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Original Contribution

PREECLAMPSIA IS ASSOCIATED WITH A DECREASE IN PLASMA COENZYME Q10 LEVELS

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Abstract—Preeclampsia is a common (~7% of all pregnancies) disorder of human pregnancy in which the normal hemodynamic response to pregnancy is compromised. Despite many years of intensive research, the pathogenesis of preeclampsia is still not fully understood. The objective of the present study was to investigate the concentration of coenzyme Q10 in normal pregnancy and preeclampsia. Pregnant women ($n = 18$), women with preeclampsia ($n = 12$), and nonpregnant normotensive women ($n = 22$) were included. Plasma levels of coenzyme Q10 were measured by high-performance liquid chromatography. Plasma coenzyme Q10 levels were significantly higher in normal pregnant women (mean = 1.08, SEM = 0.08 umol/l; $p < .005$) in comparison to nonpregnant women (mean = 0.86, SEM = 0.16 umol/l) and women with preeclampsia (mean = 0.7, SEM = 0.03 umol/l; $p < .0001$). These results demonstrated that during preeclampsia there is a significant decrease in plasma levels of coenzyme Q10 compared to normal pregnant women, and compared to those who are not pregnant. © 2003 Elsevier Inc.

Keywords—Coenzyme Q10, Pregnancy, Preeclampsia, Oxidative stress, Free radicals

INTRODUCTION

Preeclampsia is a common (~7% of all pregnancies) disorder of human pregnancy in which the normal hemodynamic response to pregnancy is compromised. It remains a leading cause of maternal morbidity and mortality, and is associated with a significant increase in perinatal mortality [1]. Despite many years of intensive research, the pathogenesis of preeclampsia is still not fully understood. In pregnancies complicated with preeclampsia, there is a marked decrease in serum and tissue antioxidant protection, i.e., superoxide dismutase, catalase, and glutathione peroxidase, compared to uncomplicated pregnancies [2,3], suggesting that oxidative stress might also be implicated in the pathophysiology of preeclampsia.

Coenzyme Q (CoQ) or ubiquinone is well defined as a crucial component of the oxidative phosphorylation process in the mitochondria, which converts the energy in carbohydrates and fatty acids into adenosine triphosphate (ATP) to drive cellular machinery and synthesis [4]. It also has antioxidant and membrane-stabilizing functions [5]. In particular, coenzyme Q10, which is a natural ubiquinone found in animal mitochondria [6], plays a well-known role in the oxidative metabolism of muscle cells during physiological [7] and pathological conditions (cardiovascular, cancer, muscular, and mitochondrial disease) [8–10].

Also, it is well known that CoQ is an important antioxidant in the inner membrane, where it scavenges radicals directly [11] and regenerates α-tocopherol from the tocopheroxyl radical [12]. CoQ is not only present in the inner mitochondrial membrane, but in low-density lipoprotein (LDL), plasma membranes, and all intracellular membranes, where it is found mainly in the reduced state [13].

Measurement of plasma coenzyme Q10 has been claimed to be of clinical significance [14,15], and the range for plasma coenzyme Q10 concentrations in humans is typically 0.5–2 umol/l [16,17]. It is already well established that plasma coenzyme Q10 concentrations are highly dependent on serum lipoproteins (cholesterol

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Table 1. Characteristics of the Women Included in the Study

	Nonpregnant (n = 22)		Normal pregnant (n = 18)		Preeclampsia (n = 12)		
	Mean	SD	Mean	SD	Mean	SD	p
Age (y)	19.5	0.32	19.7	0.7	21.7	0.9	
Gestational age (weeks)	–		39.6	0.07	37.6	0.2	<.01
Systolic BP (mmHg)	99.1	1.8	109.4	2.3	150.0	4.4	<.0001
Diastolic BP (mmHg)	65.4	1.7	70.5	2.1	103.3	3.8	<.0001

levels), which are carriers of coenzyme Q10 in the circulation [18]. Interestingly, we have found only few papers analyzing CoQ during normal pregnancy [19,20], whereas no data about CoQ and preeclampsia is available.

Thus, the objective of the present study was to investigate the plasma levels of coenzyme Q10 both in normal pregnancy and in those complicated with preeclampsia.

