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Role of *Cannabis sativa* on Male Reproduction, with Special Reference to Marijuana

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Abstract: The plant whose dried leaves, flowers, stems, and seeds are referred to as marijuana is *Cannabis sativa*, sometimes referred to as hemp. A few of cannabis' active components, such as delta-9-tetrahydrocannabinol (THC), cannabinol (CBN), and cannabidiol (CBD), have been related to the intoxicating effects of the plant. Marijuana is one of the medications that is most commonly abused globally. This study seeks to dispel several widespread myths about marijuana. The Indian hemp plant is the source of marijuana, and the component that contains the "drug" is mostly found in the flowers. Around the world, cannabis is a widely used recreational substance. Numerous studies have linked marijuana use to deregulation of the HPG axis, and in particular, a decrease in a vital hormone like LH, which in turn can alter testosterone and spermatogenesis. Using both the cannabinoid and vanilloid receptors, it appears that marijuana can actually alter sperm quality and semen parameters. Since marijuana appears to affect erectile function, it has also been related to sexual health. More clinical research must be conducted to more thoroughly explore the consequences of marijuana use in light of the legalization and decriminalization of marijuana use, as well as the fact that some studies have produced contradicting and contrasting results. All of the aforementioned studies highlight the need for clinicians to ask about marijuana use while assessing male infertility. When recommending medical marijuana, doctors should absolutely keep in mind the link to and potential effects on male fertility.

Keywords: Fertility, *Cannabis*, Sperm, Fertility, Testosterone, Marijuana

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Introduction

Cannabis sativa, often known as hemp, is the plant whose dried leaves, flowers, stems, and seeds are referred to as marijuana. The intoxicating effects of cannabis have been linked to a small number of its active ingredients, including delta-9-tetrahydrocannabinol (THC), cannabinol (CBN), and cannabidiol (CBD) (Murray *et al*., 2007; Sagar *et al*., 2018; Tendon *et al*., 2019). The plant contains several closely similar chemicals as well as the mind-altering substance delta-9-tetrahydro-cannabinol (THC). The cannabis plant can also be used to produce extracts with significant THC cannabinol (THC). The cannabis plant can also be used to produce extracts with significant THC

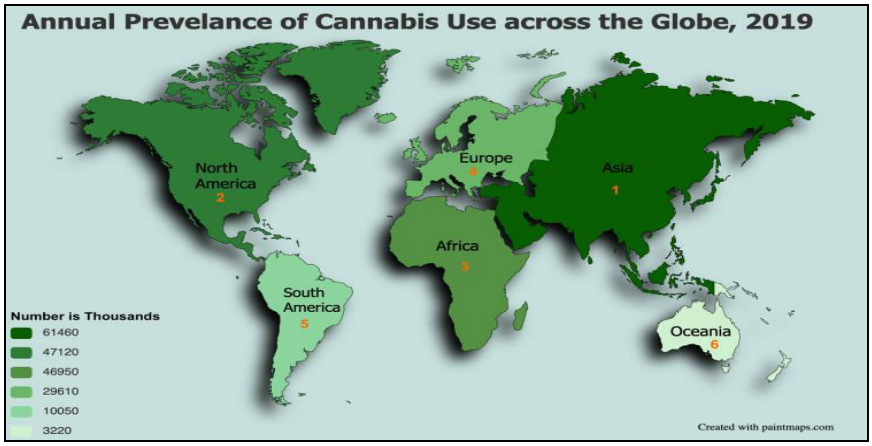

Fig. 1: Cannabis consumption worldwide annually prevalent in 2019. (Source: world drug report)

concentrations (Ahrens *et al*., 2009). One of the drugs that is most frequently abused worldwide is marijuana (Harmon *et al*., 1972). This article aims to dispel some common misconceptions regarding marijuana. Marijuana comes from the Indian hemp plant, and the part that contains the "drug" is found primarily in the flowers (commonly called the "buds") and much less in the seeds, leaves, and stems of the plant (Erowid, 2016). Cannabis is a popular recreational drug around the world **(**Fig. 1), only behind alcohol, caffeine, and tobacco. In the U.S. alone, it is believed that over 100 million Americans have tried cannabis, with 25 million Americans having used it within the past year. As a drug it usually comes in the form of dried infructescences, resin (hashish), or various extracts collectively known as hash oil (Erowid, 2016). THC creates the mind-altering effects that classifies marijuana as a "drug" (Cates *et al*., 1977).

In 2014 there were an estimated 182.5 million cannabis users worldwide (3.8% of the global population aged 15–64). This percentage did not change significantly between 1998 and 2014 (UNO, 2016). Studies show that marijuana usage can be linked to several adverse health consequences in a multi-system pattern (Hall *et al*., 2009).

Survey in India:

In India, according to a nationwide survey, 31 million people (2.8% of the total population) reported using cannabis in 2018, and 0.25% (2.5 million) also showed signs of cannabis dependence (Ambekar *et al*., 2019). In India, Marijuana is banned except for medicinal purposes, but smugglers and peddlers distribute it specially Punjab, Nagpur, Manipur and in many cities.

