



REVIEW

Treating Infertility in IVF Clinics: Therapeutic Potential of Stem Cells

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ABSTRACT

Infertility is increasing rapidly in the world because of change in major lifestyle habits and it has been estimated that near about 75 million peoples in the world are facing infertility problems including both male and females. A number of therapeutic approaches like assisted reproductive techniques (ARTs), intra-uterine-insemination (IUI), hysteroscopic surgery, hormonal and chemotherapy have been applied in infertility clinics. Stem cell therapy has been proposed a promising candidate in this field where adult stem cells can be transdifferentiated to oocyte-like cells and SSC-like cells, which can be used to treat infertility like azoospermia etc. In this study, a number of research papers regarding infertility and stem cell therapy have been reviewed and hypothesized that trans-differentiation of stem cells towards SSC-like cells or OSC-like cells, and their clinical trials may authenticate the claims of stem cell therapeutic potential to treat infertility. Transdifferentiation is a process of lineage reprogramming where the fate of a (stem) cell has been determined. Increased research on transdifferentiation of multipotent and pluripotent stem cells (induced pluripotent stem cells (iPSCs), mesenchymal stem cells MSCs) etc toward germ cell lineage in last decade hypothesized that infertility can be treated by using these kind of stem cells. Reliable successes have been achieved in the transdifferentiation of stem cells toward germ-like cells (oocyte-like cells and SSC-like cells). Clinical trials of transdifferentiated mesenchymal stem cells are the only way to answer this question. Clinical trials of trans-differentiated MSCs may give valuable results to combat increased infertility in the world. In conclusion, applications of MSCs in infertility clinics and start of their clinical trials may open new horizons of treatment in infertility.

Keywords: IVF Clinics, Transdifferentiation, lineage reprogramming, mesenchymal stem cells, clinical trials

Infertility is defined as a condition in which a sexually active couple fail to conceive, after having regular intercourse without using contraceptive techniques after 1 year. It has been reported that approximately more than 15% of couples are infertile [1-3]. The failure this natural conception may happens because of male sterility situations like azoospermia or lack of ejaculation

or because of female sterility conditions such as ovarian failure, tubal occlusion or absence of the uterus etc.[4, 5]. Azoospermia, is defined as the absence of sperm and it accounts approximately 15% of infertility in men. Azoospermic condition arises due to pre-testicular, testicular, or post-testicular problems [6]. There are four causes of infertility, i.e. 1) the female factor 2) the male factor

3) combined factors 4) unexplained infertility [7]. The exact percentage for each of these factors is unknown but generally it has been considered from literature reports that approximately 35% infertility cases are because of female factor whereas 30% cases are because of male factor. Abnormalities detected in both partners, and unidentified factors account as 20% and 15% respectively [8-10].

An idiopathic and unexplained male infertility in male is mostly based on the semen quality. Idiopathic type of male infertility has been characterized as an unexplained decline in semen quality, whereas unexplained male infertility can be described as infertility of unknown origin with normal sperm parameters. In spite of having huge research, there is no optimal or acceptable strategy to treat idiopathic male infertility. A number of reasons are involved in female infertility such as ovulatory disorders, tubal disorders, endometriosis, premature ovarian insufficiency etc.[8-10]. Statistical details of infertility causes in males and females are given in figure 2.