PARTICIPANTS AND METHODS

The study was approved by the Ethics Committee of the Biomedical Center and was conducted at the Hospital Gineco-Obstetrico Isidro Ayora (HGOIA), Quito, Ecuador. Pregnant women (n = 18) were selected according to the following criteria: nulliparity, age < 25 years, residing in Quito (2800 m altitude), blood pressure of <120/80 mmHg, and no known medical disorders. Women with preeclampsia (n = 12) had blood pressures over 140/90 mmHg on at least two occasions 6 h apart, and proteinuria greater than ++ as assessed by dipstick on the first urine of the morning (>300 mg/dl), on two occasions 4–24 h apart. Blood pressure was measured as described previously [21]. Nonpregnant normotensive women (n = 22) who attended the HGOIA for family planning were included as controls. None of these women were pregnant before. Blood samples in preeclamptic women were taken when the diagnosis was confirmed (mean = 37.6, SEM = 0.2 weeks), whereas in normal pregnant women samples were taken at admission to the hospital, at least 6 h before delivery (mean = 39.6, SEM = 0.07 weeks). A blood sample was taken from each woman at the antecubital venous puncture site, and immediately transferred into a heparinized polypropylene vial. Samples was centrifuged at 4°C for 10 min, plasma fraction transferred into another vial and stored in 500 ul aliquots at −40°C until assayed. Plasma levels of coenzyme Q10 was measured using a high performance liquid chromatography (HPLC) system (Perkin-Elmer, Shelton, CT, USA) equipped with a Lichrosorb RP18 (10 um, 125 × 4 mm; Phenomenex, Torrance, CA, USA) column and with a guard column (Merck, Darmstadt, Germany). The mobile phase was methanol/ethanol (30:70 v/v) previously filtered. Flow rate was 1 ml/min and the UV detector was set up at 275 nm. For analysis, samples were thawed on ice and 0.2 ml of plasma was mixed with 50 ul ethanolic BHT (2,6-di-tert-butyl-p-cresol; Sigma Aldrich, St. Louis, MO, USA) solution (10 mg/ml), 0.2 ml of 0.1 M aqueous sodium dodecyl sulfate (Sigma-Aldrich) and 1.6 ml of a solution of CoQ9 (Fluka Biochemica, Buchs, Switzerland) in ethanol as internal standard. The mixture was vortexed for 30 s, 2 ml of hexane (Merck, Darmstadt, Germany) was added, and the tightly screwed test tube was vigorously vortexed for 2 min. It was then centrifuged for 5 min at 1000 × g to separate the layers. One milliliter of the hexane layer was transferred to a small vial and dried under nitrogen. The residue was redissolved in methanol/(ethanol/isopropanol 95/5, v/v) (1:1 v/v). Samples were analyzed immediately and kept on ice and covered with aluminum foil to prevent photodegradation of ubiquinones. Coenzyme Q10 was a kind gift of Eisai Pharma-Chem Europe Ltd. (London, UK) and standards were prepared in absolute ethanol after an initial dilution in chloroform (Merck). Samples were measured in duplicates and the mean value was used for the statistical analysis. Detection limit of the assay was 0.1 umol/l and the average recovery of the internal standard was 85%. The intra-assay error coefficient was 4.34% whereas the inter-assay error coefficient was 15.9%. Plasma cholesterol was determined in duplicates using a spectrophotometer (Eppendorf, Germany) and a commercial available kit (Human GmbH, Wiesbaden, Germany). Finally, an aliquot of whole blood was taken to determine hematocrit, hemoglobin, and leukocyte count.

Kruskal–Wallis nonparametric analysis of variance followed by the Mann–Whitney U-test was used to compare the women with preeclampsia and the controls, the women with normal pregnancy and the controls, and the women with normal pregnancy and those with preeclampsia. In performing the Mann–Whitney U-test the nominal level of statistical significance was set at 0.05.

RESULTS

The characteristics of women included in the study are presented in Table 1. Blood tests revealed that both hematocrit and hemoglobin levels were significantly low

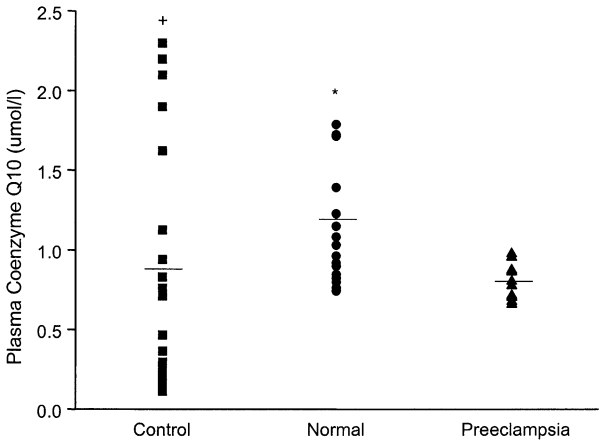

Fig. 1. Plasma levels of coenzyme Q10 (umol/l) measured by high-performance liquid chromatography in nonpregnant (n = 22), normal pregnant (n = 18) and preeclamptic (n = 12) women. Kruskal–Wallis test showed a p value <.02. *p < .006 preeclampsia compared with normal pregnant women and ^{+}p < .02 preeclampsia compared with nonpregnant women.