(i) *Overview of harmful effects caused by the use of marijuana*:

The immediate effects of taking marijuana include rapid heartbeat, disorientation, lack of physical coordination, often followed by depression or sleepiness. Some users suffer panic attacks or anxiety (Hall *et al*., 2009). But the problem does not end there. According to scientific studies, the active ingredient in cannabis, THC, remains in the body for weeks or even months. Marijuana smoke contains 50% to 70% more cancer-causing substances than tobacco smoke. One major research study reported that a single cannabis joint could cause as much damage to the lungs as up to five regular cigarettes smoked one after another. Long-time joint smokers often suffer from bronchitis, an inflammation of the respiratory tract (Osborne *et al*., 2008). The drug can affect more than physical health (Ugboma *et al*., 2012). Studies in Australia in 2008 linked years of heavy marijuana use to brain abnormalities. This is backed up by earlier research on the long-term effects of marijuana, which indicate changes in the brain similar to

those caused by long-term abuse of other major drugs. A number of studies have shown a connection between continued marijuana use and psychosis. Marijuana can change the structure of sperm cells, deforming them (Mirra *et al*., 2021). Thus even small amounts of marijuana can cause temporary sterility in men. Marijuana use can also disrupt a woman's menstrual cycle (Smith *et al*., 1983). Studies show that the mental functions of people who have smoked a lot of marijuana tend to be diminished (Chakravarty *et al*., 1975). The THC in cannabis disrupts nerve cells in the brain, affecting memory. Cannabis is one of the few drugs that causes abnormal cell division, which leads to severe hereditary defects. A pregnant woman who regularly smokes marijuana or hashish may give birth prematurely to an undersized, underweight baby (Mirra *et al*., 2021). Over the last 10 years many children of marijuana users have been born with reduced initiative and lessened abilities to concentrate and pursue life goals. Studies also suggest that prenatal (before birth) (Mendelson *et al*., 1985) use of the drug may result in birth defects (Mendelson *et al*., 1986), mental abnormalities and increased risk of leukemia (cancer of the bone marrow) in children.

(ii) *Short-term effects*:

Short term effects are -- severe anxiety, including fear that one is being watched or followed (paranoia), very strange behaviour, seeing, hearing or smelling things that are not there, not being able to tell imagination from reality (psychosis)(Dixit *et al*., 1977), panic, hallucinations, loss of sense of personal identity (Thompson *et al*., 1973), lowered reaction time, Increased heart rate (risk of heart attack), increased risk of stroke, problems with coordination (impairing safe driving or playing sports), sexual problems (for males), up to seven times more likely to contract sexually transmitted infections than non-users (for females) (Legator *et al*., 1976).

(iii) *Long-term effects*:

Long terms effects are-- decline in IQ (up to 8 points if prolonged use started in adolescent age), poor school performance and higher chance of dropping out, impaired thinking and ability to learn and perform complex tasks, lower life satisfaction, addiction (about 9% of adults and 17% of people who started smoking as teens), potential development of opiate abuse, relationship problems, intimate partner violence, antisocial behaviour including stealing money or lying, financial difficulties, Increased welfare dependence, greater chances of being unemployed or not getting good jobs (Dixit *et al*., 1977).

Effect of marijuana on human male endocrine system:

Male infertility can be caused by many factors that interfere with spermatogenesis, organic dysfunction, and psychogenic causes, including inherited, acquired, and idiopathic reasons (WHO, Infertility, 2021). Marijuana and its active component THC affect multiple endocrine systems. A suppressive effect is seen on the reproductive hormones, prolactin, growth hormone, and the thyroid axis, while the HPA axis is activated. These effects are mediated through CB1 receptor activation in the hypothalamus, which directly or indirectly modulates anterior pituitary function (Gerard *et al*., 1991; Raypole, 2021).

(i) *Hypothalamic- Pituitary-Gonadal axis*:

The secretion of sex hormones is directly controlled by the pituitary and indirectly influenced by the hypothalamus (Fig. 2). From cells in the medial basal hypothalamus, gonadotropin-releasing hormone (GnRH) is secreted in a pulsatile fashion under the influence of a variety of other factors, including endogenous opiates, catecholamines, prolactin, corticotropin-releasing hormone (CRH), and neuropeptide. GnRH stimulates the production of follicle-stimulating hormone (FSH) and luteinizing hormone (LH) in the anterior pituitary

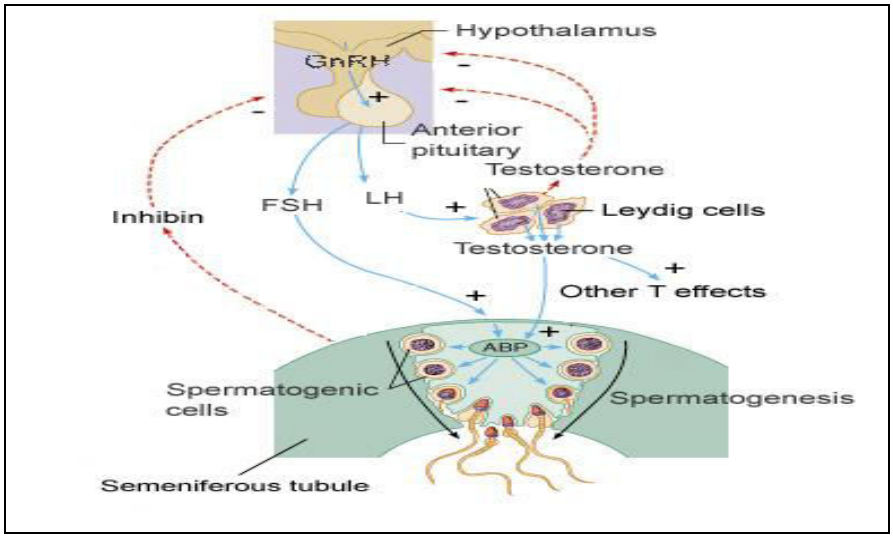

Fig. 2: Hypothalamo-Pituitary-Gonadal axis in male.

