

The Effect of Protective Factors on Induced Toxins in Genital Organs

R. Kheradmandi (MSc)¹, S.G.A. Jorsaraei (PhD)^{*2}

1.Department of Anatomy, Faculty of Medicine, Babol University of Medical Sciences, Babol, I.R.Iran.

2.Infertility and Reproductive Health Research Center, Health Research Institute, Babol University of Medical Sciences, Babol, I.R.Iran.

J Babol Univ Med Sci; 19(10); Oct 2017; PP: 48-56

Received: Apr 18th 2017, Revised: Jul 3th 2017, Accepted: Aug 11th 2017.

ABSTRACT

BACKGROUND AND OBJECTIVE: Different toxins have harmful effects on Genital organs. Exposure to contaminants as well as physical stress can increase the rate of infertility. Antioxidant products are highly considered, because they can react to pollutants and remove free oxygen radicals. This study investigates the effects of proteins on the protection of the reproductive system against toxins.

METHODS: This review article was conducted using keywords including “protective” and “organophosphorus toxins”, and searching on online databases such as Pubmed, Science Direct, Scopus and Google Scholar. The related articles were extracted and classified.

FINDINGS: The results of about 80 articles, selected out of 140 new and valid articles, suggest that elements such as arsenic, cyclophosphamide, cadmium, diazinon and gossypol reduce the reproduction rate and damage body tissues. Elements such as N-acetylcysteine, carnitine, corsetine, black seed oil, palm powder, ginseng, honey and saffron are proteins that may have protective effects against potential damage.

CONCLUSION: Based on the results of this study, the natural proteins that can be found in plants, fruits and food can be used to prevent the damage to the reproductive system caused by various toxins.

KEY WORDS: *Protective, Teratogen, Sperm, Embryo.*

Please cite this article as follows:

Kheradmandi R, Jorsaraei SGA. The Effect of Protective Factors on Induced Toxins in Genital Organs. J Babol Univ Med Sci. 2017;19(10):48-56.

* Corresponding author: S.G.A. Jorsaraei (PhD)

Address: Infertility and Reproductive Health Research Center, Health Research Institute, Babol University of Medical Sciences, Babol, I.R.Iran

Tel: +98 11 32199596

E-mail: alijorsara@yahoo.com

Introduction

A wide range of toxins have destructive effects on Genital organs. In addition to acute injury, some of them also cause irreparable damage in the long run (1,2).

Many of these mechanisms are based on the reactions of reactive oxygen species (ROS). A high dose of ROS can increase lipid peroxidation and, by changing the antioxidant system, induces DNA damage and ultimately leads to changes in the gene expression and apoptosis of the cells of the body(3). Exposure to contaminants and toxins, as well as physical stress, increases the rate of infertility, which has become a major dilemma in present societies (4–6). Some studies on animal specimens show that toxins have a negative effect on sperm parameters (7).

In addition, levels of testosterone, FSH, and LH, and the stages of spermatogenesis, steroidogenesis, and testicular cell cycle are affected by contaminants (8–10). The lipid membrane of the cell is targeted by the ROS and causes a change in its structure. The lipid peroxidation created by ROS increases the permeability and fluidity of the sperm membrane (11). Significant and irreversible consequences of lipid peroxidation in the sperm membrane include reduced motility, inhibition of cellular respiration, intracellular enzyme leakage, DNA damage and failure of sperm to penetrate the oocyte (12).

Many studies have investigated the inhibition of oxidative stress, and enzymatic and non-enzymatic antioxidants in order to maintain the quality of semen (13, 14). In addition, the production of drugs or antioxidants that respond to the toxic effects of various pollutants, especially in the removal of oxygen free radicals, was much considered (15).

Antioxidant substances are commonly found in people's diet. The molecular mechanism of the antioxidants works in a way that treats or overcomes some of the oxidative stress and organ toxicity disorders (16, 17). Even, freezing the gametes with what is common in infertility centers may lead to adverse changes in the composition of the lipid membrane and their viability and cause apoptosis and planned cell death by increasing the damage to the DNA (18, 19). Therefore, the presence of some protectives in the mechanism of freezing gametes is also justifiable. This review article investigates the materials and protectives that have a protective effect in the exposure of reproductive system to toxics and harmful drugs.

Methods

This review article was designed through a library-based search method. Initially, the subjects were evaluated and searched by selecting the words protective, toxins, drugs, teratogen, sperm, ovum, testicular tissue, ovary and embryos. The vocabulary was classified and categorized as primary and secondary in terms of validity in the process of scientometric analysis.

The main keywords were “protectives and gametes”, and other words were used as sub – keywords. Subsequently, related articles were extracted by placing words in Pubmed, Science Direct, Scopus, and Google Scholar websites. The resulting material was classified. Subsequently, the material was mentioned based on the necessity and expression of the protectives that were more comprehensive and at the same time easier to use. More than 100 titles of related articles were selected in this regard. Protectives were divided into two groups of synthetic and natural and material was mentioned to be more useful than others.

Results

The results of about 80 selected articles out of a 140 new and valid articles suggest that many synthetic materials or medicinal plants have a protective effect against elements that have harmful or possibly destructive effects on the tissues of the body, and they increase the tissue's resistance or have a restorative effect. Some studies indicate that protective substances can be used if harmful elements enter the human body unintentionally or by necessity or if exposure to them is inevitable. Olive oil can somewhat protect the damaged Genital tissues from a soy-based diet. Olive oil increases sperm parameters, including motility and count, and reduces abnormalities to some extent (20). Polydatin also has a protective effect and can significantly improve the effect of lipid peroxidation, DNA degradation and antioxidant enzymatic activities of arsenic (21).

