Potential role of Kisspeptin in infertility

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Abstract. Kisspeptin is a neuropeptide that plays a central role in fertility and neuroendocrine regulation of the hypothalamic-pituitary-gonadal axis. It has also been shown to act at the peripheral level in both men and women. Many studies have shown a correlation between kisspeptin blood levels and fertility in men. It is also involved in the maturation of sperm and even in the implantation of a pregnancy in women. In men, dysregulation of kisspeptin signaling can lead to hypogonadotropic hypogonadism. Recent studies have shown that Kisspeptin could constitute a new therapeutic target in the treatment of fertility disorders. Others have shown that the administration of exogenous Kisspeptin stimulates the release of gonadotropins in patients with fertility problems and even in healthy subjects. In addition, it plays an essential role in improving the quality of sperm in medically assisted procreation and even in the maturation of oocytes. In this literature review, we aim to examine the main functions of kisspeptin in male and female infertility.

Keywords: Kisspeptin, fertility, hypogonadism, gonadotropin-releasing hormone, sperm motility.

1 Introduction

Kisspeptin is a recognized regulator of the onset of puberty, sexual maturation, and adult reproductive activity. It is a 54 amino acid peptide, of which the KISS-1 gene is responsible for the activation of the G protein-associated receptor GPR54. In melanoma cell lines, it was first identified in 1996 as an inhibitor of metastasis [1]. It is a series of structurally similar peptides originating from the differential synthesis of preprokipeptin, a common pioneer. Kisspeptin peptides are part of a class of RF-amino (arginine (R) and amidated phenylalanine (F)) peptides, i.e., neuroactive peptides that exhibit a specific Arg-Phe-NH2 motif [2]. Kisspeptin-54 is the most common kisspeptin in the human body, which can then be converted into various peptides containing 14, 13 and 10 amino acids [3]. In the hypothalamus, there is a receptor called Kiss1R, which is connected to G proteins of a typical subgroup of G protein-related receptors.

The GPR54 receptor was cloned by RT-PCR in 1999 from rat brains [4]. It is a seven-domain transmembrane receptor with a sequence analogy to the galanin receptor. GPR54 messenger RNAs are expressed in many tissues (brain, pituitary gland, pancreas, kidney, testes, etc.), but, as for kisspeptin, the highest expression was found in the placenta [5].

Gonadotropin-releasing hormone (GnRH) is an essential component of hypothalamic regulation in the human reproductive system. Pituitary secretion of gonadotropins is regulated by GnRH, which also impacts gonadal steroid feedback and influences fertility. Since the hypothalamus produces GnRH, it mainly works in the pituitary gland.

Studies have shown that GnRH is regulated by Kisspeptin (KISS1) [6].

As an upstream regulator of the sudden, pulsatile release of GnRH, Kisspeptin has been widely praised over the past decade for its fundamental role in the regulation of reproduction [7].

According to several researches, kisspeptin has an effect on the hypothalamus, leading to the production of gonadotropins (luteinizing hormone (LH), follicle-stimulating hormone (FSH)) and downstream sex hormones (testosterone and estradiol) [8]. The first experiment called “Kisspeptin in human” took place in 2005 and confirmed that kisspeptin could stimulate female hormones [5]. Various reported reproductive

diseases are caused by disruption of the central KISS1/KISS1R system.
Mutations that inhibit KISS1R result in hypogonadotropic hypogonadism, while mutations that activate KISS1R result in precocious puberty [9].
It is undeniable that kisspeptin plays a crucial role in stimulating hypothalamic GnRH secretion.
Given all the available information regarding the essential role of Kisspeptin in stimulating GnRH secretion at the hypothalamus, it is also important to take into account the peripheral part of Kisspeptin in the tissues (testis, ovary, etc.). Recent research has revealed the possible functions of this peptide. during germ cell formation, regulation of sperm function and sperm production in the testes [10].
In this literature review, we focus on the localization and structure of kisspeptin and its receptors. We will also summarize recent advances in the roles of kisspeptin on female and male fertility as well as its therapeutic applications.

