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A brief overview of the physiological roles of reactive oxidative species in the ovaries

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Abstract
In normal physiologically healthy conditions, there exists a harmony between the reactive oxidative species and the anti-oxidants. When this harmony disrupts it results in oxidative stress. There is known literature on the effect of oxidative stress, especially in female reproduction. This review focuses on the antioxidant system-both enzymatic, and non-enzymatic and the probable mechanisms of redox cell signalling in oocyte and embryo metabolism.

Keywords: Arginine, homoarginine, lathyrus, carcass traits, sheep

Introduction

Oxidative Stress

Oxidative stress (OS) is a consequence of discrepancy amongst the antioxidants and pro-oxidants [1] due to an increase in the levels of ROS and/or reactive nitrogen species (RNS), or an associated reduction in by antioxidants [2] (Burton and Jauniaux, 2010). If the ROS production overrides the body's antioxidant defence mechanism it creates an unfitting environment for the reproductive physiology [1] (Al-Gubory *et al*., 2010).

Oxidative Stress in Female Reproduction

The oogenesis is a cascade of processes involving recruitment, development, and atrophy. Out of the many oocytes which are recruited only one oocyte attains the status of the dominant follicle [3] (Agarwal *et al*., 2012). The meiosis I of the dominant follicle is highly susceptible to oxidative stress and is the target of ROS but contrary to the suppressive role of ROS in meiosis I the antioxidants promote the advancement of meiosis II [4] (Behrman *et al*., 2001). Further, during the development of the oocyte towards the dominant follicle stage, there is an increased demand for steroidogenesis and a concurrent increase in P450 which involves ROS formation. GSH, FSH, and Catalase are generated in response to increased Estrogen under the influence of FSH, which collectively counterbalances the ROS-induced oxidative stress and apoptosis. The LH surge which is required for the ovulation acts as a precursor for ROS and a decrease in these precursors impairs the ovulation [5] (Shkolnik *et al*., 2011). After ovulation, the corpus luteum (CL) is formed which is a source of progesterone [6] (Sugino, 2006). During this transition of CL through the early to mid-luteal phase there is an increase in Cu, Zn-SOD which gradually decreases during the regression phase in line with the progesterone concentration. The decline in the levels of Cu, Zn and SOD during the CL regression can be attributed to a) an increase in the concentration of ROS [3] (Agarwal *et al*., 2012) b) increase in PGF2α c) a reduction in ovarian blood flow [4] (Behrman *et al*., 2001).

Antioxidant System

The antioxidants can be broadly classified into two categories viz., enzymatic and non-enzymatic.

Enzymatic Anti-oxidant System

Enzymatic Antioxidant defence system enzymes comprise of glutathione (GSH), superoxide dismutase (SOD), catalase, Glutathione peroxidase (GPx) and Glutathione oxidase. The enzymatic antioxidant defence system carries out the detoxification process by transferring electrons [3] (Agarwal *et al*., 2012).

Super Oxide Dismutase (SOD) Family

SOD acts by the SO anion dismutation to H_2O_2. [3] (Agarwal *et al*., 2012) and exists in three isoenzyme forms viz., SOD 1, SOD2 SOD 3. SOD 1 is positioned in the cytosol and comprises metal co-factors viz., Cu, Zn as whereas SOD 2 containing manganese (Mn) is located in the mitochondria and SOD 3 is encrypted extracellularly. SOD enzyme families are present in multiple tissues and ovaries of different mammals. High expression levels of both SOD1 and SOD2 have been detected in luteinized granulosa and theca cells. Neither SOD1 nor SOD2 has been observed in primordial and primary follicles. SOD2 has been detected in secondary follicles, while SOD1 begins to appear in theca cells after the formation of the antral cavity.

The relatively decreased SOD activity in large follicular fluid is necessary to ensure that ROS levels reach a threshold value that is required for ovulation. An oocyte in the preovulatory follicle acquires developmental competence and very active metabolism, and during this process, a large amount of ROS can be generated; thus, SOD1 is required to neutralize O2•− in the cytoplasm of oocytes, and therefore, SOD must be maintained at a certain concentration and activity level within the follicles to guarantee a balance between O2•− and H_2O_2 for normal cellular function. After ovulation, SODs are very active in the corpus luteum, because corpus luteum function is related to progesterone levels and ROS. Interestingly, progesterone fluctuation in the luteal phase is positively correlated with SOD1 activity. Reduction in SOD1 during corpus luteum regression is accompanied by increased ROS levels. In contrast to SOD1, SOD2 concentration in the corpus luteum is enhanced in the regression phase to clear the excess ROS produced in mitochondria by cytokines and inflammatory reactions. Thus, SOD1 activity in the corpus luteum is closely correlated with progesterone secretion, while SOD2 is primarily targeted to protect the luteal cells from oxidative damage caused by inflammation. SOD1 and SOD3 are expressed in the oocyte nucleus, and SOD3 is only detected in the zona pellucida. The level of SOD1 in the oocyte nucleus is enhanced in small and medium-sized follicles. SOD1 and SOD3 potentially contribute to the protection of DNA or transcription regulation of redox-sensitive genes.