Quercetin is a flavonoid or quasi-flavonoid element found in many herbs and fruits. This element has antioxidant properties, which is also a protective and can increase the daily sperm production rate, while its use with arsenic prevents the destruction of testicular tissue to a large extent (22). If N-acetylcysteine (NAC) is used together with sodium fluoride, it reduces the effect of oxidative stress and reduces damage to DNA. The use of NAC before the administration of an

element such as sodium decreases lipid peroxidation (23). Crocin is an element that increases sperm parameters, testicular weight, and increases testosterone levels. This substance inhibits the nicotine degradation effect, which reduces the level of testosterone, inhibits the sperm parameters, including the motility and count, the rate of viability, and also decreases the diameter of the seminiferous ducts, and improves them to some extent (24).

The *phyllanthus fraternus* plant can improve or inhibit the oxidative effect of cyclophosphamides, which greatly reduces the gonadosomatic indices, the epididymal sperm count, and the motility and viability of the sperm (25). Melatonin and BQ-123, especially if used together, reduce the testicular tissue damage in the exposure to cigarette smoke and prevent damage to germinal cells (26). *Spirulina platensis* is one of the microorganisms that has an antioxidant effect and has been declared by the World Health Organization as a superior food. This plant reduces the oxidative stress and toxicity of sodium arsenic (27). Carnitine has a protective effect on the toxicity of cadmium.

Cadmium reduces sperm count, germinal cells and sperm viability (28). *Satureja khuzistanica* essential oil (SKEO) is anti-apoptotic and has a protective effect against the damage of sperm and seminiferous tubules caused by busulfan, and it can improve sperm parameters by preventing the degradation of germinal tissue. The neurotrophic factor isolated from the brain is effective on sperm function, oxidative stress and cell membrane, and increases the sperm motility and prevents apoptosis induced by ROS (30).

Neuronal growth factor also affects the quality of sperm in men with asthenospermia during sperm cryopreservation, and by maintaining the sperm motility and viability rate, it can inhibit DNA fragmentation during cryopreservation (31). *Nigella sativa* oil has a protective effect and improves sperm parameters against the effects of nicotine (32). The use of dates powder is recommended as a protective element against the reduction of sperm parameters caused by nicotine (33). Selenium and diazinon have an adverse effect on sperm parameters. This becomes more intense when used together (34). Trolox is also a protective. This element has a protective effect on the quality of sperm in patients with normospermia and oligospermia during cryopreservation and can prevent DNA fracture (35). *Arctium lappa* extract reduces the toxicity of cadmium (36). Korean red ginseng improves the negative effect of busulfan to some

extent (37). Using intraperitoneal injection of selenium and nicotine will be able to reduce lipid peroxidation, enhance the activity of antioxidants and improve the metabolism of nicotine (38). Cyclophosphamide causes abnormalities in the sperm, chromosome and decreases the expression of some of the cytokines. Honey works as a protective element against its toxicity (39). Saffron neutralizes the toxicity of cadmium to a great extent (40).

Ellagic acid neutralizes the toxicity of sodium valproate in the genital system and acts as a protective and relatively short acting agent (41). The protective effect of strawberry, grape and cranberry powder is also considered to affect the acrylamide-induced toxicity and can restore the parameters of the sperm (42). Allicin, proanthocyanidin, polyphenol, ultracid and isoflavones have anti-mutagenic activity and reduce the toxicity of methotrexate by inhibition of the toxicity and rate of sperm abnormalities (43).

Gallic acid overcomes the toxicity of cyclophosphamide and compensates for some of its adverse effects (44). Humanin has a protective effect against testicular heating and hormonal disorders, and toxicity caused by cyclophosphamide and prevents apoptosis in reproductive cells (45). Dexrazoxane is capable of improving the aneuploidy induced by doxorubicin, which causes oxidative changes and DNA damage (46). Gossypol is a chemical in flaxseed that reduces fertility (47).

This disorder is partially compensable by vitamin E (48). N-acetylcysteine reduces nicotine-induced abnormalities (49, 50). The protective effect of bee pollen in endocrine disorders, as well as malathion-induced toxicity of reproductive system has been reported (51). N-acetylcysteine and L-carnitine can prevent oocyst damage caused by follicular fluid in infertile women with endometriosis to some extent (52). Green tea extract has a protective effect, but its positive effect is not much confirmed (53).

Nortriptyline has protective effect against the apoptosis of the testicular germ cells, and by decreasing malondialdehyde, it brings the activity of antioxidant enzymes to normal level (54). Punicalagin is found in pomegranate. It increases the activity of antioxidant enzymes and reduces the peroxidation of lipids (55). The polyphenolic extract of green tea preserves sperm viability and health against damage from cryopreservation (56). *Satureja khuzistanica* essential oil can improve fertility capacity and sperm parameters against cyclosporine toxicity to some

extent (57). Rolipram is one of the elements that acts as a protective element against ionizing radiation and prevents reduced sperm count and motility (58). Lycopene decreases the sperm apoptosis during cryopreservation (59). There is a chance of maintaining motility, viability and health of sperm membrane in using cholesterol cyclodextrin against oxidative damage induced by hydrogen peroxide (60). Polysaccharide in the *Rhodiola rosea* plant can somewhat improve parameters such as sperm motility, mitochondrial activity, acrosome surface health and plasma membrane (61). Zonisamide is able to reduce the LH and FSH hormones and the endocrine gene expression and hormone secretion. Sperm abnormalities and DNA fragmentation increase. However, melatonin can improve the damage of zonisamide to some extent (62).

