The Role of Antioxidant Activity in the Prevention and Treatment of Infertility Caused by Cisplatin in Rats

Article *in* Gynecologic and Obstetric Investigation · January 2015

DOI: 10.1159/000367657 · Source: PubMed

CITATIONS
12

READS
152

7 authors, including:

Ayse Nur Aksoy
Nenehatun hospital, Erzurum,Turkey
35 PUBLICATIONS **327** CITATIONS
SEE PROFILE

Suna Kabil Kucur
Dumlupinar Üniversitesi
44 PUBLICATIONS **289** CITATIONS
SEE PROFILE

Gonca Batmaz
Bezmiâlem Vakif Üniversitesi
49 PUBLICATIONS **272** CITATIONS
SEE PROFILE

Ilay Gözükara
11 PUBLICATIONS **93** CITATIONS
SEE PROFILE

Original Article

Gynecologic and Obstetric Investigation

Gynecol Obstet Invest
DOI: 10.1159/000367657

Received: March 11, 2014
Accepted after revision: August 14, 2014
Published online: ■■■

The Role of Antioxidant Activity in the Prevention and Treatment of Infertility Caused by Cisplatin in Rats

Ayse Nur Aksoy[a] Suna Kabil Kucur[b] Gonca Batmaz[c] Ilay Gözükara[b]
Mehmet Aksoy[d] Nezahat Kurt[e] Renad Mammadov[f]

[a]Department of Obsterics and Gynecology, Nenehatun Hospital, Erzurum, [b]Department of Obstetrics and Gynecology, Faculty of Medicine, Dumlupınar University, Kutahya, [c]Department of Obstetrics and Gynecology, Faculty of Medicine, Bezmialem Vakif University, Istanbul, [d]Department of Anaesthesiology and Reanimation, Faculty of Medicine, Ataturk University, Erzurum, [e]Department of Biochemistry, Faculty of Medicine, Ataturk University, Erzurum, and [f]Department of Pharmacology, Faculty of Medicine, Recep Tayyip Erdogan University, Rize, Turkey

Key Words
Antioxidant activity · Oxidative stress · Cisplatin · Infertility · Rat

Abstract
Background/Aims: To investigate the importance of antioxidant activity in infertility caused by cisplatin in rats. ***Methods:*** Rats in cisplatin control (CG), Vitamin E + cisplatin (ECG), Vitamin C + cisplatin (CCG), Hippophae rhamnoides extract (HRE) + cisplatin (HRECG), and thiamine pyrophosphate (TPP) + cisplatin (TPPCG) groups were injected intraperitoneally (ip) with (100 mg/kg) Vitamin E, Vitamin C, HRE, and TPP, respectively. One hour later, ip cisplatin was administered (5 mg/kg), and then antioxidant medications were continued for 10 days. Cisplatin + Vitamin E (CEG-1), cisplatin + Vitamin C (CCG-1), cisplatin + HRE (CHREG-1), and cisplatin + TPP (TPPCG-1) rats received cisplatin (5 mg/kg, ip) and were kept for 10 days. At the end of that period, rats received antioxidant medications for 10 days. (n = 12, for each group). Six rats from each group were sacrificed. Ovaries were removed to measure malondialdehyde, total glutathione, glutathione S-transferase, and glutathione reductase levels. The remaining rats were kept in a suitable laboratory environment. ***Results:*** Cisplatin-induced oxidative stress was best prevented by HRE, Vitamin E, Vitamin C, and TPP, in that order. However, infertility caused by cisplatin was only prevented and treated by TPP. ***Conclusion:*** Oxidative stress is not a major component in the pathogenesis of cisplatin-associated infertility.

Introduction

Early ovarian insufficiency and infertility appear in women receiving chemotherapy; anticancer drugs have been reported to induce this effect by reducing the number of primordial follicles and leading to ovarian atrophy [1]. The cisplatin is a platinum-based chemotherapeutic agent [2], and infertility develops in some patients receiving cisplatin therapy [3]. Therefore, clinical trials [4] and experimental studies [5] for the prevention of infertility resulting from chemotherapy have been carried out; research suggests that oxidative stress that rises with chemotherapy is a factor leading to infertility [5]. In addition, increased oxidative stress and decreased levels of antioxidants were reported in patients received chemotherapy

0378–7346/15/0000–0000$39.50/0

E-Mail karger@karger.com
www.karger.com/goi

Ayse Nur Aksoy
Department of Obsterics and Gynecology
Nenehatun Hospital, Dr. Refik Saydam Street
TR–25070 Palandöken, Erzurum (Turkey)
E-Mail draysenuraksoy@hotmail.com

[6, 7]. Borekci et al. [5] also determined an increase in the levels of malondialdehyde (MDA), a product of lipid peroxidation, and a decrease in levels of total glutathione (tGSH), an endogenous antioxidant, in ovarian tissue of rats receiving cisplatin. They also reported infertility in all rats with high MDA and low tGSH levels [5]. This information from the literature suggests that infertility associated with cisplatin and other chemotherapeutics arises from increasing oxidative stress in ovarian tissue. However, it has also been suggested that cytotoxicity caused by chemotherapy does not stem from an increase in free oxygen radicals and that antioxidants used together with chemotherapeutics do not reduce the antitumor effect of chemotherapeutic drugs [8, 9]. A similar study also showed that antioxidants did not reduce the antitumor effect of cisplatin and other chemotherapeutics [10]. This indicates that there are more important mechanisms in addition to oxidative stress in the pathogenesis of infertility caused by chemotherapeutics.

