Chapter

Inflammation and Ovulation

Pankaj Pant and Havagiray R. Chitme

Abstract

The ovulation is a complex physiological process which is very commonly affected in patients with PCOS. Understanding inflammatory process involved in ovulation is important with respect to its onset, diagnosis and treatment. There are multiple inflammatory factors are associated with ovulation however anovulation and contraception have not been therapeutically explored in context with inflammatory process. Therefore, this chapter is written to help readers to understand the basics of inflammation in ovulation and role of inflammatory mediators in ovulation. This chapter also describes genetic and molecular aspects linked to ovulation.

Keywords: inflammation, ovulation, cytokine, prostaglandin, TNF-alpha, PPAR-γ

1. Introduction

Mammalian ovulation is a fundamental physiological process involves the rupturing of follicle and releasing of the dominant follicle from the ovary into the fallopian tube where it has the potential to get fertilized if it exposed to sperm. Oocyte is covered up of four different layers namely the granulosa cells, which form a protected layer within oocyte and the extra follicular microenvironment, then theca layers of theca-interna and theca-externa, tunica albuginea and the outermost one is epithelium [1, 2]. A thin transparent layer between oocyte and follicular membrane is made up of secretions by the oocyte, termed as zona pellucida [2].

The developing oocyte enclosed in a ovarian follicle which is float in a dynamic fluid i.e. Follicular fluid (FF), contain variety of signaling molecules such as polysaccharides, hormones, cytokines, chemokines, growth factors, reactive oxygen species (ROS), metabolites, antioxidant enzymes, etc. The follicular fluid formed in developing antral follicles, primarily to support the development and protection of oocytes. These molecules are also acts as communicators between somatic and germ cells [3].

The duration of ovulatory process in humans, pigs, rats and rabbits takes approximately 40, 22, 12 and 10 hours respectively to complete. Substantial tissue remodeling occurs during the ovulation, the follicle increases its own size, and the layers of theca cells fuse with the tunica albuginea, resulting into thinner and permitting rupturing of follicle to release the oocyte [4]. The mature oocyte when released by the rupturing of follicle its uptake facilitated by the fingure like opening projections of fallopian tube which is known as fimbriae, if fertilization occur here then the blastocyst further transfer it towards the uterus where the pregnancy takes place [5].

Inflammation is defensive mechanism of the cells that is crucial to health and it is delineated as a local immune response of living vascularized tissues to endogenous and exogenous stimuli and its actions is to removal of injurious stimuli with

starting the healing process [6]. Inflammation is also initiated when the cells die from deficiency of nutrients or hypoxia, a condition that often is originated by the blood flow loss to the site. The chemical mediators of the inflammation generally originate from the blood plasma, platelets, white blood cells (monocytes, neutrophils, basophils, and macrophages), endothelial cells lining of the blood vessels, mast cells, and injured tissue cells. The chemical mediators responsible for the inflammation is histamine, that stimulates vasodilation and increases the vascular permeability, and lysosomal substances acting as vascular permeability enhancer which are secreted from neutrophils, and certain small proteins in the complement system, namely C3a and C5a. Various cytokines released by inflammatory cells also have vasoactive and chemotactic function. Many cells produce prostaglandins which linked to the fever and pain of inflammation; a group of fatty acids which involves in the augmentation of vascular permeability of the other substances, platelets aggregation; which is essential for coagulation [6].

The objective of this review is to understand and establish a relationship between how the inflammation as well as different mediators of inflammation that influence the ovulation process that is crucial for clinical management and prediction of gynecological complications for future study.

2. Inflammatory genetic mechanism

Inflammation is detected throughout many normal reproductive progressions, for the duration of ovulation, menstruation, implantation, as well as parturition. Ovulatory cycle is also considered as inflammatory process because the rupturing of dominant follicle undergoes the process of healing [7]. Throughout ovulation, the role of inflammation is very significant in terms of folliculogenesis and luteinization. In the course of the rupturing of follicle, there is significant surge of intra-follicular pressure which leads to weakening of follicle layer by the stimulation of gonadotropins resulted in inflammation [8]. The inflammation notably persuades in both ovulation and tissue remodeling phase which resulted in events of hyperemia, vasodilation, edema, collagenolysis, and proliferation of cells [1].