L-carnitine also maintains sperm motility and vitality, as well as testosterone hormones, estrogen, and expression of the LC3-II and Beclin-1 genes against cyclophosphamide (63). Hyaluronic acid can improve motility, the health of the sperm membrane and the acrosome region, mitochondrial activity, and the activity of the catalase and superoxide dismutase enzymes (64). Phenylhydrazine and ethyl pyruvate can also improve the parameters of the sperm to some extent (65). Decorsin prevents the oxidative stress of TM3 cells and improves sperm parameters in people with cryptospermia, increases the expression of Nrf2 gene and prevents cell apoptosis (66). Pumpkin seeds and ginger extract can prevent the damage of sperm parameters against cyclophosphamide to some extent (67). Vitamin E improves the parameters of sperm motility and count in rats receiving methyl methanesulfonate. The levels of androgen, antioxidant and testosterone are largely compensated by the antioxidant properties of vitamin E (68).

The moraceae plant prevents the reduction of sperm parameters with its protective effect on toxicity and oxidative stress caused by cadmium (69). The combination of ascorbic acid and vitamin E can have a protective effect on the motility and viability of frozen sperm (70). Methotrexate is able to increase malondialdehyde and increase the number of damaged sperms. Cranberry consumption can reduce these damages and increase the chance of fertility (71). Glutathione lipid peroxidation prevents DNA fracture, free radicals and hydrogen peroxide in frozen sperm (72). Cisplatin is a drug that induces oxidative stress and reduces sperm production. This damage is somewhat improved by resveratrol injection (73). With the increase in LDL, sperm count may reduce. Therefore, the use of vitamin E and astaxanthin as

protective elements may somewhat compensate (74). L-carnitine can maintain the parameters of frozen sperm in people with astenospermia and normospermia (75), decrease cisplatin, testosterone levels, sperm count and motility, and increase sperm abnormality rates. The lotus plant can slightly compensate for these disorders (76). Glutathione maintains the parameters of frozen sperm and can reduce sperm apoptosis (77).

Diethylhexyl phthalate reduces testicular weight, and sperm count and motility, reduces testosterone levels and ultimately causes oxidative stress and tissue abnormalities. Quercetin significantly compensates the disorders and abnormalities (78). Melatonin can prevent lipid peroxidation from ionizing radiation (79). Red palm oil compensates for the reduction of sperm parameters caused by lead acetate (80). Trolox has a negative effect on frozen sperm motility. Glutathione can reduce the percentage of apoptosis in frozen sperm (81). Nicotine increases the concentration of thiobarbituric acid and nitric oxide. Vitamin C and selenium can significantly reduce the effects of nicotine (82). Rosuvastatin can improve the motility and count of sperm, control testicular inflammation and cell death by reducing the expression of the INOS gene expression (83). The use of selenium and n-acetylcysteine in infertile men increases testosterone levels and sperm parameters. The co-administration of these two antioxidants provides better results (84). Foxil is a protein that facilitates the reaction between the sperm and the uterine tube for fertility in humans. The suppression of this protein in the sperm cryopreservation improves oxidative stress. Asialofetuin is a subunit of this protein that has protective effect on the proper performance of the plasma membrane and sperm motility (85). Egg yolk improves the fluidity of the plasma membrane and sperm viability (86).

Yangjing can compensate for tissue changes induced by cyclophosphamide and increase sperm count and motility by decreasing the apoptosis rate. It also genetically reduces the ability to express the Bcl-2 gene (87). Benztropine increases sperm abnormalities, and levels of malondialdehyde and 8-OHdG gene, and decreases the activity of glutathione peroxidase and superoxide dismutase enzymes. Vitamin E improves some of the damages of benztropine to testicles (88). Metronidazole is able to impair sperm production, antioxidant enzymes, lactate dehydrogenase, alkaline phosphatase, and lipid peroxidation in testicular tissue. Tribulus terrestris can improve such damages to some extent (89). Zinc reduces the oxidative stress on reproductive system tissue caused by lead and improves spermatogenesis (90). Melatonin can compensate for some of the side effects of cadmium (91). Considering that sperm parameters decrease with

cryopreservation due to oxidative stress, melatonin can increase sperm motility and vitality, and reduce oxidative stress and malondialdehyde (92). Humanin drug prevents apoptosis of germ cells and leukocytes as a result of receiving cyclophosphamide. Humanin inhibits the IGF-1 agent (93).

Discussion

Protectives or protective elements have a variety of applications and are widely used in medicines and foods. They are naturally found in plants, fruits and foods. They are often mentioned as accompanying elements or supplements. Many of the plants, fruits, and synthetic elements can be found in the protectives because of their antioxidant properties.