2 Localization and structure of Kisspeptin and its receptor

Kisspeptin and its receptor are located in the central and peripheral nervous systems [11]. At the core level, the Kiss1/KISS1R pair has been identified in the hypothalamus, while at the peripheral level, it is found in the placenta, testes, ovaries, pancreas, and small intestine. Kisspeptins are part of a family of RF peptide hormones, hence their name, because they contain arginine-phenylalanine residues at the amino terminus (Arg-the-NH2) [4**].**

In humans, there are various structures of Kisspeptin. All are produced from the cleavage of a common precursor known as pre-pro-kisspeptin. Prepro-kisspeptin is a peptide composed of 145 amino acids, with a molecular weight of 15.6 kDa (kilo dalton = kg/mol). Then, it is divided into four subsequently lower molecular weight peptides, namely KP-54, KP-14, KP-13, and KP-10 (see Figure 1) [8]. The number of amino acids in the long isoform varies from species to species; in rats, it comprises 52 amino acids [12].
All possess the RF-amide motif that can bind and activate Kiss1R (14). The kisspeptin most present in human circulation is kisspeptin 54 (KP-54) [8].

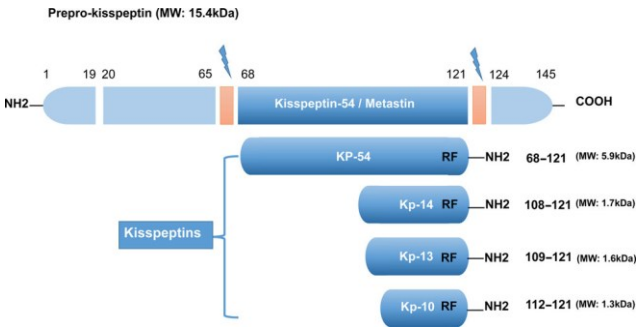

Fig. 1. Structure of kisspeptin [12].

3 Kisspeptins and gonadotropic hormone secretion

Kisspeptin has been shown to stimulate LH secretion in rats and mice in several studies. This hypothesis has been verified in other species, such as mammals, sheep, monkeys, humans, and cows [13]. Two regions of the hypothalamus house kisspeptin action neurons: the anteroventral periventricular nucleus (AVPV) and the arcuate nucleus (ARC). KP neurons in the infundibular nucleus/ARC are activated by a low level of estrogen synthesized from the ovary, resulting in pulsatile secretion of gonadotropins GnRH and GPN. In contrast, increased estrogen levels during the follicular phase stimulate KP neurons in the PeN/AVPV and block infundibular/ARC neurons that produce the GnRH/GPN surge, which promotes oocyte maturation and ovulation [14, 15].

GPR54 in the hypothalamus is activated by kisspeptin, resulting in the release of GnRH, but this action is inhibited by the administration of a GnRH antagonist [12]. Research has demonstrated that both central and peripheral administration of kisspeptin increases blood levels of the luteinizing hormone LH [16]. In humans, the teams of Dhillo et al. and Pinilla et al. showed that intravenous or subcutaneous Kisspeptin (Kp-54) could stimulate the secretion of LH and FSH in men and women [5]. Accumulating evidence has indicated that Kisspeptin stimulates the hypothalamus' release of gonadotropin-releasing hormone (GnRH). This then triggers the production of follicle stimulating hormone (FSH), luteinizing hormone (LH) and the secretion of testosterone. Research conducted by Talib, HA et al. underscores this phenomenon, revealing reduced levels of LH, FSH, testosterone, and Kisspeptin in infertile patients relative to their fertile counterparts [17].

4 Kisspeptin and women's fertility

Kisspeptin plays an essential role in female reproduction, playing a role in gonadotropin production, puberty, ovulation, and metabolic regulation of fertility.

4.1 Function on ovaries (Kisspeptin and human ovary)

While it is becoming increasingly clear that the central effects of Kisspeptin are visible, there is still the possibility of direct gonadal effects. In rat ovaries, Terao and his colleagues initially found the expression of genes responsible for Kisspeptin and its receptor in 2004, a discovery validated by their research group [18] by revealing the presence of Kisspeptin and its receptor in the ovaries of primates and humans as well. According to research by Gayan F and colleagues, it was shown that in kisspeptin receptor knockout (KO) and haploinsufficient mice exhibited premature ovarian failure (POF), which correlated with low levels of expression of the ovarian kisspeptin receptor [14]. This is functional proof of a direct effect of Kisspeptin on the

ovaries, without dependence on its central products by gonadotropin.
Ali Abbara, Channa N. Jayasena, and their colleagues explored how kisspeptin 54 impacts the induction of oocyte maturation in women undergoing in vitro fertilization (IVF) who are at a heightened risk of having ovarian hyperstimulation syndrome (OHSS)
[19]. Several concentrations of Kisspeptin (3.2, 6.4, 9.6, or 12.8 nmol/kg) were taken. The rate of oocyte maturation was lower in the group receiving 3.2 nmol/kg than in the group receiving 6.4 nmol/kg, and similarly in the group receiving 9.6 nmol/kg than in the group receiving 12. 8 nmol/kg. The 60 women who received Kisspeptin experienced oocyte maturation in 54 of them. According to the results, the dose of 12.8 nmol/kg produced 121% more mature oocytes than the others. After oocyte maturation, ICSI was performed, but fertilization failed in three women. Following ICSI, they carried out the embryo transfer. Pregnancy tests were carried out on the women to see their progress. Following administration of 9.6 nmol/kg kisspeptin-54, the highest pregnancy rates were observed (85%, 77%, and 62%, respectively), with an implantation rate of 57.7%. OHSS cases were not documented in this investigation. Kisspeptin may be a viable and secure way to encourage oocyte maturation in IVF patients, particularly in those who are more susceptible to OHSS. [20].