The Vitamins E and C, Hippophae rhamnoides (sea buckthorn) oil, and thiamine pyrophosphate (TPP) tested against cisplatin-induced infertility in this study have known antioxidant activities. These drugs are vitamins with varying levels of antioxidant capacity. Vitamin C is the most powerful of the water-soluble Vitamins [11]. However, the antioxidant effect of Vitamin E is reported to be greater than that of Vitamin C [12]. Sea buckthorn oil is extracted from the plant Hippophae rhamnoides. Hippophae rhamnoides oil is a member of the family Eleagnaceae [13]. The plant contains α, β, and γ carotene, riboflavin, Vitamin C, tocopherol, tocotrienol, folic acid, and tannin [14, 15]. Hippophae rhamnoides extract (HRE) has been shown to have antioxidant and antitoxic properties [16]. TPP has been reported to exhibit antioxidant activity against ischemia reperfusion-induced oxidative ovarian damage [17].

The aim of this study was to investigate the role and importance of antioxidant activity in the prevention and treatment of infertility caused by cisplatin. For this purpose, we compared the effectiveness of Vitamin E, Vitamin C, HRE, and TPP in the prevention and treatment of cisplatin-induced oxidative stress-related infertility.

Materials and Methods

One hundred twenty female Albino Wistar rats weighing 200–210 g for use in the study were obtained from the Ataturk University Medical Experimental Practice and Research Center. Rats were kept in groups at normal room temperature (22°C) and fed animal chow before the experiment. This study was performed in accordance with the national guidelines for the use and care of laboratory animals with the approval of Ataturk University Local Ethical Committee of Experimental Animals (the protocol number: 24).

The Thiopental sodium, Vitamin E (α-tocopherol), Vitamin C (ascorbic acid), HRE, TPP, and cisplatin (50 mg/100 ml; Cisplatin-Ebewe) used in the study were obtained from IE Ulagay-Turkey, Kocak-Turkey, Bayer-Germany, Biota-Turkey, Biopharma-Russia, and Liba-Turkey, respectively.

The rats to be used in the experiment were divided into six groups: Cisplatin control (CG), Vitamin E + cisplatin (ECG), Vitamin C + cisplatin (CCG), HRE + cisplatin (HRECG), TPP + cisplatin (TPPCG), and healthy (HG). The ECG (n = 12), CCG (n = 12), HRECG (n = 12), and TPPCG (n = 12) group rats were injected intraperitoneally (ip) with 100 mg/kg Vitamin E, Vitamin C, HRE, and TPP, respectively. One hour after administration of these drugs, all animal groups (excluding the HG group) were given ip cisplatin with a single dose of 5 mg/kg. Vitamins E and C, HRE and TPP medications were continued to be given to rats once a day for 10 days. The HG (n = 12) rats were given distilled water as solvent by the same route. At the end of that period, six rats from each group were sacrificed with a high dose of anesthesia (ip Thiopental with dose of 50 mg/kg). Then, the ovaries were removed to measure MDA, tGSH, glutathione S-transferase (GST), and glutathione reductase (GSHRd) levels. A suitable laboratory environment was established for the remaining six animals each in the CG, ECG, CCG, HRECG, TPPCG, and HG groups (n = 6, for all groups) to enable them to reproduce. Rats that did not become pregnant and give birth during three months were regarded as infertile. On the other hand, cisplatin + Vitamin E (CEG-1), cisplatin + Vitamin C (CCG-1), cisplatin + HRE (CHREG-1), and cisplatin + TPP (CTPPG-1) groups were also established. CEG-1 (n = 12), CCG-1 (n = 12), CHREG-1 (n = 12), and CTPPG-1 (n = 12) group rats received ip cisplatin with a single dose of 5 mg/kg and were kept for 10 days. At the end of that period, rats in the CEG-1, CCG-1, CHREG-1, and CTPPG-1 groups received ip Vitamin E, Vitamin C, HRE, or TPP, respectively, with a single dose of 100 mg/kg once a day for 10 days. Then, six rats from each group were sacrificed with a high-dose anesthesia and ovarian tissues were removed to measure MDA, tGSH, GST, and GSHRd levels. The remaining rats in groups (n = 6, for each group) were kept for 3 months in a suitable laboratory environment to enable them to reproduce. Animals that did not become pregnant and give birth during this period were regarded as infertile.

At this stage of the study, 0.2 g was measured from each extracted ovary. Tissues were made up to 2 ml in 1.15% potassium chloride solution for MDA assay and in pH 7.5 phosphate buffer for other measurements and homogenized in an iced environment. Then, they were centrifuged at 10,000 rpm at +4°C for 15 min. The supernatant part was used as the analysis specimen.

Tissue MDA levels were determined spectrophotometrically according to the method described by Ohkawa et al. [18]. This method is based on the spectrophotometric measurement at a wavelength of 532 nm of the absorbance of the pinkish complex formed by thiobarbituric acid with MDA at high temperature (95°C). Results were expressed as μmol/g protein.