gonadotrophs (Gerard *et al*., 1991). In both males and females, FSH and LH act on the gonads, leading to the secretion of testosterone in males and estradiol and progesterone in females (Sauer *et al*., 1983). These hormones feed back to the hypothalamus and anterior pituitary to modulate GnRH and gonadotropin release. Marijuana, Δ9 - THC, and other cannabinoids acutely alter hypothalamic-pituitary-gonadal (HPG) integrity and affect reproductive function by acting at the hypothalamus either directly through GnRH or indirectly through other modulators. These effects are likely mediated by central cannabinoid (CB1) receptors in the hypothalamus. CB1 receptors have also been found in the testes and the ovaries of experimental animals, suggesting a possible direct effect of cannabinoids on the gonads (Murphy *et al*., 1980). In addition, marijuana condensate and Δ9-THC inhibit binding of dihydrotestosterone (DHT) to the androgen receptor (Purohit *et al*., 1980; Galiegue *et al*., 1995). LH stimulates the Leydig cells in the testes to produce testosterone, while FSH primarily acts on the Sertoli cells to regulate spermatogenesis. In the adult human male, testosterone has a variety of actions throughout the body, including the maintenance of secondary sex characteristics, the facilitation of Sertoli cell function, and the promotion of sexual function (Murphy *et al*., 1980). Hypogonadism results in decreased quality of life marked by fatigue, decreased libido, diminished sense of well-being, impaired fertility, and changes in body composition, including reduced bone mineral density and lean body mass.

(ii) *Effects on HPG axis*:

Human studies investigating the effects of cannabinoids on reproductive hormones have been conflicting (Murphy *et al*., 1998). Lower testosterone levels have been reported in chronic marijuana users compared to nonusers, and acute decreases in both LH and testosterone have been observed after marijuana smoking, but multiple subsequent studies have not confirmed these findings. In one study, heavy chronic users were found to have similar testosterone levels compared to casual users at baseline and did not experience any significant alterations in testosterone after a 21-day period of intense marijuana smoking in a controlled research setting. A subsequent study of similar design by the same investigators showed no significant changes in integrated LH levels over the study period (Symons *et al*., 1976). These inconsistent observations may be due to differences in study design but also may reflect the development of tolerance, as suggested by the animal studies. Down-regulation and desensitization of CB1 receptors in the hypothalamus may underlie the weakening of effect observed with chronic cannabinoid administration. Result of LH, FSH and Testosterone levels in marijuana abuse showed

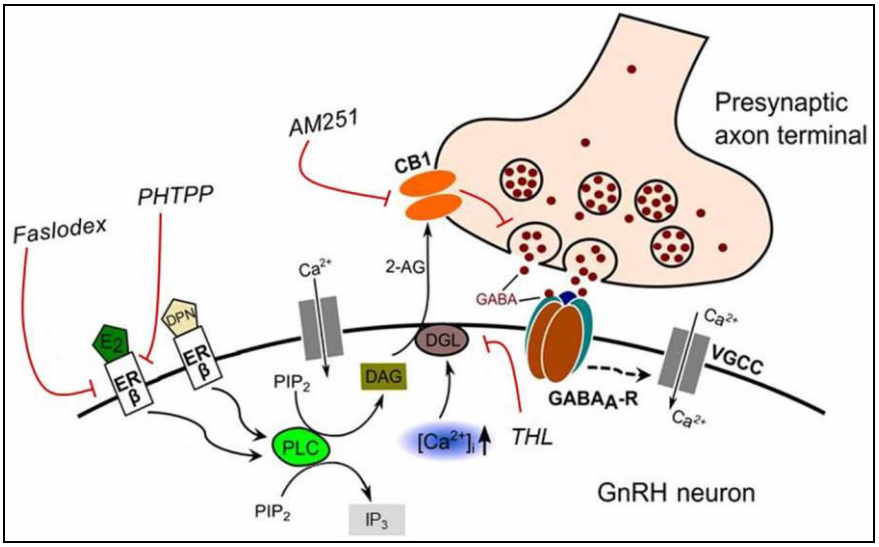

Fig. 3: Effect of endocannabinoids on GnRH hormone (Imre *et al*., 2016).

significant decrease. These results agree with the previous study who stated that in response to drug metabolic stress cannabinoids transiently depress pituitary function as reflected by decrement in LH and FSH hormones. Intra-peritoneal injection of cannabis extract at low doses induced adverse effect on testes histology which revealed significant shrinkage of tubular diameter and detrimental change in seminiferous epithelium of testes with resulting lowered serum testosterone and pituitary gonadotropins (FSH, LH) levels. It is probable that THC affects these hormones through its ability to alter various neural transmitters in the hypothalamus or neural transmitters in the CNS which impinge on the hypothalamus. The dopaminergic and serotonergic fibers seem to be particularly important. The two gonadotropins, LH and FSH, secreted by the pituitary gland are of major importance to reproduction in the male. Both gonadotropins appear to respond to a single releasing factor from the hypothalamus, GnRH, which is sensitive to catecholamine neurotrans-mitters (Fig. 3) (Imre *et al*., 2016). The THC-induced block of GnRH release results in lowered LH and FSH which is responsible for reduced testosterone production by the Leydig cells of the testis (Boris *et al*., 2011). In humans, effects on sperm production and morphology have also been observed. Dose-related oligospermia has been observed in chronic users. Similarly, a 58% decrease in sperm concentration was reported in chronic users after intensive marijuana smoking without a significant change in LH or testosterone. Reversible reductions in sperm concentration were seen 5 to 6 weeks after the initiation of intensive smoking, suggesting an effect on sperm production. In addition, abnormal sperm morphology has been noted in chronic smokers. Although these findings imply a significant effect on gonadal function in humans, the true impact of marijuana on fertility is not known. However, discontinuation of casual marijuana use is recommended for infertile men (Boris *et al*., 2011).