Protectives can be used to prevent reproductive system damage. The genital system may be exposed to destructive elements. In addition, it might be exposed to elements that cause cytotoxicity and damage the process of gamete formation. Therefore, to prevent further destruction, or to improve and heal the lesions, fruits, foods, or elements with protective properties can be used. Fruits such as grapes and apples have antioxidant properties and prevent the toxicity of oxidative stress in male Genital organs. Placing such foods that are easy to access in the food basket can protect the reproductive system. Cranberry and strawberry powder are among the antitoxic plants and are part of the protective elements. It seems that abundant phenolic compounds and anthocyanin in strawberries are involved in the repair of damages (41, 71).

Antioxidants are elements that are found in many foods, plants and fruits. Antioxidants can improve the quality of sperm and they have protective effect on the cells of the body (67). Some drugs also have anti-inflammatory effects and increase the resistance of cells to the apoptosis process, while preventing the destruction of sex cells (82). Although it may not be possible to use plants like the lotus, which is antitoxic (20), for public consumption, it can be advisable to use olive oil, especially fresh and ripe olives (76). In some cases, vegetable oils that have antioxidant properties can be used as a protective for people undergoing chemotherapy. For example, one can use savory oil is found mostly in northeastern parts of Iran or ginseng plant (29, 37). Some plants, such as nigella sativa or dates powder can control the effects of opium to some extent and placing them in the food basket is highly important (32, 33). Honey has the potential to reduce

the toxicity of sex cells and act as a protective agent. Honey increases the resistance of body cells, including sex cells, to various toxins such as agricultural pesticides (39, 51). The use of some seasonings that are consumed with different foods can be taken into consideration and be considered as a protective agent along with the main foods. Saffron is a good seasoning that protects toxicity in spermatogenesis to some extent (40). Green tea extract (53), pumpkin seeds, ginger extract (55, 67), and red palm oil (89) are some of the elements that have a protective and antitoxic effect on the testicular tissue and can prevent infertility in addition to treatment. Ginger is a food seasoning and is more effective with green tea.

Avoiding excessive consumption of certain foods such as egg yolks that exacerbate oxidative stress is recommended (85). The maintenance of sperm parameters is not limited to in vivo condition, and it is also important to maintain it under in vitro condition and in frozen form. Some people want their sperm to be stored in a good condition. Therefore, several drugs and elements such as Neural Growth Factor (30), Trolux (34), Polyphenol (56), Hyaluronic acid (64), Ascorbic acid and Vitamin E (70), L-carnitine (75) and Glutathione (80) have been suggested as protective elements in the process of cryopreservation, in order to maintain the quality of sperm.

These elements are commonly used in infertility centers and sperm banks. Of course, their use is not the routine of the assisted reproductive technology (ART) labs, and some of them are undergoing a research process. One of the most controversial chemical drugs that has been most used in recent years is selenium. Some studies do not consider selenium to be a protective agent (34), but many have also identified it as a protective agent for its antitoxic effect against opium. However, selenium is not stronger than vitamin C. Thus, there is the suggestion to use plants, foods and fruits that are rich in vitamin C. Vitamin E is an important factor in inhibiting oxidative stress. This vitamin plays a protective role. Given that this vitamin protects mitochondrial damage and oxidative stress in male Genital organs (48), it can be used according to the defined dose. Various studies indicate the use of protective elements along with food.

Acknowledgments

Hereby, we express our deepest sense of gratitude and indebtedness to all colleagues whose opinions have somehow been used in this article.