The amount of GSH in the total homogenate was measured according to the method described by Sedlak and Lindsay [19]. DTNB [5, 5′-ditiyobis (2-nitrobenzoik acid)] in the measurement environment is a disulfide chromogen and is easily reduced by sul-

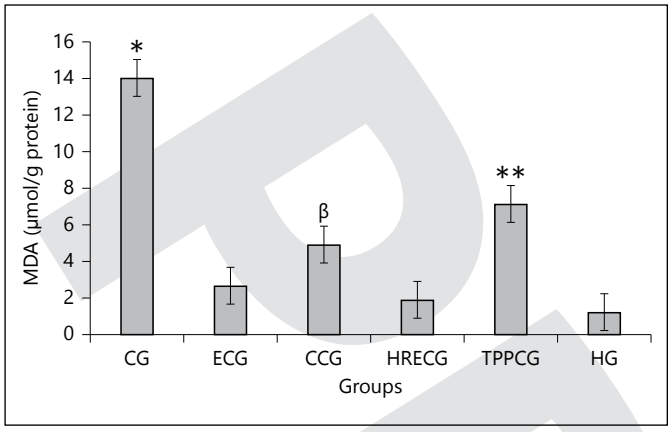

Fig. 1. Comparisons of groups in terms of MDA. * $p < 0.001$, compared with other groups; ** $p < 0.05$, compared with CCG group and ** $p < 0.001$, compared with other groups; $^{\beta}$ $p < 0.05$, compared with ECG and TPPCG groups and $^{\beta}$ $p < 0.001$, compared with other groups. CG, cisplatin group; ECG, Vitamin E + cisplatin group; CCG, Vitamin C + cisplatin group; HRECG, hippophae rhamnoides extract + cisplatin group; TPPCG, thiamine pyrophosphate + cisplatin group and HG, healthy group.

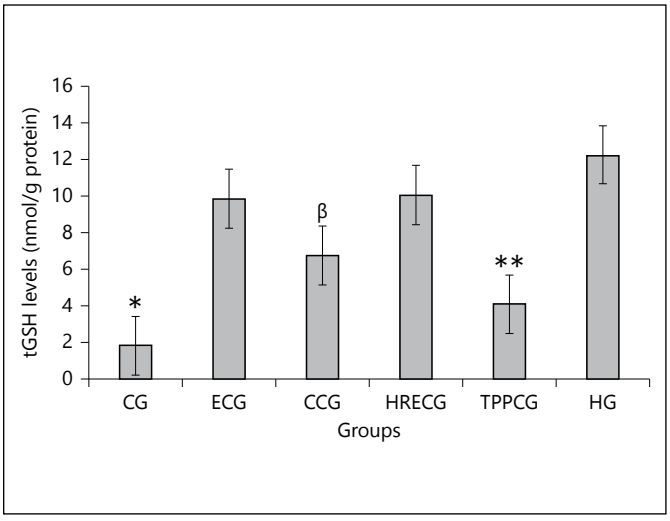

Fig. 2. Comparisons of groups in terms of tGSH levels. * $p < 0.001$, ** $p < 0.001$, compared with other groups; $^{\beta}$ $p < 0.05$, compared with ECG group and $^{\beta}$ $p < 0.001$, compared with other groups; $^{\alpha}$ $p < 0.05$ compared with HRECG group and $^{\alpha}$ $p < 0.001$, compared with other groups.

phydryl group compounds. The resulting yellow color was measured spectrophotometrically at 412 nm. Results were expressed as nmol/g protein.

GSHRd activity was determined spectrophotometrically by measuring the rate of NADPH oxidation at 340 nm according to the method described by Carlberg and Mannervik [20]. After tissue homogenization, supernatant was used for GSHRd measurement. After the NADPH and GSSG addition, the chronometer was on and absorbance was measured for 5 min by 30-min intervals with 340 nm spectrophotometric methods. Results were expressed as µ/g protein.

GST activity was determined according to the method described by Habig and Jakoby [21]. Briefly, the enzyme's activity was assayed spectrophotometrically at 340 nm in a 4-ml cuvette containing 0.1 M PBS (pH 6.5), 30 mM GSH, 30 mM 1-chloro-2,6-dinitrobenzene, and tissue homogenate. Results were expressed as µ/g protein.

The experimental results were expressed as the mean ± standard deviation (SD). Significance of differences among groups was determined using one-way ANOVA, followed by the Tukey test. Statistical analysis was performed using a statistical software package, version 18.0 (SPSS, Chicago, Ill., USA) and $p < 0.05$ was considered statistically significant.