(iii) *The Endocannibiod system*:

The endogenous arachidonate-based lipids, anandamide (*N*-arachidonoylethanolamide, AEA) and 2-arachidonoylglycerol (2-AG); these are known as "endocannabinoids" and are physio-logical ligands for the cannabinoid receptors. Endocannabinoids are all eicosanoids. The enzymes that synthesize and degrade the endocannabinoids are fatty acid amide hydrolase or monoacylglycerol lipase. The cannabinoid receptors CB_1 and CB_2, two G protein-coupled receptors are located in the central and

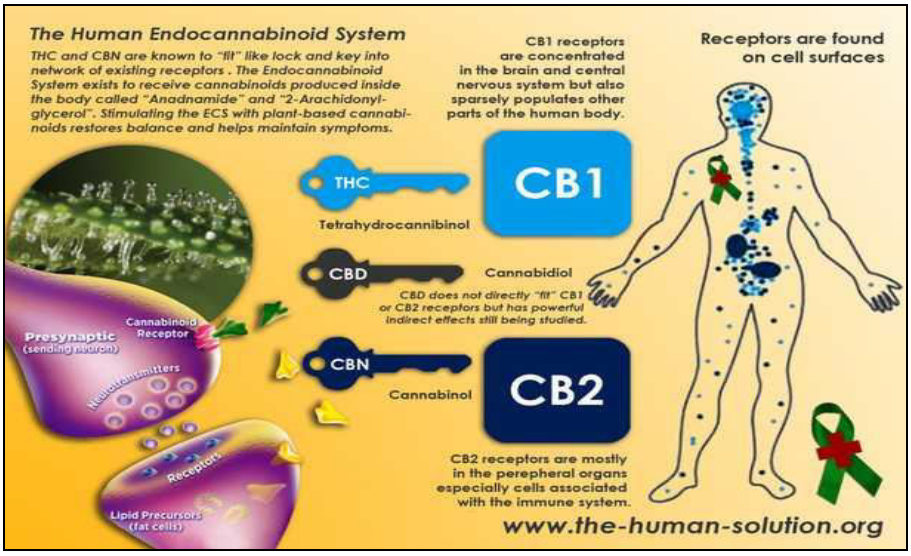

Fig. 4: Human Endocannabnoid System (www.the-human-solution.org).

peripheral nervous systems. The neurons, neural pathways, and other cells where these molecules, enzymes, and one or both cannabinoid receptor types are all co-localized form the endocannabinoid system. The endocannabinoid system has been studied using genetic and pharmacological methods.

These studies have shown that cannabis function as neuromodulators for a wide range of cognitive and physical processes, including motor learning, hunger, and pain perception. There is a significant amount of overlap between the CB1 receptor's location in the endocannabinoid system and the orexinergic projection system, which mediates many of the same physical and mental processes. The lateral hypothalamus and several orexin system output sites also colocalize CB1 with orexin projection neurons, where the two receptors physically and functionally combine to produce the CB1-OX1 receptor heterodimer (Hui Chen *et al*., 2015) (Fig. 4).

(iv) *Endocannabinoid synthesis, release, and degradation*:

During neurotransmission, the pre-synaptic neuron releases neurotransmitters into the synaptic cleft which bind to cognate receptors expressed on the post-synaptic neuron. Based upon the interaction between the transmitter and receptor, neurotransmitters may trigger a variety of effects in the post-synaptic cell, such as excitation, inhibition, or the initiation of second messenger cascades. Based on the cell, these effects may result in the on-site synthesis of endogenous cannabinoids anandamide or 2-AG by a process that is not entirely clear, but results an elevation in intracellular calcium. Expression appears to be exclusive, so that both types of endocannabinoids are not co-synthesized. This exclusion is based on synthesis-specific channel activation: a recent study found that in the bed nucleus of the stria terminalis, calcium entry through voltage-sensitive calcium channels produced an L-type current resulting in 2-AG production, while activation of mGluR1/5 receptors riggered the synthesis of an amide (Hui Chen *et al*., 2015).

(v) *Effect of marijuana actions*:

THC and the brain chemical anandamide share a similar molecular structure. Drugs can be recognised by the body and change normal brain transmission because of their structural similarity. THC and the brain chemical anandamide share a similar molecular structure. Due to structural similarities, THC can be recognised by the body and affect normal brain activity. Anandamide is an example of an endogenous cannabinoid that

performs the role of a neurotransmitter by transmitting chemical signals between nerve cells (neurons) throughout the nervous system. They have an impact on the parts of the brain that control perception of pleasure, memory, thought, concentration, movement, coordination, and timing. Due to this similarity, THC is able to bind to and activate molecules on neurons in certain brain regions known as cannabinoid receptors. This disruption of many mental and physical processes results in the effects mentioned previously. The endocannabinoid system, a neuronal communication network that makes use of these cannabinoid neurotransmitters, is crucial to the nervous system's regular operation, therefore tampering with it can have serious consequences. For example, THC is able to alter the functioning of the hippocampus and orbitofrontal cortex, brain areas that enable a person to form new memories and shift his or her attentional focus. As a result, using marijuana causes impaired thinking and interferes with a person's ability to learn and perform complicated tasks (Block *et al*., 1991). THC also disrupts functioning of the cerebellum and basal ganglia, brain areas that regulate balance, posture, coordination, and reaction time. This is the reason people who have used marijuana may not be able to drive safely and may have problems playing sports or engaging in other physical activities (Rodriguez *et al*., 1994).

People who have taken large doses of the drug may experience an acute psychosis (Thompson *et al*., 1973), which includes hallucinations, delusions, and a loss of the sense of personal identity. THC, acting through cannabinoid receptors, also activates the brain's reward system, which includes regions that govern the response to healthy pleasurable behaviours such as sex and eating. Like most other drugs that people misuse, THC stimulates neurons in the reward system to release the signalling chemical dopamine at levels higher than typically observed in response to natural stimuli. This flood of dopamine contributes to the pleasurable "high" that those use marijuana (Mendelson *et al*., 1974).