Wissing et al. isolated differentially expressed 1186 genes in human granulosa cells (GC) before and 36 h after the administration of hCG, besides 572 genes found to be up-regulated which represented angiogenesis, inflammation, extracellular matrix and growth factors and 614 genes down-regulated which denoted cell cycle and about 72 genes which has been earlier establish linked with ovarian cancer. H19/mir675, CD24, CLDN11, ANKRD22, and FBXO32 adds as new ovulation related genes and PTGS2, an inflammatory gene heavily up-regulated [5].

3. Inflammatory oxygen species and ovulation

The release of mature oocyte relies on the expansion of cumulus oocyte complex (COC), where the reactive oxygen species (ROS) function as critical modulator of inflammatory reaction. Residual growing follicles promoted to apoptosis by the ROS [8]. Simultaneously estrogen synthesis started with the influx of catalase and Glutathione (GSH) i.e. a non-enzymatic antioxidant species exists in oocytes and embryos, in growing residual follicles for the maintenance of normal ovarian function and counter the apoptotic process. The luteal phase begins with progesterone production for the maintenance of preliminary stage of pregnancy, if fertilization did not occur then degeneration of corpus luteum starts [2]. For the

induction of cell proliferation, maturation, cellular-differentiation and ovulation the physiological level of ROS is very important [9]. Augmented ROS may outcomes to DNA damage, activation of signaling cascades, and epigenetic alterations [10]. Inflammation in the course of ovulation is also responsible for the oxidative stress, damaging of DNA and moreover in the neoplastic ovarian surface epithelium (OSE) cells transformation [11].

4. PCOS and inflammation

Elevated level of inflammation also involves in the pathogenicity of many reproductive disorder such as polycystic ovary syndrome (PCOS), characterized by biochemical hyperandrogenemia, chronic anovulation, and polycystic ovaries. About 5 to 15% of reproductive age woman are suffering with this disorder in the world. It is anticipated that the metabolic disorders associated with PCOS and the pathogenesis of PCOS due to systemic inflammation as well as dysfunctioning of mitochondria. About 33% adolescent PCOS girls are more prone to metabolic disorder are obese, which is 3 to 5 folds higher if compared with same age healthy girls and body mass index (BMI) [9]. Studies reported and found the significant increased level of monocytes, lymphocytes, CRP, interleukins IL-1, IL-6, IL-18, pro-inflammatory cytokines and TNF- α in addition to increased production of advanced oxidation protein, protein carbonylation and lipid peroxidation in PCOS patients compared with same reproductive aged healthy persons [9]. The PCOS patients suffered with chronic inflammation [12, 13]. Along with the deficiency of antioxidant i.e. Vitamin C, Vitamin E and Superoxide dismutases (SOD), this leads to cause inflammatory milieu and risk to develop the obesity, type-1 diabetes, insulin resistance, hyperandrogenism, and cardiovascular ailment [1, 14].

5. Anti-inflammatory agents and ovulation

The surge of luteinizing hormone (LH) stimulate the production of cyclic adenosine monophosphate (cAMP), steroidal hormones, histamine discharge and various mediators of inflammation e.g. prostaglandins, bradykinins, C-reactive protein (CRP), Proinflammatory cytokines, etc. [1]. Christina et al. study states that the successful folliculogenesis, oocyte maturation, and ovulation require a healthy inflammatory response.

There are many findings illustrate that the importance of untroubled inflammatory response for proper folliculogenesis and ovulation, if it altered, may contribute to oocyte quality concern and reproductive dysfunctions such as anovulation, infertility, menstrual irregularities, etc. [5, 8]. It is reported that low dose of Aspirin taken by the patients suffering with higher systemic inflammation were able to reestablish the pregnancy [4, 12].