Results

Ovarian tissue MDA concentrations (µmol/g protein) were higher in CG (14 ± 2.6) compared with ECG (2.6 ± 0.4), CCG (4.9 ± 0.7), HRECG (1.9 ± 0.6), TPPCG (7.1 ± 1.3), and HG (1.2 ± 0.3) groups ($p < 0.001$, for all). Also, ovarian tissue MDA concentrations were higher in TPPCG group compared with other groups ($p < 0.05$, for the CCG group; $p < 0.001$, for other groups) (fig. 1). Ovarian tissue tGSH levels (nmol/g protein) were lower in the CG (1.8 ± 0.5) compared with the ECG (9.8 ± 0.6), CCG (6.7 ± 0.5), HRECG (10 ± 0.9), TPPCG (4.1 ± 0.8), and HG (12.2 ± 1.5) groups ($p < 0.001$, for all). Also, ovarian tissue tGSH levels were lower in the TPPCG group compared with other groups ($p < 0.001$, for all) (fig. 2). HRECG group had higher ovarian tissue tGSH levels compared with the CCG and TPPG ($p < 0.05$). Ovarian tissue GSHRd levels (µ/g protein) were similar between CG (2.5 ± 0.5) and TPPCG (5.4 ± 1.3) ($p > 0.05$), whereas they were lower in the CG group compared with the ECG (14.8 ± 1.6), CCG (8.3 ± 2.1), HRECG (15.1 ± 2.4), and HG (17.3 ± 1.7) ($p < 0.001$, for all). Also, TPPCG group had lower ovarian tissue GSHRd levels compared with the ECG, HRECG, and HG ($p < 0.001$), whereas TPPCG group had similar ovarian tissue GSHRd levels compared with the CG and CCG ($p > 0.05$) (fig. 3). Ovarian tissue GST levels (µ/g protein) were lower in the CG (3.1 ± 1.1) compared with the ECG (20.7 ± 1.8), CCG (13.3 ± 1.9), HRECG (23.1 ± 2.3), and HG (27.1 ± 1.7) groups ($p < 0.001$, for all). However, ovarian tissue GST levels were similar between the CG and TPPCG (7.0 ± 1.7) groups ($p > 0.05$). Ovarian tissue GST levels were lower in the TPPCG group compared with the CCG, ECG, HRECG, and HG groups ($p < 0.05$, for CCG group;

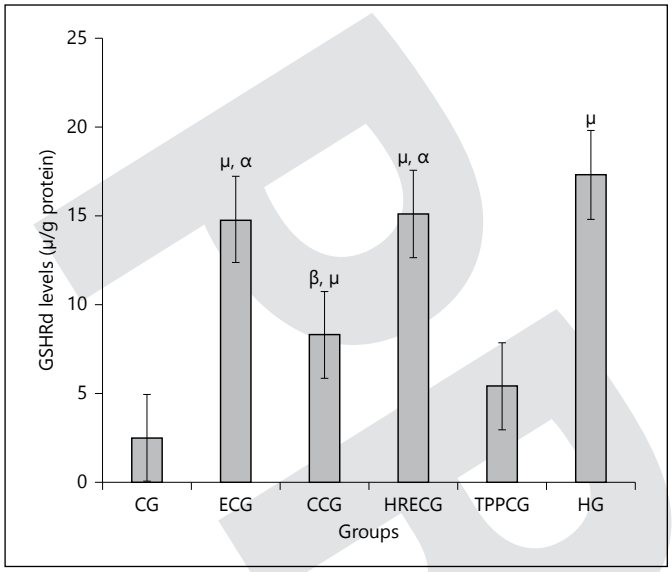

Fig. 3. Comparisons of groups in terms of GSHRd levels. $^{\mu}$ $p < 0.001$, compared with CG group; $^{\alpha}$ $p < 0.001$, compared with TPPCG group; $^{\beta}$ $p < 0.001$, compared with ECG, HRECG and HG groups.

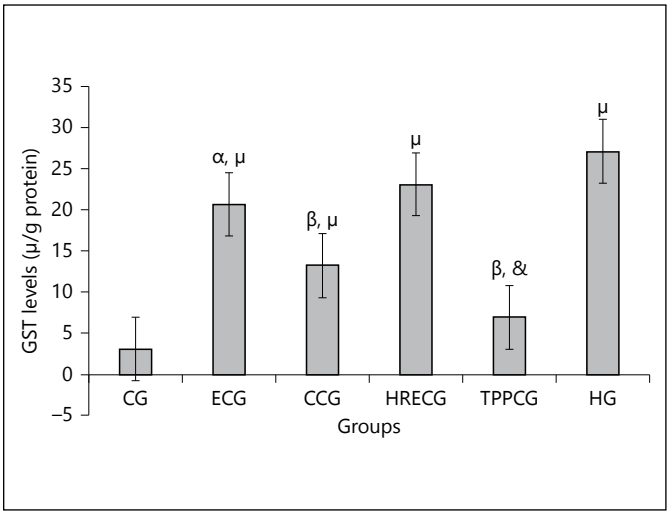

Fig. 4. Comparisons of groups in terms of GST levels. $^{\mu}$ $p < 0.001$, compared with CG group; $^{\&}$ $p < 0.05$, compared with CCG group, $^{\beta}$ $p < 0.001$, compared with ECG, HRECG and HG groups; $^{\alpha}$ $p < 0.05$, compared with HG group, $^{\alpha}$ $p < 0.001$, compared with CG, CCG, and TPPCG groups.

