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Teratogenic effect of cisplatin in rats and the protective role of sodium selenate

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ABSTRACT

Eighty pregnant Sprague-Dawley rats were used in this study. They were allotted to four equal groups. The first group served as a control without any treatment while the other groups were given cisplatin, sodium selenate, and cisplatin + sodium selenate, respectively. Cisplatin was injected intraperitoneally in a dose of 5 mg/kgb wt. on the 12th day of gestation while sodium selenate was administered orally in a dose of 0.5 mg/kgb wt throughout gestation. Animals were sacrificed on the 20th day of gestation for fetal examination. Cisplatin produced significant elevation in the percentages of late resorption sites and dead foetuses compared with the control group. The mean foetal and placental weights were significantly reduced. Dwarf foetuses and subcutaneous (s/c) haemorrhage were also recorded in cisplatin-treated group. Visceral abnormalities were revealed in the form of dilated nares, anophthalmia and/or microphthalmia, dilated brain ventricles, hypertrophy of the heart, hypoplasia of the lung, hepatomegaly and dilated renal pelvis. Skeletal examination showed wide open fontanel, incomplete ossification of parietal and interparietal bones, incomplete ossification of sternum, reduction in the number or even complete absence of phalanges, sacral and/or caudal vertebrae. Histopathological examination of placentas in cisplatin-treated group revealed severe pathological alterations. Administration of sodium selenate significantly alleviated the afore-mentioned adverse effects of cisplatin on the fetuses and their placentas so we conclude that sodium selenate as an antioxidant has an effective protective role in cisplatin teratogenic effects.

1. Introduction

Many anticancer agents have been shown to be mutagenic, teratogenic and carcinogenic in experimental systems and second malignancies are known to be associated with several specific therapeutic treatments. Anticancer agents thus represent a class of occupational carcinogens, the handling of which should involve no unnecessary exposure. Two recent case-referent studies among hospital personnel have pointed to slightly increased risks of disorders in pregnancy outcome; one of the studies has shown an excess of spontaneous abortions and other malformations in children of females with a history of work with anticancer agents (Sorsa et al., 1985).

Cisplain [*cis*-dichlorodiammineplatinum(II)], the first generation anticancer platinum complex, is one of the most effective anticancer agents currently available for the treatment of testicular, ovarian, and bladder carcinomas but its clinical usefulness has frequently been limited by undesirable side effects such as nephrotoxicity, gastrointestinal toxicity, ototoxicity, and neurotoxicity (Bruno et al., 2003; Sindhu and Kuttan, 2013). Cisplatin has a profound deleterious effect on the reproductive system and on the fertility potential of the treated male and female rats (Kinkead et al., 1992; Altuner et al., 2013).

In a healthy body, reactive oxygen species (ROS) and antioxidants remain in balance. When the balance is disrupted towards an overabundance of ROS, oxidative stress (OS) occurs. OS influences the entire reproductive lifespan of a woman and even thereafter (i.e. menopause). OS results from an imbalance between prooxidants (free radical species) and the body's scavenging ability (antioxidants). ROS are a double-edged sword; they serve as key signal molecules in physiological processes but also have a role in

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pathological processes involving the female reproductive tract. ROS affect multiple physiological processes from oocyte maturation to fertilization, embryo development and pregnancy. It has been suggested that OS modulates the age-related decline in fertility. It plays a role during pregnancy and normal parturition and in initiation of preterm labor. Ovulation-induced oxidative base damage and damage to DNA of the ovarian epithelium can be prevented by antioxidants (Agarwal et al., 2005).

Selenium is an essential element because it forms the prosthetic group of some critical selenocysteine-containing enzymes, such as glutathione peroxidase, iodothyronine 5′-deiodinase, and thioredoxin reductase. The mechanism of selenite protection against cisplatin-induced nephrotoxicity is correlated to higher levels of selenium in the kidney (about eight times) and with higher levels of glutathione in the kidney, both compared to tumors. Selenite is metabolized into selenols, specifically into methylselenol and glutathionyl selenol; this bioactivation of selenite into selenols is a glutathione-dependent process. High performance liquid chromatography (HPLC) with on-line radioactivity detection radioactive isotope of 195 mPt showed that methylselenol is capable of forming a complex with cisplatin in vitro. It is proposed that the formation of a cisplatin—selenol complex also takes place in vivo, specially in the kidney, thereby preventing cisplatin exerting its nephrotoxic activity (Yeh et al., 2008).