(p < .001) in normal pregnant women (mean = 40.8, SD = 0.4% and mean = 12.9, SD = 0.2 g/dl, respectively) compared with nonpregnant women (mean = 46.0, SD = 0.6% and mean = 14.8, SD = 0.2 g/dl, respectively) and even lower (p < .001) in those with preeclampsia (mean = 38.2, SD = 1.6% and mean = 11.9, SD = 0.5 g/dl, respectively). This unusual finding might be due to a nutritional deficiency, as reported previously [21,22].

Leukocyte count was significantly higher during pregnancy, both in normal pregnancy (mean = 9.7, SD = 0.3 × 10^6/l) and in those complicated with preeclampsia (mean = 8.1, SD = 0.3 × 10^6/l), compared to nonpregnant women (mean = 6.9, SD = 0.2 × 10^6/l; p < .001).

Women with preeclampsia showed significantly higher plasma levels of cholesterol compared to those with normal pregnancy (mean = 282, SD = 34 vs. mean = 225, SD = 47 mg/dl; p = .002) or nonpregnant women (mean = 166, SD = 31 mg/dl; p < .0001).

Plasma coenzyme Q10 levels were significantly different between all groups (p < .02). The values were higher in normal pregnant women (mean = 1.08, SEM = 0.08 umol/l; p < .02) in comparison to nonpregnant women (mean = 0.86, SEM = 0.16 umol/l) and women with preeclampsia (mean = 0.7, SEM = 0.03 umol/l; p < .006; Fig. 1).

DISCUSSION

This study conducted in an Andean population is in good agreement with previously reported values of plasma coenzyme Q10 in other populations (i.e., Finland and Japan) [17,23]. Even though the women described in this report have well-known nutritional deficiencies, we have found that pregnant women showed a significant increase in plasma levels of coenzyme Q10. This data closely resembled the few reports about this antioxidant during human gestation [19,20].

However, we were not able to find any information about coenzyme Q10 in pregnancies complicated with preeclampsia, therefore, our data constitutes the first report into the literature. We have found that during preeclampsia there is a significant decrease in plasma levels of coenzyme Q10 compared to normal pregnant women, and compared to those nonpregnant. However, this decrease was completely independent of the plasma cholesterol that was significantly higher in women with preeclampsia compared to normal pregnant and nonpregnant women, as reported by other authors [24–26]. Interestingly, the increase of coenzyme Q10 during normal pregnancy most likely depends on the increase in LDL, which are the main carriers of coenzyme Q10 in plasma.

This abnormal response that might be occurring during preeclampsia is congruent with the well-accepted suggestion that oxidative stress is implicated in the pathophysiology of preeclampsia. In pregnancies complicated with preeclampsia, there is a marked decrease in sera antioxidant protection compared with uncomplicated pregnancies [2,3]. On the other hand, it is also possible that plasma levels of coenzyme Q10, like plasma levels of reduced gluthatione [27], should be considered as an indicator of intracellular pool of coenzyme Q10. As a result, for us it is reasonable to hypothesize that energy production is affected. Therefore, abnormal placentation, another classical finding in preeclampsia [28], should be an obvious consequence.

Furthermore, assuming that in the absence of adequate levels of coenzyme Q10, the electron transport chain does not work properly, production of superoxide radical will be increased [7]. This immediately brings to mind the possibility that coenzyme Q10 is upregulating the nitric oxide (NO) vasodilatory mechanism. However, it is difficult to see how coenzyme Q10 could influence NO synthesis, because the constitutive NO synthase is a soluble enzyme that would have little direct contact with membrane-sequestered coenzyme Q10 [29]. Interestingly, we have found that free NO can react directly with coenzyme Q in a reversible manner, resulting in inhibition of mitochondrial complex I activity [30].

We recognize that weaknesses of our study were its modest-sized and cross-sectional design. But, our study also had strengths; we used a rigorous case definition for preeclampsia and well-established methods for the measurement of coenzyme Q10. It was undertaken in women with established preeclampsia and it is not possible to determine whether the decrease in plasma levels of coenzyme Q10 was a cause or a consequence of the disease. In conclusion, the observed link between CoQ10

and preeclampsia might bear deep physiopathological significance and deserves to be further elucidated.

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