Table 1. Ovarian tissue MDA and tGSH levels in the study group received antioxidan therapy after 10 days following cisplatin administration

Parameters	CG	CEG-1	CCG-1	CHREG-1	CTPPG-1	HG
MDA, μmol/g protein	14±2.6*	1.8±0.2**	3.0±0.6	1.3±0.3**	4.9±0.8	1.2±0.3**
tGSH, nmol/g protein	1.8±0.5*	10.2±0.8†	7.2±0.6$^{\ddagger, \S, \|}$	10.4±0.6†	5.0±0.6$^{\ddagger, \S}$	12.2±1.5¶

Results were presented as mean ± standard deviation.
* $p < 0.001$, compared with other groups; ** $p < 0.001$, $^{\|}$ $p < 0.05$ compared with the CTPPG-1 group; ‡ $p < 0.001$, ¶ $p < 0.05$ compared with CEG-1 and CHREG-1 groups; † $p < 0.05$, § $p < 0.001$, compared with the HG group.

Table 2. Ovarian tissue GSHRd and GST levels in study group received antioxidan therapy after 10 days following cisplatin administration

Parameters	CG	CEG-1	CCG-1	CHREG-1	CTPPG-1	HG
GSHRd levels, μ/g protein	2.5±0.5	15.7±2.0$^{\|, \dagger, **}$	10.6±1.3$^{\|}$	16±2.6$^{\|, \dagger, \ddagger}$	6.2±1.7*	17.3±1.7$^{\|, \dagger}$
GST levels, μ/g protein	3.1±1.1	22.8±2$^{\|, \dagger}$	16±2.3$^{\|, \dagger, \P}$	22.2±4.3$^{\|, \dagger, \S}$	7.8±1.7$^{*, \P}$	27.1±1.7$^{\|, \dagger}$

Results were presented as mean ± standard deviation.
* $p < 0.05$, $^{\|}$ $p < 0.001$ compared with the CG group; † $p < 0.001$, compared with the CTPPG-1 group; ** $p < 0.001$, ‡ $p < 0.05$, compared with the CCG-1 group; ¶ $p < 0.001$, § $p < 0.05$, compared with the HG group.

$p < 0.001$, for other groups). Also, HRECG group had similar ovarian tissue GST levels compared with ECG and HG groups (fig. 4).

Ovarian tissue MDA, tGSH, GSHRd, and GST levels in second study groups are given in tables 1 and 2. Ovarian tissue MDA concentrations (μmol/g protein) were higher in the CG (14 ± 2.6) compared with the CEG-1 (1.8 ± 0.2), CCG-1 (3.0 ± 0.6), CHREG-1 (1.3 ± 0.3), CTPPG-1 (4.9 ± 0.8), and HG (1.2 ± 0.3) groups ($p < 0.001$, for all). However, CTPPG-1 group had higher

ovarian tissue MDA concentrations compared with the CG, CEG-1, CHREG-1, and HG groups ($p < 0.001$, for all). Ovarian tissue tGSH levels (nmol/g protein) were found to be lower in the CG (1.8 ± 0.5) than those of CEG-1 (10.2 ± 0.8), CCG-1 (7.2 ± 0.6), CHREG-1 (10.4 ± 0.6), CTPPG-1 (5.0 ± 0.6), and HG (12.2 ± 1.5) groups ($p < 0.001$, for all). Lower ovarian tissue tGSH levels were found in the CTPPG-1 group compared with the CG, CCG-1, CHREG-1, and HG groups ($p < 0.05$, for CCG-1 group; $p < 0.001$, for others) (table 1). Ovarian tissue GSHRd levels (μ/g protein) were lower in the CG (2.5 ± 0.5) compared with the CEG-1 (15.7 ± 2.0), CCG-1 (10.6 ± 1.3), CHREG-1 (16 ± 2.6), CTPPG-1 (6.2 ± 1.7), and HG (17.3 ± 1.7) groups ($p < 0.05$ for the CTPPG-1 group; $p < 0.001$, for others). Ovarian tissue GSHRd levels were similar among CEG-1, CHREG-1, and HG groups ($p > 0.05$), whereas they were lower in the CTPPG-1 group compared with the CEG-1, CHREG-1, and SG groups ($p < 0.001$). Ovarian tissue GST levels (μ/g protein) were found to be lower in the CG group (3.1 ± 1.1) compared with the CEG-1 (22.8 ± 2), CCG-1 (16 ± 2.3), CHREG-1 (22.2 ± 4.3), CTPPG-1 (7.8 ± 1.7), and HG (27.1 ± 1.7) ($p < 0.05$, for the CTPPG-1 group; $p < 0.001$, for others). Ovarian tissue GST levels were similar between CEG-1 and HG, whereas HG group had higher ovarian tissue GST levels compared with the CG, CCG-1, CHREG-1, and CTPPG-1 ($p < 0.05$, for CHREG-1 and $p < 0.001$, for others).

None of the six rats taken for reproduction from CG, ECG, and CCG groups gave birth within 3 months. One rat in the HRECG group gave birth on the 40th day, whereas the other five did not give birth. Five of the six rats taken for reproduction in the TPPCG gave birth, on days 35, 36, 38, 40, and 41, whereas one was infertile. All of the HG group rats gave birth, on days 30, 31, 34, 36, 37, and 39. No rats from the CEG-1, CCG-1, and CHREG-1 groups gave birth in 3 months, whereas all the rats in the CTPPG-1 group gave birth, on days 31, 34, 39, 46, 47, and 51.