References

1. Santos FW1, Graça DL, Zeni G, Rocha JB, Weis SN, Favero AM. Sub-chronic administration of diphenyl diselenide potentiates cadmium-induced testicular damage in mice. *Reprod Toxicol*. 2006;22(3):546-50
2. Rikans LE, Yamano T. Mechanisms of cadmium mediated-acute hepatotoxicity. *Toxicol Mol Toxicol*. 2000;14(2):110-7.
3. Stohs SJ, Bagchi D, Hassoun E, Bagchi M. Oxidative mechanisms in the toxicity of chromium and cadmium ions. *Environ Pathol Toxicol Oncol*. 2001;20(2):77-88.
4. Meeker JD1, Rossano MG, Protas B, Diamond MP, Puscheck E, Daly D, et al. Cadmium, lead, and other metals in relation to semen quality: human evidence for molybdenum as a male reproductive toxicant, *Environ. Health Perspect*. 2008;116(11):1473-9.
5. Marchetti F1, Eskenazi B, Weldon RH, Li G, Zhang L, Rappaport SM, et al. Occupational exposure to benzene and chromosomal structural aberrations in the sperm of Chinese men, *Environ. Health Perspect*. 2012;120(2):229-34.
6. Jorsaraei G, Beiki A, Yousefnia Pasha Y, Alizadeh Navaei R. The in vitro effects of hinosa and diazinon on human sperm parameters. *J Babol Univ Med Sci*. 2005;7(2):30-4. [In Persian].
7. Lu Z1, Wang S, Sun Z, Niu R, Wang J. In vivo influence of sodium fluoride on sperm chemotaxis in male mice, *Arch. Toxicol*. 2014;88(2):533-9.
8. Reddy PS1, Pushpalatha T, Reddy PS. Suppression of male reproduction in rats after exposure to sodium fluoride during early stages of development. *Naturwissenschaften*. 2007;94(7):607-11.
9. Pushpalatha T, Srinivas M, Reddy PS. Exposure to high fluoride concentration in drinking water will affect spermatogenesis and steroidogenesis in male albino rats. *Biometals*. 2005;18(3):207-12.
10. Huang C, Niu R, Wang J. Toxic effects of sodium fluoride on reproductive function in male mice. *Fluoride*. 2007;40:162-168.
11. Awda BJ, Mackenzie-Bell M, Buhr MM. Reactive oxygen species and boar sperm function. *Biol Reprod*. 2009 Sep;81(3):553-61.
12. Bucak MN1, Ateşşahin A, Varışlı O, Yüce A, Tekin N, Akçay A. The influence of trehalose, taurine, cysteamine and hyaluronan on ram semen microscopic and oxidative stress parameters after freeze thawing process. *Theriogenol*. 2007;67(5):1060-7.
13. Maia Mda S1, Bicudo SD, Sicherle CC, Rodello L, Gallego IC. Lipid peroxidation and generation of hydrogen peroxide in frozen-thawed ram semen cryopreserved in extenders with antioxidants. *Anim Reprod Sci*. 2010;122(1-2):118-23.
14. Santiani A, Evangelista S, Sepúlveda N, Risopatrón J, Villegas J, Sánchez R. Addition of superoxide dismutase mimics during cooling process prevents oxidative stress and improves semen quality parameters in frozen/thawed ram spermatozoa. *Theriogenol*. 2014;82(6):884-9.
15. Frei B, Higdon JV. Antioxidant activity of tea poly phenols *in vivo*: Evidence from animal studies. *J Nutr*. 2003;133(10):3275-84.
16. Havsteen BH. The biochemistry and medical significance of the flavonoids. *Pharmacol Ther*. 2002;96(2-3):67-202.
17. Justice T, Christensen G. Sperm cryopreservation methods. *Spermatog Meth Protocol*. 2013;209-15.
18. Malo C, Gil L, Gonzalez N, Martínez F, Cano R, de Blas I, Espinosa E. Antioxidant supplementation improves boar sperm characteristics and fertility after cryopreservation: comparison between cysteine and rosemary (*Rosmarinus officinalis*). *Cryobiol*. 2010;61(1):142-7.
19. Venkatesh S, Singh A, Shamsi MB, Thilagavathi J, Kumar R, Mitra DK, et al. Clinical significance of sperm DNA damage threshold value in the assessment of male infertility. *Reprod Sci*. 2011;18(10):1005-13.
20. El-Kholy TA, Al-Abbadi HA, Qahwaji D, Al-Ghamdi AK, Shelat VG, Sobhy HM, et al. Ameliorating effect of olive oil on fertility of male rats fed on genetically modified soya bean. *Food Nutr Res*. 2015;59:10-34.
21. Ince S1, Avdatek F2, Demirel HH3, Arslan-Acaroz D4, Goksel E5, Kucukurt I. Ameliorative effect of polydatin on oxidative stress-mediated testicular damage by chronic arsenic exposure in rats. *Andrologia*. 2016;48(5):518-24.
22. Jahan S, Rehman S, Ullah H, Munawar A, Ain QU, Iqbal T. Ameliorative effect of quercetin against arsenic-induced sperm DNA damage and daily sperm production in adult male rats. *Drug Chem Toxicol*. 2016;39(3):290-6.
23. Feng D, Huang H, Yang Y, Yan T, Jin Y, Cheng X, et al. Ameliorative effects of N-acetylcysteine on fluoride-induced oxidative stress and DNA damage in male rats' testis. *Gen Toxicol Environ Mutag*. 2015;792:35-45.
24. Salahshoor MR, Khazaei M, Jalili C, Keivan M. Crocin improves damage induced by nicotine on a number of reproductive parameters in male mice. *Int J Fertil Steril*. 2016;10(1):71-8.
25. Singh S, Lata S, Tiwari KN. Antioxidant potential of *Phyllanthus fraternus* Webster on cyclophosphamide induced changes in sperm characteristics and testicular oxidative damage in mice. *Ind J Experiment Biol*. 2015;53:647-56.