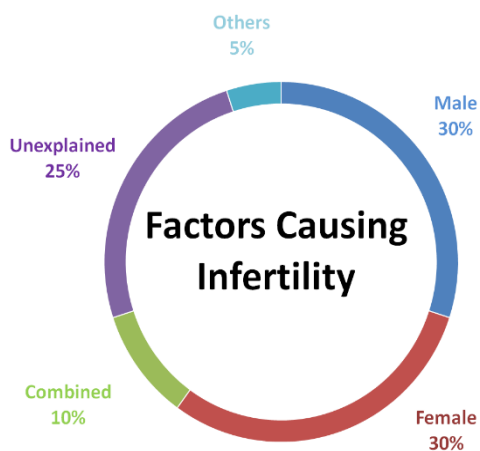
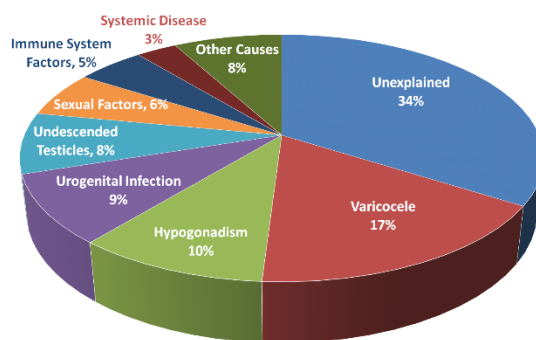
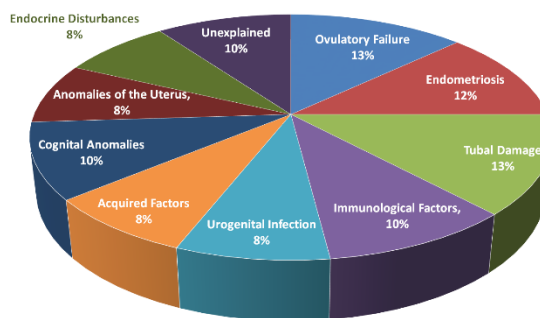


Figure 1: Factors causing infertility in young couples



Factors Causing Male Infertility

[A]



Factors Causing Female Infertility

[B]

Figure 2: Factors causing infertility. A) Causes of male infertility, b) Causes of female infertility

Traditional Infertility Treatment Techniques:

Infertility can be treated by using a number of techniques. In medical terms, this treatment can be categorized into two major categories, (1) specific and (2) non-specific way of treatments. By specific way of infertility treatments, different etiologies like hypogonadotrophic hypogonadism, male accessory gland infection, retrograde ejaculation, and positive antisperm anti-body (ASA) are treated by using following strategies.

Hypogonadotrophic hypogonadism can be treated by gonadotropin replacement therapy, which has been considered as an effective therapy where spermatogenesis and testosterone production have been inhibited.

Male accessory gland infection is caused by the presence of leukocytes and microorganisms which

can be treated by using specific antibiotics for 14-21 days. Alpha adrenergic agonists have been used to treat the patients with ejaculation disorders. In ASA, sperm-egg interaction blockage happens because of the immobilized spermatozoa which can be treated by using assisted reproduction techniques. In non-specific way of treatment, empirical medical treatment (EMT) has much value which can be applied to treat idiopathic infertile males. EMT efficacy is limited in scientific literature because of large, randomized and controlled studies. EMT follows different mode of actions which can be further categorized into (1) hormonal treatment and (2) anti-oxidant supplementation [11-14]. A number of other techniques like ovulation induction via clomifene, Weight loss, Intra-uterine insemination and IVF/ICSI (*in vitro* fertility / intra cytoplasmic sperm injection) and using donated eggs or sperms are also going to be applied in IVC clinics to treat infertility [15].

Stem Cells and Their Characteristics:

Stem cells can be defined as the cells that have self-renewal capacity, can differentiate to one or more lineages and have an enormous proliferative potential for the maintenance of the tissues where they reside as they can be found in almost all types of body organs in quiescent form [16]. They perform their specific physiological role such as highly coordination in growth, differentiation and apoptotic induction [17]. Their self-renewal behaviour allow them to be replenished by the aged cells in almost all organs of the body such as blood, bone, gametes, epithelia, nervous system, muscle etc.[18]. Stem cells have been classified into many categories like totipotent, pluripotent, multipotent, and unipotent. This classification is based on their developmental and differentiation potential toward other lineages, for example a totipotent stem cell has a full potential to give rise to a new individual, a pluripotent stem cell can differentiate to all somatic and germ lineages and an adult multipotent stem cell can differentiate into multiple cell types of a single lineage [19,

20]. A unipotent or precursor cell, have limited self-renewal ability and can develop only, one mature cell type of its own lineage. The unipotent or precursor cells can also be reprogramed to be pluripotent stem cells by adding few defined growth factors while in culture which can be applied in clinics for various diseases [21, 22].