Discussion

This study examined the value of antioxidant activity in the prevention and treatment of infertility caused by cisplatin and investigated the effectiveness of Vitamins E and C, HRE and TPP in preventing and treating cisplatin-related infertility in rats. Our results showed that cisplatin-induced increases in oxidant parameters and decreases in antioxidant parameters were best prevented by HRE administration. However, cisplatin-related infertility was prevented and treated by TPP administration, whereas it was not prevented and treated by Vitamins E and C, and HRE.

Various reasons such as ischemia reperfusion injury and using chemotherapeutic agents may lead to oxidative ovarian damage [22–25]. Studies demonstrated an increase in oxidant parameters and a decrease in antioxidant levels in ovarian tissue with oxidative damage in comparison with healthy tissues [22–25]. The antioxidant defense system in the human body includes enzymatic (GSHRd, GST, etc.) and nonenzymatic (tGSH, ascorbate, vitamin C, etc.). components [26]. In our study, cisplatin-caused oxidative damage was found to be associated with an increase in ovarian tissue MDA levels and a decrease in ovarian tissue tGSH, GSHRd, and GST levels. Similar to our results, Altuner et al. [24] found increased oxidative stress and decreased levels of antioxidants in rats that received cisplatin. They also reported that infertility develops in association with this ovarian oxidative damage and this damage-related infertility is prevented with mirtazapine. In another study, Li et al. [25] reported that cisplatin-damaged ovarian reserve is associated with an increase in oxidative stress and a decrease in antioxidation capability. They also reported that Mesna (2-mercaptoethane sulfonate) may protect ovarian reserve from cisplatin-induced damage through potent antioxidation without obvious inhibition of the anti-tumor efficacy of cisplatin.

In our study, however, Vitamin E, which was the most powerful antioxidant and widely used in infertility treatment, was found to be insufficient to prevent cisplatin-related infertility. Vitamin E is an antioxidant agent that neutralizes peroxides and oxygen radicals, prevents the lipid breakdown reaction chain, and protects against oxidative damage by settling on cell and organelle membranes [12, 27]. The drugs used in chemotherapy are thought to exhibit cytotoxic effects by establishing oxidative stress [5–7]. In theory, therefore, antioxidants are thought to reduce the side-effects of these agents by scavenging free radicals. However, there are many reports in the literature that antioxidants do not reduce the effectiveness of chemotherapy [7, 28]. In experiments in which antioxidants have been administered together with chemotherapeutics, free radical scavengers have been shown not to reduce the antitumor effect of cytotoxic drugs such as cisplatin [7, 29]. Although chemotherapy has been shown to lead to free radical formation, it has not been implicated in free oxygen radical formation [9, 10].

Vitamin C is another antioxidant drug used in this study. The antioxidant activity of Vitamin C is the most

powerful among the water-soluble vitamins, albeit not as great as that of Vitamin E [11]. However, our study showed that, similar to Vitamin E, Vitamin C was unable to prevent cisplatin-related infertility. Vitamin C traps peroxyl radicals in the aqueous phase. In addition, it enters into a reaction with tocopheroxyl radical, and this results in tocopherol. The function of Vitamin C is to increase radical trapping potential by regenerating tocopherol and protecting membranes against peroxidation [30]. It has been reported in the literature that chemotherapy establishes oxidative stress by leading to antioxidant loss, that this oxidative stress causes cell mutation [31], and that Vitamins C and E protect cells against this mutagenic effect [10, 32]. This is not in agreement with our own experimental results.

Interestingly, only one rat from the HRECG group gave birth, whereas almost all those from the TPPCG did so in this study, as TPP's antioxidant activity is lower than those of HRE and Vitamins C and E. In contrast, HRE has powerful antioxidant and antitoxic effects [16]. These antioxidant effects arise from its carotene α, β and γ, riboflavin, Vitamin C, tocopherol, tocotrienol, folic acid, and tannin contents [14, 15]. The TPP we used is an active metabolite of thiamine. TPP is a coenzyme of carboxylase enzyme that plays a role in the oxidative decarboxylation of mitochondria [33, 34]. Yapca et al. [35] reported that infertility in rats subjected to unilateral ovariectomy and ischemia/reperfusion injury in the contralateral ovary is prevented by TPP whereas it is not prevented by thiamine. Borekci et al. [5] also showed that TPP is effective against cisplatin-induced infertility in rats.