6. Correlation between hormone and inflammation

The level of LH also positive correlation with release of prostaglandins and eicosanoids that are the source to trigger the fibroblasts, promotes the angiogenesis and hyperemia, collagenase activation, release of proteolytic enzymes, some of which degrade the follicular connective tissue resulting ovulation, and cause the inflammation. The gene hyaluronan (HA) synthase-2 (Has2) associated with COC matrix formation. Bradykinin play a key role in vasodilation which appears to be 10

folds increased during ovulation. The serum C- reactive protein (CRP), a marker of inflammation also raised to stimulate the production of interlukin-6 from macrophages, tumor necrosis factor α (TNFα) and the competent system of inflammatory response further activated by the adipocytes [13].

7. Role of proinflammatory cytokines

The role of proinflammatory cytokines is also important throughout folliculogenesis and induction of ovulation [5]. Higher level of follicular TNF-α resulted in the poor quality of oocyte which compromised with the fertility, also the elevated level of interleukin (IL-6) associated with less chances of conceiving while the another interleukin (IL-1) found to be regulated by FSH and its higher follicular level has been resulted in the higher chance on embryo implantation [7, 15].

8. Nucleotide leukin rich polypeptide -3 inflammasomes

A recent finding added a new mechanism for ovulatory process regulation, suggested that the NLRP3 activation of nucleotide leukin rich polypeptide 3 (NLRP3) inflammasomes started before the ovulation lasting completion of ovulation. They induces the follicular development by 52 hours' treatment using Pregnant mare serum gonadotropin (PMSG). It was found that the expression of NLRP3 inflammasomes and adaptor protein apoptosis-associated speck-like protein (ASC) significantly increased, and it was appeared a dramatic surge in caspase-1 activity and production of IL-1β [16].

9. Gonadotropin in inflammation

Gonadotropin surge trigger the ovlation with the parllel stimulation of two gens of preovulatory follicles in granulosa cells, prostaglandin-endoperoxide synthase 2 (PTGS2) and progesterone receptor (PGR). Secretion of LH stimulates the induction of both PTGS2 and PGR in preovulatory granulosa cells. Expression of PTGS2 stimulates inflammation by releasing pro-inflammatory prostaglandins wheras anti-inflammatory action through the PGR by the supression of pro-inflamatory genes or thru the stimulation of antiinflammatory genes. Higher level of PGE2 and PTGS2 are associated with the ovarian disorders such as ovarian carcinoma, ovarian hyperstimulation syndrome (OHSS) as well as polycystic ovarian syndrome (PCOS) [11].

10. Inflammatory prostaglandins

Prostaglandins (PGs) are signaling molecules derived from dietary fats with clinically relevant roles in reproductive biology. PGE2, for instance, promotes ovulation downstream of the luteinizing hormone surge. Excess consumption of nonsteroidal anti-inflammatory drugs, which inhibit prostaglandin-endoperoxide synthase (cyclooxygenase or Cox), is associated with reversible female infertility, likely due to failed ovulation. On the other hand, proinflammatory cytokines increase PGF2α associated with corpus luteum development and immune cell recruitment [17].

11. Adipokines

A study (Bongrani et, al.) based on the adipokines roles in the pathophysiology of PCOS, they analyzed the adipokines profile in the normal-weight PCOS patients and obese women with PCOS and comparison of these with the women whose only have a Polycystic ovary morphology. Whereas they found the PCOS patient reported with lower adiponectin level in serum as well as FF, and also the lower expression in adipose tissue of AdipoR1 and AdipoR2. In granulosa cells AdipoR1 expression was positively correlated with the follicular numbers, oocytes count and embryos, on the other hand there was no significant difference in AdipoR2 reporters was found. No correlation was established among the FF adiponectin concentration and expression of its receptor, AdipoR1/AdipoR2 in GCs. Dysregulation of adiponectin may be likely mechanisms which could be responsible for impairment of insulin-sensitivity in PCOS patient, and it seems to be independent of insulin resistance severity and a potential role of adiponectin in folliculogenesis.