26. Aslan H, Kesici H, Karaca Zi, Özyurt B, Taş U, Ekici F, et al. Beneficial effects of melatonin and BQ-123 on the rat testis damage caused by cigarette smoke. *Turk J Med Sci*. 2015;45(1):11-7.
27. Bashandy SA, El Awdan SA, Ebaid H, Alhazza IM. Antioxidant potential of spirulina platensis mitigates oxidative stress and reprotoxicity induced by sodium arsenite in male rats. *Oxid Med Cel longevit*. 2016 (2016).
28. Yari A, Asadi MH, Bahadoran H, Dashtnavard H, Imani H, Naghii MR. Cadmium toxicity in spermatogenesis and protective effects of L-carnitine in adult male rats. *Biolog Trace Element Res*. 2010;137(2):216-25.
29. Nasimi P, Vahdati A, Tabandeh M, Khatamsaz S. Cytoprotective and anti-apoptotic effects of Satureja khuzestanica essential oil against busulfan-mediated sperm damage and seminiferous tubules destruction in adult male mice. *Andrologia*. 2016;48(1):74-81.
30. Najafi A, Amidi F, Gilani S, Moawad A, Asadi E, Khanlarkhni N, et al. Effect of brain-derived neurotrophic factor on sperm function, oxidative stress and membrane integrity in human. *Andrologia*. 2017;49(2).
31. Saeednia S, Nashtaei MS, Bahadoran H, Aleyasin A, Amidi F. Effect of nerve growth factor on sperm quality in asthenozoospermic men during cryopreservation. *Reproduct Biol Endocrinol*. 2016;14(1):1.
32. Cho Ping N, Hashim NH, Hasan Adli DS. Effects of Nigella sativa (Habbatus sauda) oil and nicotine chronic treatments on sperm parameters and testis histological features of rats. *Evid-Base Complement Alternat Med*. 2014.
33. Saeed K, Tahir M, Lone KP. Effect of Phoenix dactylifera (date palm) pit powder on nicotine induced spermatotoxicity in adult albino mice. *J Pak Med Assoc*. 2015;65(1):43-8.
34. Toman R, Hluchy S, Cabaj M, Massanyi P, Roychoudhury S, Tunegova M. Effect of separate and combined exposure of selenium and diazinon on rat sperm motility by computer assisted semen analysis. *J Trace Element Med Biol*. 2016;38:144-9.
35. Nekoonam S, Nashtaei MS, Zangi BM, Amidi F. Effect of Trolox on sperm quality in normozoospermia and oligozoospermia during cryopreservation. *Cryobiol*. 2016;72(2):106-11.
36. de Souza Predes F, Diamante M, Foglio M, Dolder H. Effects of arctium lappa on cadmium-induced damage to the testis and epididymis of adult wistar rats. *Biol Trace Element Res*. 2016;173(2):362-371.
37. Jung S-W, Kim H-J, Lee B-H, Choi S-H, Kim H-S, Choi Y-K, et al. Effects of Korean Red Ginseng extract on busulfan-induced dysfunction of the male reproductive system. *J Gin Res*. 2015;39(3):243-9.
38. Sreekala S, Indira M. Effects of exogenous selenium on nicotine-induced oxidative stress in rats. *Biological Trace Element Res*. 2009;130(1):62-71.
39. Zoheir KMA, Harisa GI, Abo-Salem OM, Ahmad SF. Honey bee is a potential antioxidant against cyclophosphamide-induced genotoxicity in albino male mice. *Pakistan J Pharmaceutical Sci*. 2015;28(3):973-81.
40. Yari A, Sarveazad A, Asadi E, Raouf Sarshoori J, Babahajian A, Amini N, et al. Efficacy of crocus sativus L. on reduction of cadmium-induced toxicity on spermatogenesis in adult rats. *Andrologia*. 2016;48(10):1244-52.
41. Girish C, Shweta O, Raj V, Balakrishnan S, G'boy Varghese R. Ellagic acid modulates sodium valproate induced reproductive toxicity in male Wistar rats. *Indian J Physiol Pharmacol*. 2014;58(4):416-22.
42. Zhao M, Liu X, Luo Y, Guo H, Hu X, Chen F. Evaluation of protective effect of freeze-dried strawberry, grape, and blueberry powder on acrylamide toxicity in mice. *J food Sci*. 2015;80(4):869-74.
43. Ying W, GU SB, Hao L, HE JY, Li L, YANG JB. Evaluation of Protective Effects of Bioactive Phytochemicals against Methotrexate in Salmonella typhimurium TA1535/pSK1002 coupled with Micronucleus Assay. *Biomed Environment Sci*. 2016;29(2):148-52.
44. Oyagbemi A, Omobowale T, Saba A, Adedara I, Olowu E, Akinrinde A, et al. Gallic acid protects against cyclophosphamide-induced toxicity in testis and epididymis of rats. *Andrologia*. 2016;48(4):393-401.
45. Surampudi P, Chang I, Lue Y, Doumit T, Jia Y, Atienza V, et al. Humanin protects against chemotherapy-induced stage-specific male germ cell apoptosis in rats. *Androl*. 2015;3(3):582-9.
46. Attia S, Ahmad S, Bakheet S. Impact of dexrazoxane on doxorubicin-induced aneuploidy in somatic and germinal cells of male mice. *Cancer Chemoth pharmacol*. 2016;77(1):27-33.
47. Neda MahdinezhadGorji, Seyed Gholam Ali Jorsaraei, Vida Hojati, Ebrahim Zabihi, Asieh Khalilpour, Zainab Abedian, Eisa Tahmasbpour. A comparison between the cytotoxicity induced by gossypol in two testicular cell lines .Iran J Toxicol . 2015;27(1):1188-95. [In Persian].
48. Santana AT, Guelfi M, Medeiros HC, Tavares MA, Bizerra PF, Mingatto FE. Mechanisms involved in reproductive damage caused by gossypol in rats and protective effects of vitamin E. *Biol Res*. 2015;48(1):43.
49. Okamura AMNC, Gomes PXL, de Oliveira GV, de Araújo FYR, Tomaz VS, Chaves Filho AJM, et al. N-acetylcysteine attenuates nicotine-induced kindling in female periadolescent rats. *Pro Neuro-Psychopharmacol Biol Psychiat*. 2016;67(3):58-65.