This study investigated the effects of TPP, HRE, and Vitamins C and E in groups in which they were administered at the same time as cisplatin therapy (ECG, CCG, HRECG, and TPPCG) and in groups in which they were administered after cisplatin therapy (ECG-1, CCG-1, HRECG-1, and TPPCG-1). The reason for administering drugs in parallel to cisplatin therapy (prophylaxis) was to investigate the effects of drugs against toxic radical formation before injury induced in ovarian tissue with cisplatin. Drugs were administered after cisplatin therapy in order to reveal their effects against developing toxic radicals and the oxidative damage caused by radicals. However, in addition to preventing radical formation before injury, antioxidants repair oxidative damage that has already occurred, neutralize various reactive side products, and prevent the reduction of oxidized biomolecules [36]. This information from the literature is compatible with our study results, because TPP, HRE, and Vitamins C and E significantly prevented a rise in MDA concentrations and a decrease in tGSH, GSHRd, and GST levels, both in groups in which they were administered before cisplatin therapy and when they were administered afterward. MDA is the final product of lipid peroxidation, and excess production leads to cell damage [37]. GSH, a nonenzymatic antioxidant, has an important role in protecting cells from oxidative damage. GSHRd and GST are enzymatic antioxidants and have higher activity in healthy tissue compared with damaged tissue [38]. GSH, GSHRd, and GST are important endogenous defense mechanisms in cell protection [38, 39]. A rise in MDA concentrations and a decrease in GSH levels were observed in rat ovarian tissue that was administered cisplatin; cisplatin-induced infertility was also prevented by the administration of TPP [5]. Our scan of the literature revealed no previous comparative studies regarding the prevention and treatment of infertility with Vitamins C and E, HRE, and TPP in rats given cisplatin.

In conclusion, we observed an increase in oxidant parameters and a decrease in antioxidant parameters in rat ovarian tissues administered cisplatin. Infertility developed in all rats that were administered cisplatin. An increase in oxidant parameters and a decrease in antioxidant parameters occurring with cisplatin were best prevented by HRE, Vitamin E, Vitamin C, and TPP, in that order. However, infertility caused by cisplatin in rats was prevented and treated by TPP, whereas it was not prevented and treated by HRE, Vitamin E, and Vitamin C. We suggested that oxidative stress is not a major component of cisplatin-related infertility. We also concluded that the administration of TPP to patients treated or still being treated with cisplatin may be beneficial in preventing and treating possible future cisplatin-related infertility.

References

1 Sanchez AM, Giorgione V, Viganò P, Papaleo E, Candiani M, Mangili G, Panina-Bordignon P: Treatment with anticancer agents induces dysregulation of specific Wnt signaling pathways in human ovarian luteinized granulosa cells in vitro. Toxicol Sci 2013;136:183–192.

2 Pabla N, Dong Z: Cisplatin nephrotoxicity: mechanisms and renoprotective strategies. Kidney Int 2008;73:994–1007.

3 Reinmuth S, Hohmann C, Rendtorff R, Balcerek M, Holzhausen S, Müller A, Henze G, Keil T, Borgmann-Staudt A: Impact of chemotherapy and radiotherapy in childhood on fertility in adulthood: the FeCt-survey of childhood cancer survivors in Germany. J Cancer Res Clin Oncol 2013;139:2071–2078.