Stem Cells in Clinical Trials:

Stem cell therapy in recent years is going to be much popular as one of the modern therapeutic approaches to treat diseases and mesenchymal stem cells (MSCs) are going to be the promising candidate in stem cell therapy [23-25]. MSCs are pluripotent, according to some scientists these are multipotent cells having self-renewal capacity. MSCs has a number of advantages over other adult stem cells for therapeutic purposes because of their accessibility, ease of culture and proliferation *in vitro*, potential to modulate tissue repair and biological stability in long-term culture [26]. For now, they are isolated from almost all parts of the body, for example, skin, blood, umbilical cord blood, dentine, pancreas, adipose, liver, brain, heart, lungs, and kidneys [27-30]. Adipose tissue has been named as the most reliable and largest source of these stem cells. MSCs are being applied in clinics in daily bases and their clinical trials are going to be increased every year. Data extracted from the website of US government clinical trials (www.clinicaltrials.gov), using the keywords “mesenchymal stem cell therapy” showed that 440 studies are registered in clinical trials from which 416 are registered in last ten years. Years based arrangement of data indicate the increased number of clinical trials per year around the world as shown in **figure 00**.

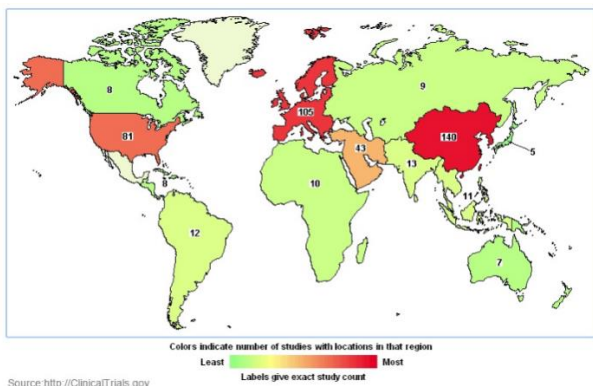


Figure 3: Geographical distribution of clinical studies using mesenchymal stem cell studies around the world.

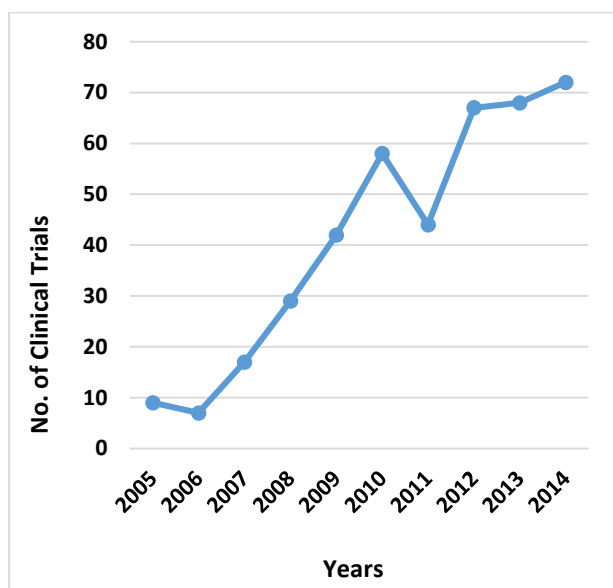


Figure 4: Number of Clinical trials using mesenchymal stem cells study in the last decade.

More than 70% of all these clinical trials have been done in five countries i.e. China, USA, Spain, South Korea and Iran. Global map of mesenchymal stem cell therapy is given in figure 3.

Stem Cells in Infertility Treatment:

In andrology, stem cells have been considered as potential therapeutic agents to treat erectile dysfunction and infertility in males [31]. Primordial germ cells (PGCs) are the potent cells

of germ cell lineage in mammals which undergoes sexually dimorphic development and generate spermatozoa and oocytes in males and females, respectively [32-34]. Mature spermatozoa and oocytes after fertilization give rise to zygotes, which are termed as totipotent cells having full developmental potential developing an individual from a diploid cell. By using lineage reprogramming and transdifferentiation techniques, stem cells like iPSCs (induced pluripotent stem cells) and MSCs can be reprogrammed or transdifferentiated towards germ cell lineages, producing mature and normal oocytes and spermatozoa [35]. In conclusion, stem cells or more specifically adult stem cells have a huge potential to be applied in clinics in order to treat infertility in males as well as in females [36-38].

