak pattern.²⁰

In conclusion, the present study demonstrates that Ensorb™ has a higher bioavailability than the commercial formulation used in this study. The presence of surfactants in the Ensorb™ formulation would contribute to the enhanced solubilization and release of CoQ.

Acknowledgements. This study was fully funded by the NDS Pty Ltd, Willoughby, NSW, Australia. Thanks go to all participants for their cooperation.

Bioavailability of two different formulations of coenzyme Q₁₀ in healthy subjects

ML Wahlqvist, N Wattanapenpaiboon, GS Savige, D Kannar

Asia Pacific Journal of Clinical Nutrition (1998) Volume 7, Number 1: 37-40

健康成人對兩種不同輔酶 Q₁₀製劑的生物利用率

摘要

作者比較了軟膠囊 (Ensorb™) 乳劑輔酶 Q₁₀ (泛醌) 和硬膠囊粉劑輔酶 Q₁₀ (泛醌) 的生物利用率, 用隨機交叉試驗, 經過 3 周清洗期後, 再重複試驗一次, 試驗對象包括 23 位健康成人 (男 12 人, 女 11 人), 年齡在 20—43 歲, 試驗對象服用 2 個 50mg 輔酶 Q 膠囊, 服膠囊後第 2、4、6、8、10、24 和 36 小時分別抽取血液樣本測定輔酶 Q₁₀, 結果發現, 服膠囊後血漿輔酶 Q₁₀ 濃度在 3—4 小時出現高峰, 乳劑軟膠囊的曲線面積較粉劑硬膠囊高 927% (P<0.0001)。因此作者指出: 乳製劑的生物利用率較粉製劑為高。

References

1. Ernster L, Dallner G. Biochemical, physiological and medical aspects of ubiquinone function. *Biochim Biophys Acta* 1995; 1271: 195-204.
2. Mohr D, Bowry VW, Stocker R. Dietary supplementation with coenzyme Q₁₀ results in increased levels of ubiquinol-10 within circulating lipoproteins and increased resistance of human low-density lipoprotein to the initiation of lipid peroxidation. *Biochim Biophys Acta* 1992; 1126: 247-254.
3. Kontush A, Hübner C, Finckh B, Kohlschütter A, Beisiegel U. Low density lipoprotein oxidizability by copper correlates to its initial ubiquinol-10 and polyunsaturated fatty acid content. *FEBS Lett* 1994; 341: 69-73.
4. Kontush A, Hübner C, Finckh B, Kohlschütter A, Beisiegel U. Antioxidative activity of ubiquinol-10 at physiologic concentrations in human low density lipoprotein. *Biochim Biophys Acta* 1995; 1258: 177-187.
5. Kalén A, Appelkvist E, Dallner G. Age-related changes in the lipid compositions of rat and human tissues. *Lipids* 1989; 24: 579-584.
6. Söderberg M, Edlund C, Kristensson K, Dallner G. Lipid composition of different regions of the human brain during aging. *J Neurochem* 1990; 54: 415-423.

7. Stadtman ER, Oliver CN. Metal-catalyzed oxidation of proteins. Physiological consequences. *J Biol Chem* 1991; 266: 2005-2008.
8. Linnane AW, Marzuki S, Ozawa T, Tanaka M. Mitochondrial DNA mutations as an important contributor to ageing and degenerative disease. *Lancet* 1988; 25: 642-645.
9. Watts GF, Caselluccio C, Rice-Evans C, Taub NA, Baum H, Quinn PJ. Plasma coenzyme Q (ubiquinone) concentrations in patients treated with simvastatin. *J Clin Pathol* 1993; 46: 1055-1057.
10. Ghirlanda G, Oradei A, Manto A *et al*. Evidence of plasma CoQ₁₀-lowering effect of HMG-CoA reductase inhibitors: a double blind, placebo-controlled study. *J Clin Pharmacol* 1993; 33: 226-229.
11. Laaksonen R, Ojala J, Tikkanen MJ, Himberg J. Serum ubiquinone concentrations after short- and long-term treatment with HMG-CoA reductase inhibitors. *Eur J Clin Pharm* 1994; 46: 313-317.
12. Weber C, Bysted A, Holmer G. The coenzyme Q₁₀ content of the average Danish diet. *Internat J Vit Nutr Res* 1997; 67: 123-129.
13. Weber C, Jakobsen TS, Mortensen SA, Paulsen G, Holmer G. Antioxidative effect of dietary coenzyme Q₁₀ in human blood plasma. *Internat J Vit Nutr Res* 1994; 64: 311-315.
14. Edlund PO. Determination of CoQ, α -tocopherol and cholesterol in biological samples by coupled-column liquid chromatography with coulometric and ultra-violet detection. *J Chromatol* 1988; 425: 87-97.
15. Thakker KM, Sitren HS, Gregory JF III, Schmidt GL, Baumgartner TG. Dosage form and formulation effects on the bioavailability of vitamin E, riboflavin, and vitamin B-6 from multivitamin preparations. *Am J Clin Nutr* 1987; 45: 1472-1479.
16. Horwitt MK, Elliot WH, Kanjanagulpan P, Fitch CD. Serum concentration of α -tocopherol after ingestion of various vitamin E preparations. *Am J Clin Nutr* 1984; 40: 240-245.
17. Bateman NE, Uccellini DA. Effect of formulation on the bioavailability of retinol, d- α -tocopherol and riboflavin. *J Pharm Pharmacol* 1984; 36: 461-464.
18. Gibaldi M. *Biopharmaceutics and clinical pharmacokinetics*. 3rd edn. Lea & Febiger, Philadelphia, PA, USA, 1984.
19. Bateman NE, Uccellini DA. Effect of formulation on the bioavailability of retinol, D- α -tocopherol and riboflavine. *J Pharm Pharmacol* 1984; 36: 461-464.
20. Weis M, Mortensen SA, Rassing MR, Møller-Sonnergaard J, Poulsen G, Rasmussen SN. Bioavailability of four oral coenzyme Q₁₀ formulations in healthy volunteers. *Molec Aspects Med* 1994; 15 (Suppl.): S273-S280.