4 Oktay K, Buyuk E, Libertella N, Akar M, Rosenwaks Z: Fertility preservation in breast cancer patients: a prospective controlled comparison of ovarian stimulation with tamoxifen and letrozole for embryo cryopreservation. J Clin Oncol 2005;23:4347–4353.
5 Borekci B, Yapca OE, Turan MI, Gul MA, Isaoglu U: Prevention of ovarian oxidative stress – related infertility associated with cisplatin in rats with thiamine pyrophosphate. Lat Am J Pharm 2013;32:512–517.
6 White AC, Sousa AM, Blumberg J, Ryan HF, Fanburg BL, Kayyali US: Plasma antioxidants in subjects before hematopoietic stem cell transplantation. Bone Marrow Transplant 2006;38:513–520.
7 Block KI, Koch AC, Mead MN, Tothy PK, Newman RA, Gyllenhaal C: Impact of antioxidant supplementation on chemotherapeutic efficacy: A systematic review of the evidence from randomized controlled trials. Cancer Treat Rev 2007;33:407–418.
8 Crohns M, Liippo K, Erhola M, Kankaanranta H, Moilanen E, Alho H, Kellokumpu-Lehtinen P: Concurrent decline of several antioxidants and markers of oxidative stress during combination chemotherapy for small cell lung cancer. Clin Biochem 2009;42:1236–1245.
9 Jaakkola K, Lähteenmäki P, Laakso J, Harju E, Tykkä H, Mahlberg K: Treatment with antioxidant and other nutrients in combination with chemotherapy and irradiation in patients with small-cell lung cancer. Anticancer Res 1992;12:599–606.
10 Weijl NI, Cleton FJ, Osanto S: Free radicals and antioxidants in chemotherapy-induced toxicity. Cancer Treat Rev 1997;23:209–240.
11 Silalahi J: Anticancer and health protective properties of citrus fruit components. Asia Pac J Clin Nutr 2002;11:79–84.
12 Sokol RJ: The coming of age of vitamin E. Hepatology 1989;9:649–653.
13 Rousi A: The genus Hippophue L. A taxonomic study. Ann Bot Fennici 1971;8:177–227.
14 Guliyev VB, Gul M, Yildirim A: Hippophae rhamnoides L.: chromatographic methods to determine chemical composition, use in traditional medicine and pharmacological effects. J Chromatogr B Analyt Technol Biomed Life Sci 2004;812:291–307.
15 Andersson SC, Rumpunen K, Johansson E, Olsson ME: Tocopherols and tocotrienols in sea buckthorn (Hippophae rhamnoides L.) berries during ripening. J Agric Food Chem 2008;56:6701–6706.
16 Geetha S, Sai Ram M, Mongia SS, Singh V, Ilavazhagan G, Sawhney RC: Evaluation of antioxidant activity of leaf extract of Seabuckthorn (Hippophae rhamnoides L.) on chromium(VI) induced oxidative stress in albino rats. J Ethnopharmacol 2003;87:247–251.
17 Demiryilmaz I, Sener E, Cetin N, Altuner D, Akcay F, Suleyman H: A comparative investigation of biochemical and histopathological effects of thiamine and thiamine pyrophosphate on ischemia-reperfusion induced oxidative damage in rat ovarian tissue. Arch Pharm Res 2013;36:1133–1139.
18 Ohkawa H, Ohishi N, Yagi K: Assay for lipid peroxides in animal tissues by thiobarbituric acid reaction. Anal Biochem 1979;95:351–358.
19 Sedlak J, Lindsay RH: Estimation of total, protein-bound, and nonprotein sulfhydryl groups in tissue with Ellman's reagent. Anal Biochem 1968;25:192–205.
20 Carlberg I, Mannervik B: Glutathione reductase. Methods Enzymol 1985;113:484–90.
21 Habig WH, Jakoby WB: Assays for differentiation of glutathione S-transferases. Methods Enzymol 1981;77:398–405.
22 Ozler A, Turgut A, Görük NY, Alabalik U, Basarali MK, Akdemir F: Evaluation of the protective effects of CoQ_{10} on ovarian I/R injury: an experimental study. Gynecol Obstet Invest 2013;76:100–106.
23 Celik M, Aksoy AN, Aksoy H, Aksoy Y, Halici Z: Sildenafil reduces ischemia-reperfusion injury in rat ovary: biochemical and histopathological evaluation. Gynecol Obstet Invest 2014;78:162–167.
24 Altuner D, Gulaboglu M, Yapca OE, Cetin N: The effect of mirtazapine on cisplatin-induced oxidative damage and infertility in rat ovaries. Scientific World Journal 2013;2013:327240.
25 Li X, Yang S, Lv X, Sun H, Weng J, Liang Y, Zhou D: The mechanism of mesna in protection from cisplatin-induced ovarian damage in female rats. J Gynecol Oncol 2013;24:177–185.
26 Carden DL, Granger DN: Pathophysiology of ischaemia-reperfusion injury. J Pathol 2000;190:255–266.
27 MacDonald-Wicks LK, Garg ML: Vitamin E supplementation in the mitigation of carbon tetrachloride induced oxidative stress in rats. J Nutr Biochem 2003;14:211–218.
28 Simone CB 2nd, Simone NL, Simone V, Simone CB: Antioxidants and other nutrients do not interfere with chemotherapy or radiation therapy and can increase kill and increase survival, part 1. Altern Ther Health Med 2007;13:22–28.
29 Seifried HE, Anderson DE, Fisher EI, Milner JA: A review of the interaction among dietary antioxidants and reactive oxygen species. J Nutr Biochem 2007;18:567–579.
30 Schäfer L, Thorling EB: Lipid peroxidation and antioxidant supplementation in old age. Scand J Clin Lab Invest 1990;50:69–75.
31 Ladas EJ, Jacobson JS, Kennedy DD, Teel K, Fleischauer A, Kelly KM: Antioxidants and cancer therapy: a systematic review. J Clin Oncol 2004;22:517–528.
32 Bairati I, Meyer F, Gélinas M, Fortin A, Nabid A, Brochet F, Mercier JP, Têtu B, Harel F, Mâsse B, Vigneault E, Vass S, del Vecchio P, Roy J: A randomized trial of antioxidant vitamins to prevent second primary cancers in head and neck cancer patients. J Natl Cancer Inst 2005;97:481–488.
33 Klooster, A, Leuvenink H, Gans R, Bakker S: Tissue thiamine deficiency as potential cause of delayed graft function after kidney transplantation: thiamine supplementation of kidney donors may improve transplantation outcome. Med Hypotheses 2007;69:873–878.
34 Gangolf M, Czerniecki J, Radermecker M, Detry O, Nisolle M, Jouan C, Martin D, Chantraine F, Lakaye B, Wins P, Grisar T, Bettendorff L: Thiamine status in humans and content of phosphorylated thiamine derivatives in biopsies and cultured cells. PLoS One 2010;5:e13616.
35 Yapca OE, Turan MI, Cetin N, Borekci B, Gul MA: Use of thiamine pyrophosphate to prevent infertility developing in rats undergoing unilateral ovariectomy and with ischemia reperfusion induced in the contralateral ovary. Eur J Obstet Gynecol Reprod Biol 2013;170:521–525.
36 Sorg O: Oxidative stress: a theoretical model or a biological reality? C R Biol 2004;327:649–662.
37 Southorn PA, Powis G: Free radical in medicine. I. Chemical nature and biologic reactions. Mayo Clin Proc 1988;63:381–389.
38 Valko M, Rhodes CJ, Moncol J, Izakovic M, Mazur M: Free radicals, metals and antioxidants in oxidative stress-induced cancer. Chem Biol Interact 2006;160:1–40.
39 Kaya H, Oral B, Ozgüner F, Tahan V, Babar Y, Delibaş N: The effect of melatonin application on lipid peroxidation during cyclophosphamide therapy in female rats. Zentralbl Gynakol 1999;121:499–502.