To investigate SOD regulation in relation to steroids, oestradiol and SOD were measured in the follicular fluid of patients who underwent in vitro fertilization (IVF). Interestingly, researchers found a strong positive correlation between SOD enzyme activity and intrafollicular oestradiol levels. In contrast, SOD was shown to have inhibitory effects on oestrogen synthesis by inhibiting FSH-induced aromatase activity in cultured granulosa cells, and this inhibition was found to occur at one or more post-FSH receptor sites in rat granulosa cells in vitro. LH can increase the mRNA and protein levels of SOD1, SOD2, and catalase as well as SOD activity in the bovine corpora lutea. SOD1, SOD2, and catalase mRNA levels varied in different luteal phases and reached the highest expression in the mid-luteal phase. In addition, the authors suggested that the LH-induced upregulation of antioxidant enzymes increased cell viability and maintained corpus luteum function during the luteal phase. Conversely, corpus luteum-derived SOD2 was found to serve as an LH-release inhibitory factor in sheep.

Glutathione (GSH) family of enzymes

The GSH family comprises of GPx, GST, and GSH reductase. GPx reduces the H_2O_2 to H_2O and O_2 thereby oxidizing the GSH to GSSG, GST degrades peroxidases by acting as an H+ donor and using the reduced form of GSH whereas the Glutathione reductase recycles the GSH by utilising the proton from the donor NADPH to GSSG [7] (Perkins, 2006).

Glutathione peroxidase

Glutathione peroxidase occurs in five isoforms viz., GPx1, GPx2, GPx3, GPx4 and GPx5 (Perkins, 2006). GPx1 is located in the cytoplasm, GPx2 in the gastrointestinal tract, GPx3 in plasma and epididymal fluid, GPx 4 in most of the biological membranes and specifically aids in the detoxification of the phospholipid hydroperoxide [8] (Fujii *et al*., 2005) and GPx5 is found within the epididymis [7] (Perkins, 2006). Usually, GPx4 deficiency results in cell death and Vit E protects the cell from apoptosis during the GPx4 deficiency [9] (Maiorino *et al*., 2003). Within the cell cytosol Glutathione is formed from cysteine, glutamate and glycine under the influence of enzymes γ-glutamyl cysteine synthetase and glutathione synthetase [10, 9] (Ruder *et al*., 2009 and Fujii *et al*., 2005). According to Agarwal *et al*. (2012) [3], GPX in its reduced state is the chief thiol buffer in cells and conjugates with few of the hazardous endogenous and xenobiotic compounds thereby maintaining cell homeostasis.

Glutathione

GSH is a low molecular weight thiol that is predominantly expressed in mammalian cells. GSH maintains cells in a reduced state and functions as an electron donor for some antioxidant enzymes. GSH is involved in many cellular functions, including cell proliferation, differentiation, and apoptosis. GSH can be synthesized de novo from glutamate, cysteine, and glycine via catalysis by glutamate-cysteine ligase and glutathione synthetase. According to the literature, GSH increases gamete viability and fertilization. GSH content was reported to be decreased by approximately 10-fold in unfertilized mouse oocytes. In addition, researchers have described GSH contribution to spindle formation in bovine oocytes via depletion of GSH in oocytes; after GSH depletion, the spindle poles became wider, and the spindle area increased significantly. Furthermore, high GSH content in oocytes during follicular development is related to improved development competence of the follicle. In addition to GSH, GPX has been reported to play significant roles in gametogenesis and in vitro fertilization. The activity of GPX in the follicular fluid of follicles that were subsequently fertilized was higher than that in fluid from nonfertilized follicles.

Both in vivo and in vitro studies have shown that the oocyte GSH concentration is modulated by gonadotropin signalling during the preovulatory period. In vivo, studies revealed that FSH stimulation enhanced ovarian GSH content. In addition, the GSH content in cumulus cells was gradually increased during oocyte maturation in pigs, cattle, and horses. FSH and BMP15 cotreatment promoted development competence in oocytes by distributing metabolism equally throughout the oocyte. FSH promoted glucose metabolism, while BMP15 accelerated glutathione recycling to protect against cellular OS via increased NADPH production.