The endocannabinoids and male fertility:

The endocannabinoid system (ECS) and male reproduction (Smith *et al*., 1976) are inter connected (Fig. 5). The presence of the ECS has been demonstrated in various cell types that are involved in male reproduction. As previously mentioned, endocannabinoids and cannabinoid receptors have been shown to be present in testicular tissue, including Sertoli and Leydig cells as well as spermatozoa in various species from invertebrates to mammals (Rosenkrantz *et al*., 1980). It is therefore clear that the ECS is deeply involved in the control of the male reproductive system and function of spermatozoa. ECS and the hypothalamus-pituitary-gonadal axis, a fully functional HPG axis is needed to properly orchestrate and maintain the process of spermatogenesis (Mendelson *et al*., 1978). GnRH is released from the hypothalamus which, in turn, stimulates specific nuclei in the pituitary to synthesize and release follicle-stimulating hormone (FSH) and luteinizing hormone (LH). These two gonadotropins act on their respective target tissues in the gonads. Basically, FSH stimulates the Sertoli cells to support developing spermatozoa, while LH leads to the release of testosterone from Leydig cells (Mendelson *et al*., 1978). The ECS has been closely associated with the HPG pathway at multiple levels as CB1 receptors are expressed in the anterior pituitary, Leydig cells and Sertoli cells. CB2 was found in Sertoli cells while other components of the ECS, such as AEA, FAAH, and EMT have also been observed in testicular tissues (Stefan *et al*., 2015). For example, administration of AEA, which usually binds to postsynaptic CB1 receptors, decreased serum LH. This action could be prevented by a specific CB1 antagonist (SR141716). It has been demonstrated that endocannabinoid activates CB1 which, in turn, inhibits spontaneous gamma amino butyric acid (GABA) release. Postsynaptic GABA receptors, located on GnRH neurons, are not activated, and as a consequence, GnRH is not released. Interestingly enough, the inhibitory effect of AEA was higher than that of 2-AG. It has

been postulated that the difference between the effects of AEA and 2-AG on the serum levels of LH is due to the difference in receptor activation as AEA can activate both CB1 and TRPV1 receptors while 2-AG acts only on the CB1 receptor. Furthermore, CB1 receptor expression varies between males and females, thereby indicating that males are more sensitive to cannabinoid-induced changes and subsequently the secretion of pituitary hormones (Collu *et al*., 1975). CB1 receptors have also been found to be present in the Leydig cells of mice and rats. LH and testosterone secretion were decreased in CB1 receptor-inactivated mice (Kolodny *et al*., 1974, 1976). However, in wild-type mice, AEA suppressed the levels of both of these hormones. When they are activated through the endogenous cannabinoid AEA, it not only results in a drop in testosterone levels, but this alteration in sex steroid level can also disturb the spermatogenic process (Mendelson *et al*., 1974). It was furthermore showed that these CB1 receptors are responsible for the actions of exogenous cannabinoids. Sertoli cells play an important role during germ cell development as they nurture the developing spermatozoa. Sertoli cells not only have CB1 and CB2 receptors but also have TRPV1 receptors. AEA can act via these receptors to induce apoptosis of these cells. FSH acts on its receptor on the Sertoli cell to activate two separate pathways. It activates adenylate cyclase which, in turn, causes PKA activation via cAMP, thereby causing increased expression of FAAH. This subsequently causes an increase in FAAH expression by activation of the FAAH promoter through the estrogen response element. FAAH helps to hydrolyze AEA and thereby decrease the intracellular level of AEA. Thus, FAAH has a protective role in preventing AEA-induced apoptosis. Interestingly, studies showed that the CB2 receptor can also play a protective role by decreasing programmed cell death (Stefan *et al*., 2015) (Fig. 5).

Effects on sperm function:

Both CB1 and CB2 receptors are present on spermatozoa. CB1 has been localized to the plasma membrane of the acrosomal region, midpiece, and tail of the spermatozoon, while CB2 receptors are mostly localized in the postacrosomal region as well as mid piece and tail (Sheena *et al*., 2012). The transporters as well as enzymes responsible for synthesis and hydrolysis of endocannabinoids have also been identified in the male gametes of various species, including humans. Francavilla *et al*. (2009) therefore concluded that human spermatozoa exhibit a completely functional ECS. As AEA is present in human seminal plasma spermatozoa are therefore also exposed to this compound in the epididymis , and it is inevitable that the ECS thus play a potential modulatory role in sperm function. AEA, as mentioned earlier is a primary endocannabinoid (Rubino *et al*., 2000). It is synthesized from the membrane phospholipid N-archidonylphosphatidyl ethanolamine (NAPE) by the enzyme NAPE-PLD inside the spermatozoa from where it is transported to the outside via the EMT. AEA can also move back into the cell via the EMT. Once outside, it can act on both CB1 and CB2 receptors. Activation of these receptors modulates the motility of spermatozoa. CB1 receptor activation was found to not only decrease motility and viability of spermatozoa but also inhibit the capacitation-induced acrosomal reaction (Sheena *et al*., 2012). Similarly, the CB1 antagonist, rimonabant (SR141716), increased sperm motility and viability, while it also induced capacitation and the acrosome reaction. It had an overall lipolytic action on the spermatozoa, and it also induced energy expenditure possibly through induction of the pAkt and pBc12 proteins that control pro-survival pathways and regulate metabolism. Studies also showed that CB2 modulated the motility of spermatozoa. It was shown that CB2 activation caused an increase in the slow/sluggish progressive sperm population and CB1 activation increased the immobile spermatozoa. In humans, the endogenous agonists activate both CB1 and CB2 receptors. Therefore, motility will depend on the dose of the agonist