4.2 Role of kisspeptin during embryo implantation and pregnancy

According to numerous studies, kisspeptin plays a regulatory role in the invasion of trophoblasts during embryo implantation in the early stages of pregnancy [3,17]. The study by Janneau et al. The team reported no significant difference between kisspeptin expression in women at term and early pregnancy [21]. Using the same research approach, the groups of Torricelli et al. and Horikoshi et al. respectively showed that the expression of kisspeptin in the placenta is higher during a premature pregnancy than during a term pregnancy. At the same time, they observed a decline in KISS1 expression as placental maturation progressed [22]. Alternatively, kisspeptin levels experience a substantial increase throughout pregnancy, increasing 900-fold during the first trimester and skyrocketing to a staggering 7,000-fold increase in the third trimester compared to levels seen in non-pregnant women [23].

Furthermore, subsequent studies showed that the concentration of kisspeptin in the placenta was higher at the beginning of pregnancy than at the end of it [24].

Kisspeptin levels in plasma or serum may indicate placental function and, consequently, the viability of a pregnancy. Recent research increasingly indicates that the levels of kisspeptin in serum or plasma, specifically kisspeptin-54, 14, and 10, serve as accessible markers for distinguishing between confirmed miscarriage and ongoing intrauterine pregnancy at the time of diagnosis [21.17]. The results of these studies demonstrated a notable disparity in kisspeptin levels between the miscarriage and intrauterine pregnancy groups.
Nowadays, miscarriages constitute 20% of pregnancy problems [15]. According to research, Kisspeptin has been shown to have the ability to control embryo implantation by altering endometrial function and improving stromal cell decidualization [3,11,19]. One of the roles of Kisspeptin in implantation during pregnancy is to regulate the infiltration of natural killer (NK) cells. Park, D.-W et al. According to the researchers, Expression of kisspeptin in trophoblast cells from pregnancies experiencing recurrent spontaneous abortions was observed to be diminished and correlated with the levels of peripheral and decidual natural killer (NK) cells [25]. Additionally, an interesting finding in several women with idiopathic infertility who underwent IVF shows a lower concentration of Kisspeptin, associated with minor embryo implantation ability [22,25].
These studies show that low concentrations of kisspeptin may be associated with greater miscarriages.

5 Kisspeptin and male fertility (testicular function and spermatozoa)

Human plasma has been detected with the presence of kisspeptin and measured in different health disorders. Kisspeptin levels are significantly higher in fertile men than in those who are infertile [26]. In instances of hypogonadotropic hypogonadism, elevated blood levels of kisspeptin have been reported. Nevertheless, administration of GnRH leads to a decrease in circulating kisspeptin levels, attributed to the restoration of sex steroid feedback mechanisms at the hypothalamic level. Despite gonadotropin stimulation, testosterone biosynthesis and spermatogenesis are not consistently restored in clinical cases of inactivating KISS1R mutations, implying the necessity of testicular Kiss1R signaling for steroidogenesis. The impact of Kisspeptin on Testicular Function during Pregnancy.
The testes are glands that produce male gametes, and sperm respectively. Male germ cells (spermatogonia) are converted into mature sperm during spermatogenesis, which occurs through cell division and differentiation. The process of spermatogenesis in men takes place in the seminiferous tubules, more precisely in the Sertoli cell cavities. Several factors and hormones regulate spermatogenesis, such as testosterone, a regulatory and supporting hormone for sperm production, produced by Leydig cells, and the hypothalamic-pituitary axis, of which GnRH secreted by the hypothalamus stimulates spermatogenesis. The hormones LH and FSH form the pituitary gland, which influences the testicles. When FSH is produced, it affects Sertoli cell function and spermatogenesis, while LH affects Leydig cells and testosterone production. Sertoli cells produce testosterone and inhibin B which regulate the production of LH and FSH. Impairment of these mechanisms can lead to infertility in men.
A study in mice showed that Kisspeptin and its receptor are localized in sperm collected from the rat epididymis [27].