Protective effect of sodium selenite (Se) on the nephrotoxicity of cis-diamminedichloroplatinum (CDDP) was studied in mice by Araya (1990) and Araya et al. (1990). They concluded that co-administration of Se inhibited the increase of blood urea nitrogen (BUN) and urinary *N*-acetylglucosaminidase (NAG) and depressed the degeneration of proximal tubule cells. The mechanism of its protective effect is related to the glutathione peroxidase enzyme. Vermeulen et al. (1993) concluded that sodium selenite protects rodents against cisplatin-induced nephrotoxicity without influencing the systemic availability of cisplatin and total platinum

Cancer is a very dangerous disease which may be detected during pregnancy and Cisplatin, as an antineoplastic drug, may be urgently used in its treatment resulting in serious side effects on both dams and their embryos. Therefore, our study aims at studying the embryotoxic and teratogenic effects of cisplatin and trials to alleviate these toxic effects using sodium selenate as an antioxidant.

2. Materials and methods

2.1. Cisplatin

It is an antineoplastic cytostatic metallic drug. It is a colorless liquid solution produced by MYLAN, pharmaceuticals Co. (Morgantown, West Virginia, USA) in the form of 50 mg cisplatin/50 ml aqueous solution.

2.2. Sodium selenate

It is the sodium salt of selenic acid, used as a mineral supplement and produced by Sigma-Aldrich chemical Co. (St. Louis, MO, USA).

2.3. Animals

Eighty pregnant Sprague-Dawley rats were used in this study. They were allotted to four equal groups. The first group served as a control without any treatment while the other groups were given cisplatin, sodium selenate, and cisplatin + sodium selenate, respectively. Cisplatin was injected intraperitoneally in a dose of 5 mg/kgb wt. on the 12th day of gestation (Köpf-Maier et al., 1985) while sodium selenate was administered orally by gavage in a dose of 0.5 mg/kgb wt. throughout gestation (Ozdil et al., 2004). Pregnant dams were sacrificed on the 20th day of gestation for foetal examination according to the method adopted by Manson and Kang (1994). The fetuses obtained from control and treated groups were examined for external, visceral and skeletal abnormalities. The placentas were subjected to histopathological examination according to Bancroft and Stevens (1996).

2.3.1. (a) External examination

All treated and control female groups were killed under anesthetic chloroform just prior to the calculated date of delivery (at the 20th day of gestation). After that, an incision was made in the abdominal wall to expose the abdominal viscera. The gravid uterus of each dam was exteriorized then the numbers of uterine implants, early and late resorptions, live and dead fetuses were counted.

The isolated uteri were cut open, exposing the lining of the uterus and amniotic sacs. The amniotic sacs were ruptured at a time to exteriorize fetuses and placentas.

Table 1
Morphological examination of rat fetuses obtained from control and treated dams.

Group	Parameter											
	No. of pregnant dams	No. of uterine implants	Late resorption		Dead fetuses		Live fetuses		Mean fetal weights by g	Mean placental weights by g	External morphological abnormalities	
											Dwarfism	S/c edema and Hemorrhage
			No.	%	No.	%	No.	%			No. %	No. %
Control	20	200	0	0	1	0.5	199	99.5	4.09 ± 0.03	0.59 ± 0.003	0 0	0 0
Cisplatin	20	175	30	17.14[a]	45	25.71[a]	100	57.14[a]	3.09[a] ± 0.04	0.48[a] ± 0.003	25 14.28[a]	29 16.57[a]
Sodium selenate	20	196	0	0	1	0.5	195	99.48	4.09 ± 0.19	0.59 ± 0.001	0 0	0 0
Cisplatin + Sodium selenate	20	189	14	7.41[a,b]	15	7.94[a,b]	160	84.65[a,b]	3.92[b] ± 0.041	0.56[b] ± 0.001	14 7.40[a,b]	15 7.93[a,b]

[a] Significant difference between control and treated groups at $p \leq 0.05$.
[b] Significant difference between cisplatin and sodium selenate group at $p \leq 0.05$.