Catalase

Catalase plays a critical role in ROS metabolism. For this reason, catalase has been intensely studied in recent years. Regarding catalase regulation in follicular growth, the activity

of catalase in granulosa cells from large follicles has been observed to be several times higher than that in small and medium follicles in various mammals, such as pigs, goats, and rats. In rat ovarian granulosa and theca cells, increased catalase activity can be observed during ovarian development and luteinisation. The catalase activity in total ovary homogenate was highest in the metestrus phase, declined in the oestrous and pro-oestrous stages, and reached the lowest levels in the diestrus phase. Furthermore, the activity of catalase in both rat and pig ovaries has been shown to be positively correlated with the amount of ferredoxin and cytochrome P450scc, which are two constituents of the steroidogenic electron transport chain. In steroidogenic biogenesis, oxygen free radicals such as superoxide are produced and then catalysed by SOD to H2O2. Accordingly, catalase acts as a protective factor to neutralize H2O2 to maintain ROS balance and normal steroid levels. These studies show that catalase contributes to follicular development, the oestrous cycle, and steroidogenic events in the ovaries.

Non-enzymatic antioxidants

Glutathione

Amongst the nonenzymatic antioxidants, Glutathione is the foremost and is majorly found in oocytes and embryos. According to Behrman *et al.* (2001) [4] the antioxidant property of Glutathione can be attributed to the presence of a reducing agent i.e., thiol group of cysteine component which permits the oxidation and reduction of glutathione to a stable form and Agarwal and Allamaneni (2004) [11] was with an opinion that it also has got a role to play in the formation of glutathione disulphide which at the expense of NADPH and glutathione reductase as a catalyst can be reverted back to GSH. Its levels in the body are constantly regulated by its formation and the reactions are catalysed by the enzyme's gamma-GCS and glutathione synthetase [8, 10].

Mechanisms of redox cell signalling in oocyte and embryo metabolism

Redox reactions result in oxidized or reduced states due to the involvement of electron transfer [3] and this explains profound role of ET in the oocyte and embryo metabolism redox states. In the graffian follicles, the major sources of ROS are granulosa cells, neutrophils, macrophages and during the process of folliculogenesis, oocytes are guarded by the anti-oxidants like glutathione transferase, paraoxanase, catalase, heat shock protein (HSP) 27, SOD, and protein isomerase from the damage caused by oxidative stress [12]. According to Agarwal *et al*., 2008 this damage need not be always a direct process involving the reaction with the molecules or disrupting the cellular components but can also involve the signalling process their by bringing the negative outcomes or as stated by Irani (2000) [13] it can be involving the secondary messengers and targeting them either directly or indirectly. One of the major mechanisms of damage caused by ROS can be attributed to the expression of cytokines modulation and pro-inflammatory substrates, mediated by the AP-1, p53, and NF-kappa B activation which are redox-sensitive transcription factors. The deleterious effects can be mediated by any of the mechanism as shown in the hypothetical diagram (fig).

Pathways targeted by the Oxidative stress

The main signalling pathways that are negatively altered by the Oxidative stress are

i) Mitogen-activated protein kinases (MAPK) signalling pathways promotes the actions of receptor tyrosine kinases, protein tyrosine kinases, receptors of cytokines, and growth factors which is regulated by phosphorylation and dephosphorylation of serine and/or threonine residues [14, 15].
ii) c-Jun N-terminal kinases (JNK) which prevents phosphorylation because of its inhibition by GST and phosphorylation can be promoted by addition of H_2O_2 there by disturbing the complex [14, 15]. The JNK pathway by virtue of its inhibition via GST enzyme prevents phosphorylation and this cascade can be disrupted by promoting the phosphorylation by means of addition of H_2O_2 [16, 17].
iii) p38 pathways.
iv) Dissociate the ASK1–Trx complex [18] and
v) Calmodulin-dependent pathways [19, 20].

By means of negatively altering the above-mentioned cell signalling pathways the oxidative stress disrupts the homeostasis of the cell thus altering the growth of the cell and its proliferation.

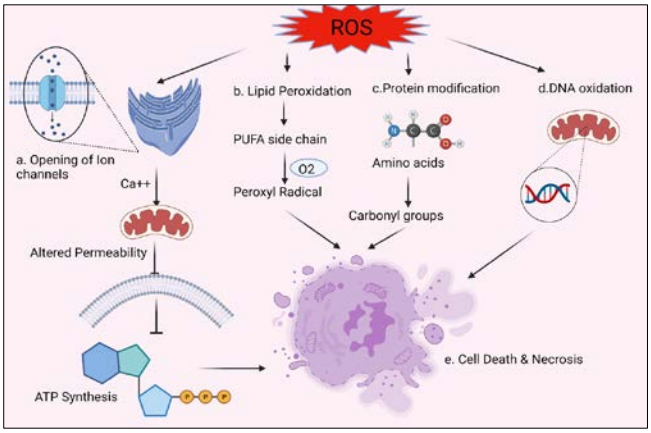

Fig 1: Hypothetical diagram depicting the deleterious effects of ROS. ROS can result in e) cell death & necrosis by a) opening of ion channels at the endoplasmic reticulum levels resulting in the release of Ca++ which in turn alters the permeability of Mitochondria inhibits the ATP synthesis b) lipid peroxidation of the PUFA side chain resulting in the peroxy radical formation c) protein modification resulting in the carbonyl groups formation d) Mitochondrial DNA oxidation [21]