Male fertility is a multi-staged process in which spermatozoas are produced throughout life efficiently via a process termed as spermatogenesis in individual testis. Spermatogenesis is a highly coordinated process involving the regular proliferation and differentiation of germ cells [39, 40]. Sertoli cells and interstitial leydig cells controlling production of spermatozoa, are providing them vital aqueous environment and producing testosterone, respectively [41, 42]. The male germ line stem cells are responsible for the production of spermatozoa in a unique and sequential step. These germline cells are also responsible for the inheritance of characters for the next generation [43]. Spermatogonial stem cells (SSCs) are the male germ line stem cells, which are responsible for the production of sperms throughout life [44, 45]. Problems in spermatogenesis caused by any factor may lead to male infertility. As it has been discussed that this is a cellular coordinated mechanism with the fine regulation and coordination of germ cells, sertoli cells etc., it can be concluded, that male infertility has a promising potential to be treated via cell therapeutic approaches [46-48]. In 1994, Brinster demonstrated that germ cells can be transplanted to treat infertility by stimulating donor derived spermatogenesis [49]. In another experiment, transplantation of spermatogonial stem cells were used and germ cells were found to be

responsible for spermatogenesis and their transplantation may help to treat fertility [48, 50, 51].

After having a number of studies on successful transplantation of SSCs done, transplanted cells were used for in vitro fertilization (IVF) supported the hypothesis of germ cell transplantation (allogeneic and xenogeneic) to treat infertility, but a number of challenges like the arrest of spermatogenesis remained a question [52-56]. However, objections raised on the spermatogenesis failure of xenogeneic transplanted SSCs received an authentic explanation when fertility was performed successfully after the transplantation of primordial germ cells between rainbow trout and salmon [56]. Transplantation of male germ cells help us to improve our understanding regarding the potential causes of male infertility and the possible ways to treat it. Genetic modification in germ lines to treat and preserve fertility in infertile patients and endangered animals are underway to have improved efficiency [57]. This journey of transplantation will help to enhance our understanding in male fertility which is a major cause of couple infertility in the world. Preclinical and a number of clinical experiments, regarding the restoration of fertility following SSCs transplantation have been considered as a promising therapeutic approach [42].

Female fertility is defined as the successful conception of sperm by egg and successful fertilization, leading to the development of an individual. The fertility of a female depends on the function of ovum, how it behaves and perform its functions normally to receive a sperm [58]. As it is discussed above, a number of reasons like ovulatory disorders, tubal disorders, endometriosis, premature ovarian insufficiency etc. are involved in female infertility. A number of strategies are also applied to treat female infertility. In recent years, stem cell therapy has been proposed as a new therapeutic strategy for restoring the structural and functional activities of an organ or tissue in the body [59]. In early

1950s, it was believed that ovarian failure is irreversible and female infertility cannot be treated, but now development in scientific technologies, made it possible and stem cell therapy has shown a promising role in the treatment of female infertility [60]. Successful isolation, identification and characterization of germline stem cells (i.e. PGCs, Oogonial stem cells OSCs etc.) and their differentiation towards oocytes present a landmark innovation in reproductive biomedicine [61-63]. It has been discussed earlier that differentiation of germline cells, in mammals known as primordial germ cells (PGCs) into oocytes can be performed and these cells can be used for in vitro maturation (IVM) and in vitro fertilization (IVF). It has been proposed that OSCs can be a promising agent for future fertility management [59]. It also has been observed that OSCs, along with their potential to develop oocytes and embryos are safe, as they do not form teratomas [64].

Conclusion:

Stem cell therapy as being a promising and recognized way of therapy is being applied in almost all fields of medical and paramedical sciences as a regenerative agent of damaged or necrotic tissues. Stem cells have also been considered to a hopeful agent to treat the birth disorders like infertility etc. A huge number of clinical trials using stem cells to treat birth disorders and to identify the potential of stem cells in re-organogenesis, are required.

Conflict of Interest:

There is no conflict of interest with any person or organization regarding this manuscript.

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