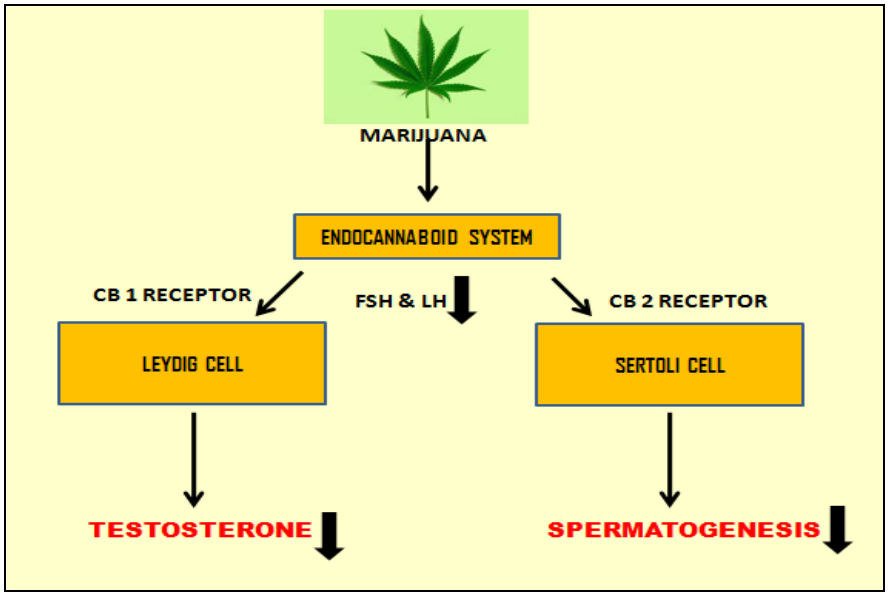

Fig. 5: Effects on leydig cells and sertoli cells, which leads to decrease testosterone and spermatogenesis process.

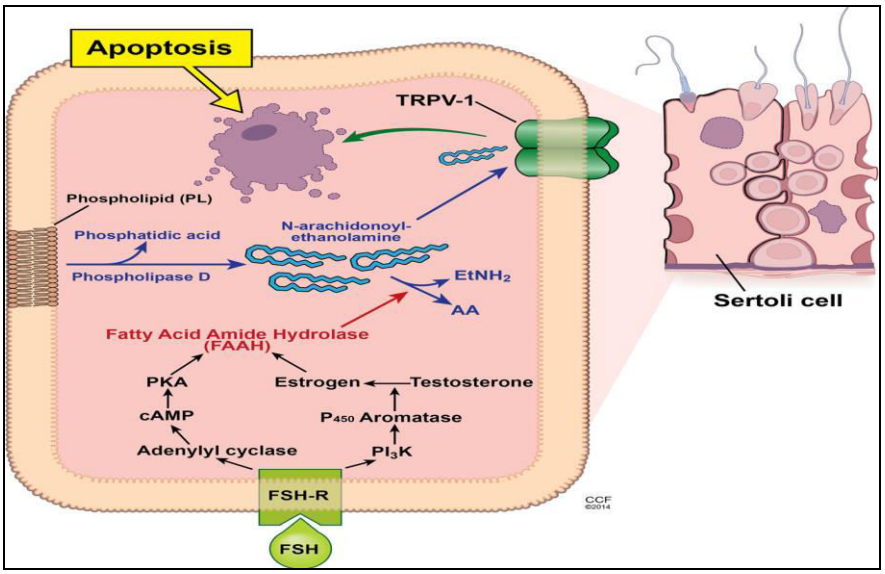

Fig. 6: The involvement of cannabinoids, vanilloid receptors, and FSH in Sertoli cell function (AA: arachidonic acid, EtNH2 ethylamine, FSH: follicle stimulating hormone) (Stefan *et al*., 2015).

(Stefan *et al*., 2015). This is particularly important as the exogenous cannabinoids might cause an inappropriate decrease in motility of spermatozoa. If these substances cause poor motility, it will result in inappropriate completion of capacitation in an area of the female reproductive tract prior to meeting the oocyte (Hembree *et al*., 1976, 1978) (Fig 6). Spermatozoa also express the vanilloid TRPV1 receptor. Along with CB1 receptors, the TRPV1 receptor has been found to play a role in spermatozoa capacitation (Zimmerman *et al*., 1979). The activation of the TRPV1 receptor through AEA binding helps to prevent the spontaneous acrosome reaction to occur in an untimely manner before reaching the oocyte. Unlike the CB1 and CB2 receptors, binding to the TRPV1 receptor occurs intra-cellularly. Supporting the physiological observations

mentioned previously, a study of 86 men presenting at an infertility clinic showed that the levels of AEA in the seminal plasma of both asthenozoospermic and ospermic men were significantly lower compared to normozoospermic men compared to normozoospermic men (Fig. 6) (Murphy *et al*., 1994; Stefan *et al*., 2015).