References

1. Al-Gubory KH, Fowler PA, Garrel C. The roles of cellular reactive oxygen species, oxidative stress and antioxidants in pregnancy outcomes. International Journal of Biochemistry & Cell Biology. 2010;42(10):1634-50.
2. Burton GJ, Jauniaux E. Oxidative Stress. Best Practice & Research Clinical Obstetrics & Gynaecology. 2010;25:287-299.
3. Agarwal A, Aponte-Mellado A, Premkumar BJ, Shaman A, Gupta S. The effects of oxidative stress on female reproduction: A review. Reproductive Biology and Endocrinology. 2012, 10(49).
4. Behrman HR, Kodaman PH, Preston SL, Gao S. Oxidative stress and the ovary. Journal of the Society for Gynecologic Investigation. 2001;8:S40-S42.
5. Shkolnik K, Tadmor A, Ben-Dor S, Nevo N, Galiani D, Dekel N. Reactive oxygen species are indispensable in

ovulation. Proceedings of the National Academy of Sciences of the United States of America. 2011;108(4):1462-7.
6. Sugino N. Roles of reactive oxygen species in the corpus luteum. Animal Science Journal. 2006;77:556-565.
7. Perkins AV. Endogenous anti-oxidants in pregnancy and preeclampsia. Australian and New Zealand Journal of Obstetrics and Gynaecology. 2006;46:77-83.
8. Fujii J, Iuchi Y, Okada F. Fundamental roles of reactive oxygen species and protective mechanisms in the female reproductive system. Reproductive Biology and Endocrinology. 2005;3:43.
9. Maiorino M, Bosello V, Ursini F, Foresta C, Garolla A, Scapin M, *et al*. Genetic variations of gpx-4 and male infertility in humans. Biology of Reproduction. 2003;68:1134-1141.
10. Ruder EH, Hartman TJ, Goldman MB. Impact of oxidative stress on female fertility. Current Opinion in Obstetrics and Gynecology. 2009;21(3):219-222.
11. Agarwal A, Allamaneni SSR. Role of free radicals in female reproductive diseases and assisted reproduction. Reproductive Biology and Endocrinology. 2004;9:338-47.
12. Angelucci S, Ciavardelli D, Di Giuseppe F, Eleuterio E, Sulpizio M, Tiboni GM, *et al*. Endothelial dysfunction in heart failure. Pharmacological Reports. 2008;60:119-126.
13. Irani K. Oxidant signaling in vascular cell growth, death, and survival: A review of the roles of reactive oxygen species in smooth muscle and endothelial cell mitogenic and apoptotic signaling. Circulation Research. 2000;87:179-183.
14. Boutros T, Chevet E, Metrakos P. Mitogen-activated protein (MAP) kinase/MAP kinase phosphatase regulation: roles in cell growth, death, and cancer. Pharmacological Reviews. 2008;60(3):261-310
15. Brown MD, Sacks DB. Protein scaffolds in MAP kinase signalling. Cell Signal. 2009;21(4):462-469.
16. Kamata H, Honda S, Maeda S, Chang L, Hirata H, Karin M. Reactive oxygen species promote TN Falpha-induced death and sustained JNK activation by inhibiting MAP kinase phosphatases. Cell. 2005;120(5):649-61.
17. Nagai H, Noguchi T, Takeda K, Ichijo H. Pathophysiological roles of ASK1- MAP kinase signaling pathways. Journal of Biochemistry and Molecular Biology. 2007;40:1-6.
18. Matsuzawa A, Ichijo H. Redox control of cell fate by MAP kinase: physiological roles of ASK1-MAP kinase pathway in stress Signaling. Biochimica et Biophysica Acta. 2008, 1780
19. Droge W. Free radicals in the physiological control of cell function. Physiological Reviews. 2002;82:47-95.
20. Harvey AJ, Kind KL, Thompson JG. REDOX regulation of early embryo development. Reproduction. 2002;123:479-486.
21. Vinaya Sree C. Effect of L-Homoarginine on Follicular development in Ewes-Thesis, PVNRTVU, 2022.