50. Bowers M, Jackson A, Maldoon P, Damaj M. N-acetylcysteine decreased nicotine reward-like properties and withdrawal in mice. *Psychopharmacol.* 2016;233(6):995-1003.
51. Kandiel MM, El-Asely AM, Radwan HA, Abbass AA. Modulation of genotoxicity and endocrine disruptive effects of malathion by dietary honeybee pollen and propolis in Nile tilapia (*Oreochromis niloticus*). *J Adv Res.* 2014;5(6):671-84.
52. Giorgi VS, Da Broi MG, Paz CC, Ferriani RA, Navarro PA. N-acetyl-cysteine and L-carnitine prevent meiotic oocyte damage induced by follicular fluid from infertile women with mild endometriosis. *Reprod Sci.* 2016;23(3):342-51.
53. Mosbah R, Yousef MI, Mantovani A. Nicotine-induced reproductive toxicity, oxidative damage, histological changes and haematotoxicity in male rats: The protective effects of green tea extract. *Exp Toxicol Pathol.* 2015;67(3):253-9.
54. Yazdani I, Ghazi-Khansari M, Saeedi Saravi S, Nobakht M, Majdani R, Rezayat S, et al. Nortriptyline protects testes against germ cell apoptosis and oxidative stress induced by testicular ischaemia/reperfusion. *Andrologia.* 2017;49(2):
55. Rao F, Tian H, Li W, Hung H, Sun F. Potential role of punicalagin against oxidative stress induced testicular damage. *Asian J Androl.* 2016;18(4):627-32.
56. Kitaji H, Ookutsu S, Sato M, Miyoshi K. Preincubation with green tea polyphenol extract is beneficial for attenuating sperm injury caused by freezing-thawing in swine. *Animal Sci J.* 2015;86(11):922-8.
57. Najafi G, Farokhi F, Jalali AS, Akbarizadeh Z. Protection against cyclosporine-induced reprotoxicity by Satureja khuzestanica essential oil in male rats. *Int J Fertil Sterilit.* 2016;9(4):548.
58. Lee W, Son Y, Jang H, Bae MJ, Kim J, Kang D, et al. Protective effect of administered rolipram against radiation-induced testicular injury in mice. *World J Mens Health.* 2015;33(1):20-9.
59. Liang Z, Guo K, Dai X, Liu L, Xu S, Zhao L, et al. Protective effect of lycopene on human spermatozoa during cryopreservation and its mechanism. *Nat J Androl.* 2015;21(6):521.
60. Naseer Z, Ahmad E, Aksoy M, Küçük N, Serin I, Ceylan A, et al. Protective effect of cholesterol-loaded cyclodextrin pretreatment against hydrogen peroxide induced oxidative damage in ram sperm. *Cryobiol.* 2015;71(1):18-23.
61. Yang S-M, Wang T, Wen D-G, Hou J-Q, Li H-B. Protective effect of rhodiola rosea polysaccharides on cryopreserved boar sperm. *Carbohydr Polym.* 2016;135:44-7.
62. Khalil W, Abdu F. Protective effect of melatonin against zonisamide-induced reproductive disorders in male rats. *Arch Med Sci.* 2015;11(3):660-9.
63. Zhu B, Zheng Y-f, Zhang Y-y, Cao Y-s, Zhang L, Li X-g, et al. Protective effect of L-carnitine in cyclophosphamide-induced germ cell apoptosis. *J Zhejiang Univ Sci B.* 2015;16(9):780-7.
64. Qian L, Yu S, Zhou Y. Protective effect of hyaluronic acid on cryopreserved boar sperm. *Int J Biological Macromol.* 2016;87:287-9.
65. Mozafari AA, Shahrooz R, Ahmadi A, Malekinjad H, Mardani K, editors. Protective effect of ethyl pyruvate on mice sperm parameters in phenylhydrazine induced hemolytic anemia. *Veter Res Forum.* 2016;7(1):63-8.
66. Bae WJ, Ha US, Choi JB, Kim KS, Kim SJ, Cho HJ, et al. Protective effect of decursin extracted from angelica gigas in male infertility via Nrf2/HO-1 signaling Pathway. *Oxidative Med Cell long.* 2016;2016:5901098.
67. Aghaie S, Nikzad H, Mahabadi JA, Taghizadeh M, Azami-Tameh A, Taherian A, et al. Protective effect of combined pumpkin seed and ginger extracts on sperm characteristics, biochemical parameters and epididymal histology in adult male rats treated with cyclophosphamide. *Anatomical science international.* 2016;91(4):382-90.
68. Tang Z, Ding W, Wang L, Jiang W, Zhang Q, Chen H, et al. Protective effect of vitamin E on methyl methanesulfonate-induced teratozoospermia in adult Sprague-Dawley rats. *Mol Med Report.* 2015;12(3):4422-6.
69. Adaramoye O, Akanni O. Protective effects of Artocarpus altilis (Moraceae) on cadmium-induced changes in sperm characteristics and testicular oxidative damage in rats. *Andrologia.* 2016;48(2): 152-63.
70. Zhao X, Li Y, Cao S, Hu J, Wang W, Hao R, et al. Protective effects of ascorbic acid and vitamin E on antioxidant enzyme activity of freeze-thawed semen of Qinchuan bulls. *Genet Mol Res.* 2015;14(1):2572-81.
71. Zarei L, Shahrooz R, Sadrkhanlou R, Malekinejad H, Ahmadi A, Bakhtiary Z. Protective effects of cornus mas extract on in vitro fertilization potential in methotrexate treated male mice. *Veter Res Forum.* 2015;6(1):55-61.
72. Ghorbani M, Vatannejad A, Khodadadi I, Amir I, Tavilani H. Protective effects of glutathione supplementation against oxidative stress during cryopreservation of human spermatozoa. *Cryo Letters.* 2016;37(1):34-40.
73. Reddy KP, Madhu P, Reddy PS. Protective effects of resveratrol against cisplatin-induced testicular and epididymal toxicity in rats. *Food Chem Toxicol.* 2016;91:65-72.