Similarly, the levels of CB1 mRNA were also decreased significantly in the spermatozoa from these men. The endocannabinoid AEA was found to decrease the mitochondrial activity of spermatozoa (Huang *et al*., 1978), likely through CB1-mediated inhibition, which, in turn, will hamper sperm viability and functions such as motility in a dose-dependent manner (Dalterio *et al*., 1982). AEA also affected motility, capacitation, and acrosome reaction in human spermatozoa in a similar dose-dependent manner. These findings suggest a possible role for the cannabinoid system in the pathogenesis of some forms of male infertility (Sigman *et al*., 1998). CB1 receptors play a central role in preventing the acquisition of motility at too early a stage in the epididymis. Endocannabinoids inhibit the biochemical and physiological changes needed for sperm to undergo capacitation through a CB1-mediated mechanism (Glass *et al*., 2001) and subsequently reduces the ability to AR in various species. In addition, capacitated spermatozoa show a general down regulation of the expression of ECS elements compared to non capacitated sperm. The distinct compartmentalization of CB1/CB2 receptors and of TRPV1 in spermatozoa as well as their levels of expression may critically regulate sperm function. Additionally, the presence of an endocannabinoid gradient in both the male and female reproductive tract (Dalterio *et al*., 1983) can lead to differential spatiotemporal activation of these receptors, thereby affecting sperm function and the various fertilization steps (Fig. 7).

Effect on sperm parameters and function:

Numerous research have looked into how cannabis affects numerous sperm parameters because the ECS is so intricately linked in the control of the male reproductive system. The blood-testis barrier shields the testicles from dangerous substances, just as the blood-brain barrier does for the brain (Mendelson *et al*., 1984). Cannabinoids are lipophilic, though, and they accumulate in testicular/epididymal fat and membranes, where they can be released gradually. This exposure may have an impact on spermatozoa and how they function. A decrease in sperm concentration has been reported in both humans and animals after regular exposure to cannabis (Stefan *et al*., 2015). It also appears that sperm counts are inversely proportional to the amount of drug taken. There is limited evidence for marijuana use to be associated with morphological abnormalities in human spermatozoa.

Morphological abnormalities due to cannabis have been well documented in animal studies. Interestingly, it appears as if THC and CBN, but not CBD, leads to more morphological abnormalities. Human seminal plasma, mid-cycle fallopian tubal fluid as well as follicular fluid contains AEA which suggests that human spermatozoa are sequentially exposed to AEA, indicating a potential modulatory role for the ECS on sperm function (Fig. 8). From the literature, it is evident that sperm motility and viability is mediated via endocannabinoids and CB receptors. Met AEA (stable form of AEA) was shown to decrease human sperm motility and viability via its action through CB1. Several other *in vitro* studies on human spermatozoa are in agreement with these findings. These observations are supported by both *in vitro* and *in vivo* animal studies where it is clearly shown that THC attenuates sperm motility and viability (Asch *et al*., 1979). The fact that THC impairs sperm motility and viability can be explained partially by the fact that it inhibits mitochondrial respiration and activity; therefore, the exposed spermatozoa are starved from energy. These findings are supported by the marked reduction in sperm ATP levels due to THC. It was also shown that THC inhibits fructose metabolism. With fructose being a major energy source for spermatozoa, this could further hamper sperm motility Glycolysis combined with oxidative phosphorylation also

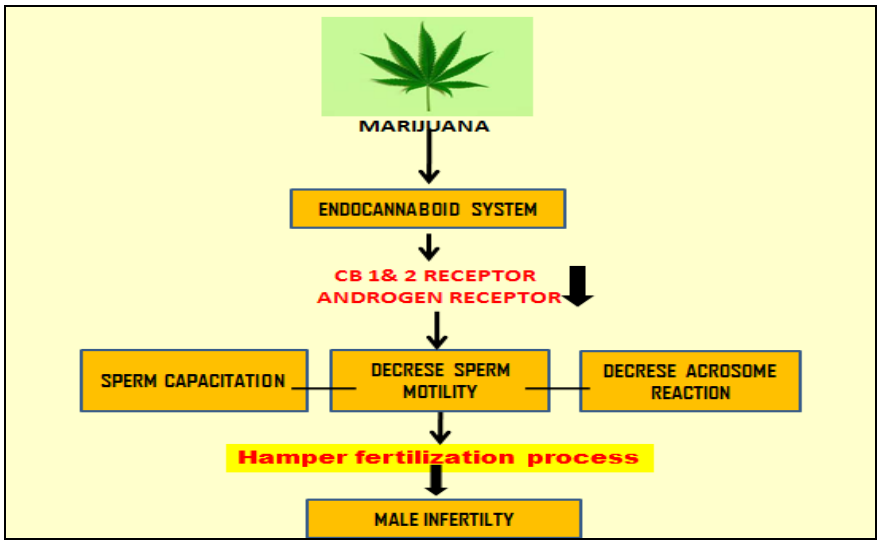

Fig. 7: Effects of marijuana on male fertility by altering the fertilization process.

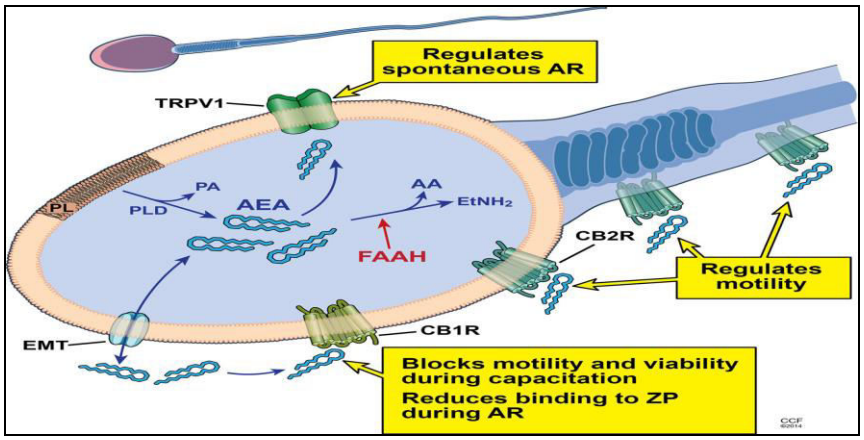

Fig. 8: The influence of the acid, AEA Narachidonoylethanolamine or anandamide, CB1R cannabinoid receptor 1, CB2R cannabinoid receptor 2, EMT endocannabinoid membrane transporter, EtNH2 ethylamine, FAAH fatty acid amide hydrolase, PA phosphatidic acid, PL phospholipid, PLD phospholipase D, TRPV1 transient receptor potential cation channel subfamily V member 1) (Stefan *et al*., 2015).