It can be said that Kisspeptin is involved in sperm maturation [21].
In a pioneering Chinese study [28], it was demonstrated for the first time that overall kisspeptin levels exhibited a positive correlation with sperm concentration, count, and motility. They compared blood levels of kisspeptin to seminal levels in infertile and healthy subjects. The aim was to see if there was a correlation between the two. After administration, they found that seminal kisspeptin levels were higher than blood kisspeptin levels. Several studies have suggested that the testes may serve as a significant contributor of Kisspeptin in the bloodstream [7,13,24]. This hypothesis was supported by the study of [29], who performed a gonadectomy on mice and observed a reduction in blood levels of kisspeptin.
These studies showed a positive effect of Kisspeptin on testicular function, but other studies also showed an inhibitory effect. [27,9,2]. Extended administration of Kisspeptin (over 30 days) induced testicular degeneration in male rats, characterized by reductions in testicular weight and seminiferous tubules, along with decreases in inhibin B and testosterone levels [15]. This may be due to desensitization of the hypothalamic-pituitary axis, as chronic use of Kisspeptin can cause desensitization at the GPR54 receptor (KISS1R).

5.1 Effect of Kisspeptin on sperm function

Research conducted in humans has revealed the expression of the kisspeptin gene and its receptor in sperm. They were located in the sperm head (post-acrosomal region), in the middle piece, and at the flagellum [28]. This suggests that Kisspeptin and its receptor Kiss1R play a direct role in male gamete infertility. Kisspeptin 13 at a dose of 1 µM triggers a progressive increase in sperm intracellular calcium and a transient hyperactivation of sperm. Studies by Rehman and colleagues found that kisspeptin levels were higher in men with normal spermatogenesis compared to those with oligospermia or asthenospermia [28,30]. Recent surveys increasingly confirm these hypotheses. In 2019, P. Wang, Le Zou, and colleagues examined kisspeptin levels in the semen and serum of 666 student volunteers and found links to sperm quality taking into account factors such as age, BMI, smoking, and immobilization time [31]. Kisspeptin concentrations in seminal plasma were observed to be higher than those in blood plasma. A positive relationship was found between the overall concentration of kisspeptin in seminal fluid and three sperm parameters: sperm concentration, total sperm count, and total motility.

Numerous facets of sperm physiology, such as motility, capacitation, and the acrosomal response, rely on the mobilization of calcium from intracellular reservoirs. Some research on the action of kisspeptin in sperm physiology has highlighted a specific role in calcium mobilization. Kp-10 causes intracellular calcium mobilization in mouse sperm, while Kp-13 impacts progressive sperm motility and transient hyperactivation, but not the acrosomal reaction in humans. Calcium plays an important role in sperm mobilization, such as motility, capacitation, and acrosomal reaction. In the literature, several studies have shown that Kisspeptin induces calcium mobilization in sperm, leading to progressive motility and transient hyperactivation.

6 Therapeutic applications of Kisspeptin fertility (testicular function and spermatozoa)

Presently, Kisspeptin holds promise as a therapeutic option for addressing fertility disorders. Globally, infertility affects one in six individuals, and existing hormonal and surgical treatments often entail notable side effects and can have high failure rates. Numerous investigations have demonstrated that administering kisspeptin infusion notably boosts plasma levels of LH, FSH, and testosterone. Dhillo and colleagues published in 2008. This hypothesis was supported: after intravenous infusion of Kisspeptin in healthy men, they found an increase in plasma levels of gonadotropins and testosterone [1]. In 2007, a similar team found increased plasma LH levels in healthy premenopausal women after a subcutaneous injection of Kisspeptin. It is important to remember that Kisspeptin has a very short shelf life. Once in the blood, many proteases break it down quickly and its metabolites are very small.

To explore the impacts of extended stimulation, women with hypothalamic amenorrhea were subjected to repeated injections of KP54 twice daily for two weeks. Following these injections, there was an initial rise in LH and FSH levels; however, this effect waned after two weeks. Then, in the same cohort, twice-weekly administration of KP54 gave a satisfactory result since they observed a response over eight weeks [5].
A recent study also showed that incubating sperm with Kisspeptin for 15 minutes could improve sperm quality [32]. This suggests that administering Kisspeptin to sperm could significantly increase sperm motility.

Conclusion

Kisspeptin could be a new biomarker for the diagnosis of fertility disorders. Involved in gonadal maturation, it acts on hormonal secretion by stimulating the secretion of LH and FSH. It could be used as an additive to improve sperm quality in medical reproductive techniques.

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