INTRODUCTION

The Anthracycline antibiotic Adriamycin (Doxorubicin) is widely used in clinical medicine for cancer chemotherapy. Unfortunately, the clinical use of Adriamycin (ADR) is accompanied by side effects of which cardiotoxicity is the most serious. The cumulative dose-dependent cardiomyopathy severely limits the total dose of ADR that can be administered in the treatment of neoplastic diseases(1). The prevention of ADR cardiotoxicity without interference with the drug's antitumor activity would be of considerable advantage in achieving additional therapeutic benefit from this agent.

The identification of several biochemical activities of ADR in vitro has suggested different mechanisms for ADR cytotoxicity. These include direct interaction with nucleic acid, inhibition of coenzyme Q-dependent enzymes, direct cell surface interaction. However, the current leading hypothesis for the mechanism of ADR cardiotoxicity is that ADR produces reactive free radicals (2) that damage myocardial tissue by non-specific oxidation of membrane and cytosol molecules, ultimately leading to cell death.

In an effort to avoid the development of chronic cardiotoxicity of ADR, several potential cardioprotective compounds including selenium (Se), vitamin E, glutathione and other sulphydryl donors have been tested. There have been few studies in which the specific effect of Se deficiency on the toxicity of ADR has been investigated. DOROSHOW 1980 (3) found that Se deficiency increased the sensivity of mice to acute ADR toxicity. FACCHINETTI 1983 (4) showed that Se-supplemented rats had better resistance to chronic ADR treatment than Se-deficient rats.

In addition, mammalian cells are protected by several enzymatic systems e.g. superoxide dismutase, catalase and glutathione peroxidase. This latter enzyme is present in two different forms: selenium-independent glutathione peroxidase (GPx) and a selenium-dependent glutathione peroxidase (Se-GPx). The latter, which contains selenium, has a potential dual function by reducing hydrogen peroxide and already formed lipid hydroperoxides. Thus, exogenously administered selenium eliminates preferential oxygen centered free radicals thereby protecting the tissue from oxidative damage. This paper reports on the influence of selenium intake on endogenous antioxidant protective systems and therefore on cardiac functions in chronically adriamycin-treated rats.
MATERIALS AND METHODS

Rats, diets and drug

Experiments were performed on male wistar rats of initial weight 180-200 g, supplied by IFFA CREDO, France. Sixty rats were allotted to six groups of 10 rats each, were housed in stainless steel cages and received granulated food and deionized distilled water ad libitum. Three groups were fed a Se-deficient basal diet obtained from UA R society (Villemoisson, France) with the following composition:

- Beer yeast: 30%
- Sucrose: 55.7%
- Corn oil: 8%
- Salt mixture: 5%
- Vitamin mixture: 1%
- DL Methionine: 0.3%
- Selenium: 0.058 ppm

The analysis of the basal diet showed that it contained 60 ug Se/kg diet. The other three groups were fed the same basal diet supplemented with 1000 ug Se/kg diet as Na2SeO3. Rats were weighed weekly. Two weeks after beginning Se deficiency, two groups received intraperitoneal injections of ADR for 2,5 weeks (two injections of 4mg of ADR /kg body weight per week) for total doses of 20 mg ADR/kg body weight. ADR (Adriblastine), purchased from Roger Bellon, was reconstituted in deionized distilled water at a concentration of 1 mg/ml immediately before injection. Control rats (non drug treated) received an equivalent volume of sucrose solution at 10 mg/ml.

Two weeks after the last injection of ADR, rats were killed and hearts were quickly removed and mounted on the perfusion apparatus as described by LANGENDORFF 1895 (5). Hearts were perfused for 5 min with a constant perfusion pressure of 60 mm Hg through the coronary vessels, by a Krebs-Henselei bicarbonate buffer. Ischaemia was performed at 37°C by a total stop of perfusion for 15 min. The reperfusion was allowed during 2 min after which hearts were immediately immersed in liquid nitrogen. During perfusion and reperfusion phases, coronary flow and cardiac frequency were recorded.

Biochemical analyses

After 18 hours of fasting, the rats were killed and tissues were removed and immersed in liquid nitrogen. Blood was collected from the chest cavity after heart had been excised. Se deficiency was verified by plasma Se determination (electrothermal AAS) and by measurement of Se-GPx in plasma, red blood cells (RBC) and in liver and heart homogenates by a modification of GUNZLER method 1994 (6). Plasma and tissue samples were analyzed for lipid peroxidation products: malondialdehyde (MDA) by YAGI method 1990 (7), dienes conjugated (D.C) as described by DORMANDY 1981 (8) and lipid hydroperoxides (LHP) by the enzymatic method of TAPEL 1979 (9). Other antioxidant enzymatic and non enzymatic systems were also determined: Superoxide dismutase (SOD) was evaluated by MARKLUND method (10) glutathione reductase (GRx), glutathione transferase (GTx), catalase by the enzymatic method. The contents of reduced glutathione (GSH) and oxidized glutathione (GSSG) in the total blood and in tissues were determined as described by TIEZE 1969 (11). Alpha-tocopherol and xanthine oxidase activity were also evaluated.

STATISTICS

The results are expressed as means + standard deviation (S.D). Data were analysed statistically by using Student’s t-test, and differences of P < 0.05 were considered statistically significant.