12. Omentin

The concentration of Omentin in FF was found to be positively correlated with BMI, higher in obese patients compared to the normal weight patients. They also point out that the omentin may possibly be controlled by means of inflammation, because the expression of omentin altered in inflammatory conditions [18].

13. Oxidative stress

ROS is necessary to maintain the normal female reproductive physiology, it is involved in the oocyte maturation, corpus luteum apoptosis as well as embryonic development process. The release of mature oocyte depends on the expansion of cumulus oocyte complex (COC), where the reactive oxygen species (ROS) function as critical modulator of inflammatory reaction. The exposure of ROS may lead to under transformative alterations of epithelial cells in the ovary and fallopian tubes [10].

14. Tumor necrosis factor-α

It has a significant role in the process of ovulation as well as to excrete out the damaged corpous luteum from the ovarian tissue. It works by the ligand gated receptors, TNFR-I and TNFR-II [10, 19]. It is also linked with the various pathological conditions when its level elevated. It is also reported that the infertile women with PCOS reported to have higher free fatty acids and blood serum level of TNF-α when compared with the healthy patients. Oxidative stressed cells also found to release higher levels of TNF-α than the normal ovarian epithelial cells which results in an autocrine surge of TNF-α mRNA as well as in the form of expression in other pro-inflammatory cytokines, chemokines, and angiogenic factors [10, 20, 21].

15. Interleukins

Interleukin-15 is an important interleukin which is negatively associated with the oocyte maturation. It belongs to the cytokines family having four α-helix

bundle, i.e. pleiotropic glycoprotein. It was reported to be higher in women with an unsuccessful assisted reproductive techniques outcomes (median value 1.4 pg./ml) than of the women those succeeded the clinical pregnancy (median value 0.8 pg./ml) [22]. The IL-6 is a regulator of cumulus cell-oocyte complex (COC) expansion and responsible for the quality of murine oocyte during the in-vitro fertilization.

16. Matrix metalloproteinases

The another factor which has been found to be involved in follicular development as well as in the ovulation process are Matrix metalloproteinases (MMPs). The matrix metalloproteinases activities regulated by the specific tissue inhibitors called metalloproteinases (TIMPs) and endogenous inhibitors. The balance of both these is very essential for their activity and to maintain the normal ovarian physiology. It is well noticed that the MMP-2 and MMP-9 level increased during and before the 3 hours of ovulation if compared to 20–22 hour before the ovulation. Augmented level of MMP-9 also postulated in the PCOS pathophysiology and is also linked with progression and etiological to many other ailments such as cystic fibrosis, asthma, ulcerative colitis, cardiovascular disorders, atherosclerosis, etc. [23, 24].

17. Peroxisome proliferator-activated receptor gamma

The Peroxisome Proliferator-Activated Receptor Gamma (PPAR-γ) is established to have an ability to prevent the expression of various signaling molecules also regulates the levels of prostaglandins through regulation of cyclooxygenase-2 and differentiation of immune cells especially those which are a part of inflammation. Thereby controls ovarian function, fertilization, and ovulation. Its proinflammatory activity is linked to the formation of prostaglandin E by downregulation of COX-2 mRNA of granulosa cells whereas it is upregulated twice in PCOS [25]. It is also being proven that the PPAR-γ controls genes responsible for expression of TNF-alpha and Interleukins along with others. The study is supported by use of PPAR-γ agonist which affected the functioning of ovaries by involving signal transduction of insulin and IGF [20].

18. Chemokines

Earlier study reported the expression of chemokine receptor-2 (C-C motif) (CCR2) in the human ovarian cumulus-oocyte complexes, theca cells, preovulatory follicles and in feline ovarian follicle walls. This receptor is believed to be involved in folliculogenesis and determines the reproductive lifespan of female [25] (**Figure 1**).

19. Conclusions

Inflammation is believed to be involved in triggering the process of ovulation. There are several factors, genes, receptors, proinflammatory mediators playing important but diverse role in ovulation. The extent of their role and intensity of reaction induced by them required to be studied to understand their clinical applicability.