74. Roushandeh AM, Salehi I, Mortazavi M. Protective effects of restricted diet and antioxidants on testis tissue in rats fed with high-fat diet. *Iran Biomed J.* 2015;19(2):96-101.
75. Zhang W, Li F, Cao H, Li C, Du C, Yao L, et al. Protective effects of l-carnitine on astheno-and normozoospermic human semen samples during cryopreservation. *Zygote.* 2016;24(02):293-300.
76. Saral S, Ozcelik E, Cetin A, Saral O, Basak N, Aydın M, et al. Protective role of Diospyros lotus on cisplatin-induced changes in sperm characteristics, testicular damage and oxidative stress in rats. *Andrologia.* 2016;48(3):308-17.
77. Razliqi RN, Zhandi M, Shakeri M, Towhidi A, Sharafi M, Emamverdi M, et al. Protective role of glutathione in buck semen cryopreservation. *Iran J vet Res.* 2015;16(3):298.
78. Abd-Ellah M, Aly H, Mokhlis H, Abdel-Aziz A. Quercetin attenuates di-(2-ethylhexyl) phthalate-induced testicular toxicity in adult rats. *Hum Experiment Toxicol.* 2016;35(3):232-43.
79. Khan S, Adhikari JS, Rizvi MA, Chaudhury NK. Radioprotective potential of melatonin against 60 Co γ -ray-induced testicular injury in male C57BL/6 mice. *J Biomed Sci.* 2015;22(1):61.
80. Jegede A, Offor U, Azu O, Akinloye O. Red Palm Oil attenuates lead acetate induced testicular damage in adult male Sprague-Dawley rats. *Evid-Base Complement Alternat Med.* 2015;(2015).
81. Anel-López L, Garcia-Alvarez O, Maroto-Morales A, Iniesta-Cuerda M, Ramón M, Soler A, et al. Reduced glutathione addition improves both the kinematics and physiological quality of post-thawed red deer sperm. *Anim Reproduct Sci.* 2015;162:73-9.
82. Ahmed MA, Hassan K, Hassanein KM, Waly H. Role of vitamin C and selenium in attenuation of nicotine induced oxidative stress, P53 and Bcl2 expression in adult rat spleen. *Pathophysiol.* 2014;21(3):211-7.
83. Heeba GH, Hamza AA. Rosuvastatin ameliorates diabetes-induced reproductive damage via suppression of oxidative stress, inflammatory and apoptotic pathways in male rats. *Life Sci.* 2015;141:13-9.
84. Safarinejad MR, Safarinejad S. Efficacy of selenium and/or N-acetyl-cysteine for improving semen parameters in infertile men: a double-blind, placebo controlled, randomized study. *J Urol.* 2009;181(2):741-51.
85. Huang VW, Lee C-L, Lee Y-L, Lam KK, Ko JK, Yeung WS, et al. Sperm fucosyltransferase-5 mediates spermatozoa-oviductal epithelial cell interaction to protect human spermatozoa from oxidative damage. *Molecul Hum Reproduct.* 2015;21:516-526.
86. Casas I, Miller-Lux Y, Osborne B, Bonet S, Althouse GC. Testing an egg yolk supplemented diet on boars to aid in sperm adaptation at 5° C. *Sys Biol Rep Med.* 2015;61(5):253-62.
87. Zhao H, Jin B, Zhang X, Cui Y, Sun D, Gao C, et al. Yangjing Capsule Ameliorates Spermatogenesis in Male Mice Exposed to Cyclophosphamide. *Evid-Base Complement Alternat Med.* 2015;(2015).
88. Li W, Dong T, Yang K, Wu M, Tu B. Vitamin E prevents the toxic effect of benzo a pyrene on reproductive system in male SD rats. *J Cent South Univ Med sci.* 2015;40(11):1192-8.
89. Kumari M, Singh P. Tribulus terrestris ameliorates metronidazole-induced spermatogenic inhibition and testicular oxidative stress in the laboratory mouse. *Ind J pharmacol.* 2015;47(3):304.
90. Anjum MR, Madhu P, Reddy KP, Reddy PS. The protective effects of zinc in lead-induced testicular and epididymal toxicity in Wistar rats. *Toxi Indust Health.* 2017;33(3):43.
91. Li R, Luo X, Li L, Peng Q, Yang Y, Zhao L, et al. The protective effects of melatonin against oxidative stress and inflammation induced by acute cadmium exposure in mice testis. *Biolog Trace Element Res.* 2016;170(1):152-64.
92. Karimfar M, Niazvand F, Haghani K, Ghafourian S, Shirazi R, Bakhtiyari S. The protective effects of melatonin against cryopreservation-induced oxidative stress in human sperm. *Int J Immunopathol Pharmacol.* 2015;28(1):69-76.
93. Lue Y, Swerdloff R, Wan J, Xiao J, French S, Atienza V, et al. The potent humanin analogue (HNG) protects germ cells and leucocytes while enhancing chemotherapy-induced suppression of cancer metastases in male mice. *Endocrinol.* 2015;156(12):4511-21.