provides fuel for many other energy dependent processes including capacitation and the acrosome reaction. Disturbing the ECS homeostasis will subsequently adversely affect these energy-dependent processes with implications for gaining fertilizing potential. The ECS is important in keeping the spermatozoa from undergoing capacitation before reaching of the oocyte. This is essential in preventing the spermatozoa from undergoing untimely capacitation in an unusual location. The fact that the process of capacitation is inhibited by cannabinoids means that this effect can be extrapolated to marijuana. It was shown that Met AEA, which is the stable analogue of AEA, inhibits capacitation via the activation of CB1 receptor. Cannabinoids (AEA, THC) has an effect on the acrosome reaction too. CB1 receptor activation prevents the acrosome reaction from occurring. Similar inhibitory findings were observed for both the spontaneous and induced AR after *in vitro* treatment of spermatozoa with either therapeutic or recreational concentrations of THC (Sassenrath *et al*., 1979). Fertilizing ability of spermatozoa also appears to be affected as hyper activated motility, necessary for penetration of zona pellucida, as well as hemizona binding

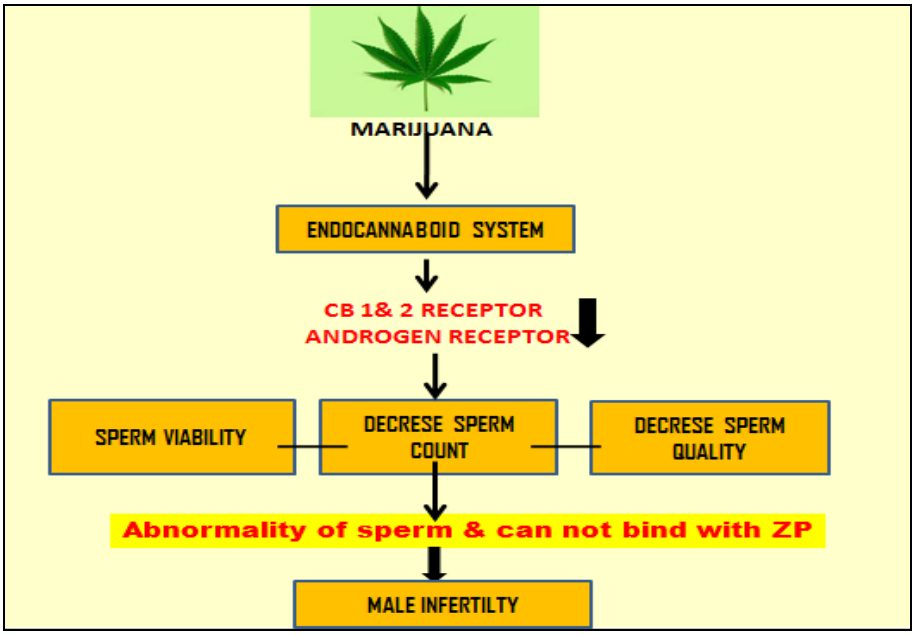

Fig. 9: Effects of marijuana on male fertility by altering sperm parameter.

were negatively affected in AEA-treated spermatozoa (Fig. 9). Interestingly, low/ physiological concentrations of AEA stimulated hyperactivated motility while it was attenuated at higher dosages. This biphasic effect was shown between 1 to 6 h of incubation in AEA (Fig. 9) (Stefan *et al*., 2015).

Conclusion

There is no question that the use of marijuana for recreational and medical purposes will rise and spread. Given the ECS's significant role in the control of male reproduction and the direct effect of exogenous cannabinoids have on the ECS's homeostasis, marijuana's potential influence on the intricately timed events related to male fertilization must undoubtedly be taken into account. Surprisingly few research have examined how marijuana directly affects male fertility. This can mainly be ascribed to legislation and ethical considerations making it virtually impossible to pursue *in vivo* human studies. The current body of knowledge pertaining to this topic mainly consists of a number of earlier human studies and more recently animal, *in vitro*, and retrospective studies. Despite these limitations, it is clear that marijuana and its compounds can influence male fertility at multiple levels. A number of studies have attributed dysregulation of the HPG axis, and in specific reduction in a key hormone such as LH, which, in turn, can affect testosterone and spermatogenesis to marijuana. It appears as if marijuana can actually affect semen parameters and sperm function by acting through both the cannabinoid and vanilloid receptors. Furthermore, sexual health has also been linked to marijuana as it seems to have an effect on erectile function. With the change in legislation and decriminalization of marijuana use, as well as the fact that some studies report conflicting and contradictory findings, it is paramount that more clinical studies should be undertaken to examine the effects of marijuana use in greater detail. The evidence supports the idea that marijuana usage has a negative impact on male reproductive capacity, despite the fact that there are currently few human research and that they are observational in nature. A recent study demonstrated that cigarette smokers overuse cannabis more frequently, and that male smokers of infertile couples had smaller ejaculate volumes despite having higher testosterone levels. It would be interesting to investigate how marijuana use affects these smokers. All of the aforementioned

studies highlight the need for clinicians to ask about marijuana use while assessing male infertility. When recommending medical marijuana, doctors should absolutely keep in mind the connection to and potential effects on male fertility.

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