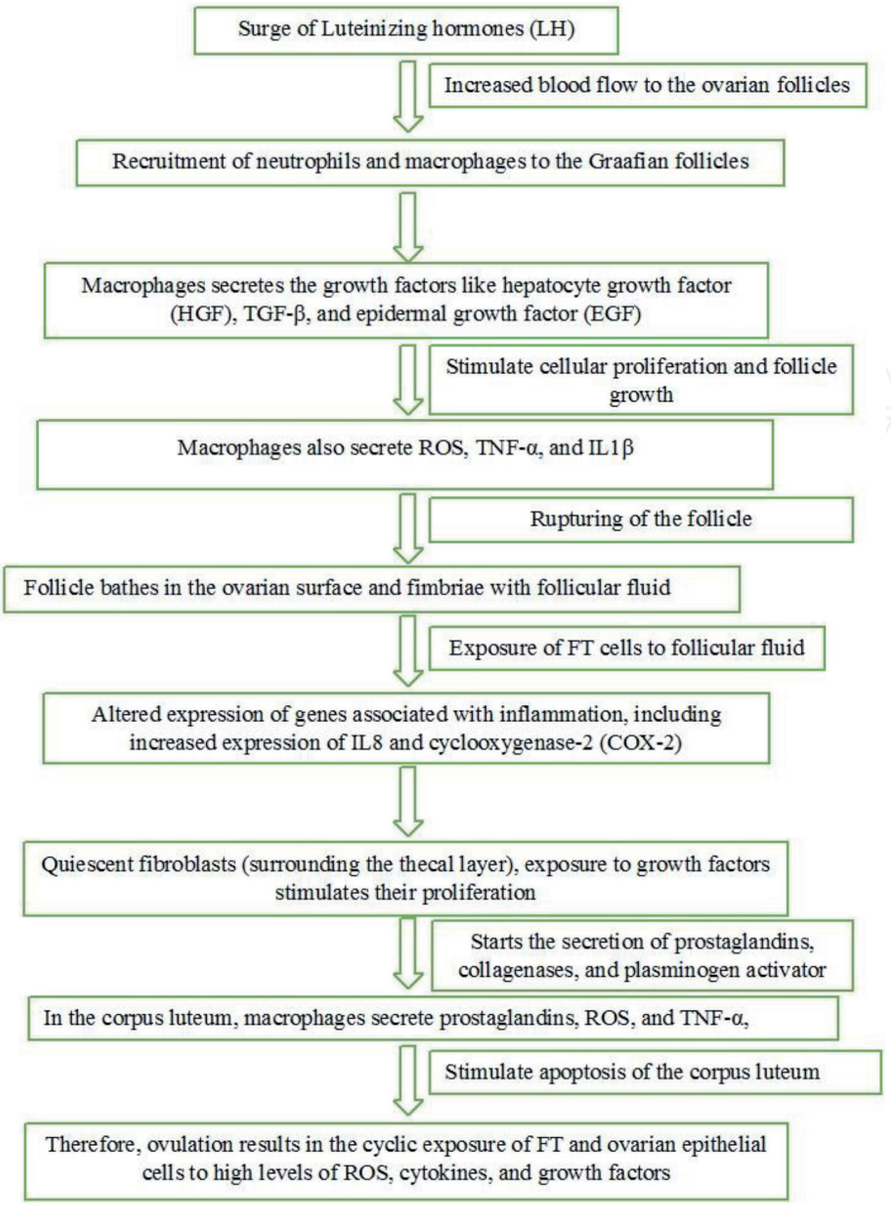

Figure 1.
Inflammatory process in ovulation.

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Conflict of interest

The authors declare no conflict of interest.

Author details

Pankaj Pant and Havagiray R. Chitme*
Faculty of Pharmacy, DIT University, Dehradun, Uttarakhand, India

*Address all correspondence to: hrchitme@gmail.com

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