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Stem cells in reproductive medicine: ready for the patient?

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STUDY QUESTION: Are there effective and clinically validated stem cell-based therapies for reproductive diseases?

SUMMARY ANSWER: At the moment, clinically validated stem cell treatments for reproductive diseases and alterations are not available.

WHAT IS KNOWN ALREADY: Research in stem cells and regenerative medicine is growing in scope, and its translation to the clinic is heralded by the recent initiation of controlled clinical trials with pluripotent derived cells. Unfortunately, stem cell 'treatments' are currently offered to patients outside of the controlled framework of scientifically sound research and regulated clinical trials. Both physicians and patients in reproductive medicine are often unsure about stem cells therapeutic options.

STUDY DESIGN, SIZE, DURATION: An international working group was assembled to review critically the available scientific literature in both the human species and animal models.

PARTICIPANTS/MATERIALS, SETTING, METHODS: This review includes work published in English until December 2014, and available through Pubmed.

MAIN RESULTS AND THE ROLE OF CHANCE: A few areas of research in stem cell and reproductive medicine were identified: *in vitro* gamete production, endometrial regeneration, erectile dysfunction amelioration, vaginal reconstruction. The stem cells studied range from pluripotent (embryonic stem cells and induced pluripotent stem cells) to monopotent stem cells, such as spermatogonial stem cells or mesenchymal stem cells. The vast majority of studies have been carried out in animal models, with data that are preliminary at best.

LIMITATIONS, REASONS FOR CAUTION: This review was not conducted in a systematic fashion, and reports in publications not indexed in Pubmed were not analyzed.

WIDER IMPLICATIONS OF THE FINDINGS: A much broader clinical knowledge will have to be acquired before translation to the clinic of stem cell therapies in reproductive medicine; patients and physicians should be wary of unfounded claims of improvement of existing medical conditions; at the moment, effective stem cell treatment for reproductive diseases and alterations is not available.

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Introduction

Research in stem cells and regenerative medicine is a growing field with promising clinical trials in their initial stages. Unfortunately, together with scientifically sound research, there is a flourishing industry offering unproven and often unsafe stem cell-based 'therapies'. Untested treatments are also affecting the field of reproductive medicine, often leaving both the physician and the patients unsure about their options. The International Society for Stem Cell Research has in fact published a handbook directed at patients with the aim of helping them understand the claims of stem cell treatments, and indicating the issues they should keep in mind when thinking about stem cell-based therapies for any disease. Recently, the first report of significant improvement in patients treated with retinal pigment epithelium derived from human embryonic stem cells (Schwartz *et al.*, 2012) indicates that the technology is on the brink of being applied in several clinical trials worldwide.

Infertility affects up to 15% of reproductive-aged couples worldwide (WHO, 2010). Current infertility treatment options include hormonal stimulation, intrauterine insemination, IVF or ICSI, gamete donation, and uterine surrogacy. For both men and women experiencing infertility, most treatment options rely on the premise that both partners produce functional gametes. For those couples where one or both partners are unable to produce functional gametes, no treatment options are currently available other than the use of donor gametes. Several factors contribute to the absence of functional gametes, of which genetic syndromes and medical treatments such as chemo- or radiotherapy and immunosuppressive treatments are the most common (Schlegel, 2009). Donation of gametes coincides with the loss of a genetic link with the child, which many couples do not accept. As an alternative to gamete donation, the possibility of patient-specific pluripotent stem cell-derived gametes has been raised through somatic cell nuclear transfer (SCNT) or induced pluripotent stem cells (iPSC) generation.

Another approach to produce functional gametes is to use germline progenitors which are present in the gonads, such as spermatogonial stem cells (SSC) in the male and the controversial ovarian stem cells (OSC) in the female. Finally, stem cell therapy has been proposed as a possible solution to a wider array of reproductive problems, which do not involve gametes, from endometrial damage to erectile dysfunction and vaginal atrophy.

In the present context of stem cells' great promises, we review the current knowledge in stem cell biology and cell therapies related to reproductive medicine, and offer an opinion on the use of such therapies in the clinic.

Background knowledge

The chain of potency

Human embryonic development starts with the fusion of the spermatozoon and the oocyte, forming the zygote (Fig. 1). The zygote is a 'totipotent' cell, which means that it can form any human cell type including extra-embryonic tissues. A slightly less plastic level of potency is called 'pluripotency'; cells in the pluripotent state are able to form all tissues in the adult organism, and are found in certain stages of pre- and post-implantation development of the embryo, including the inner cell mass (ICM) of the blastocyst. However, pluripotent cells cannot give rise to extra-embryonic tissues. The pluripotent state is characteristic for embryonic stem cells (ESC), iPSC, SCNT, embryonic carcinoma cells (EC) and embryonic germ cells (EG). Adult stem cells, such as mesenchymal stem cells or bone marrow stem cells, are considered to be 'multipotent', because they can give rise to a variable but limited number of cell types restricted to the same lineage. Finally, 'unipotent' stem cells correspond to tissue-specific progenitors, which can usually give rise to just one cell type *in vivo*.

Derivation of different types of stem cells

Human ESC (hESC) are mostly derived from the ICM of a blastocyst stage embryo, and were first reported in 1998 (Thomson *et al.*, 1998); they can also be obtained from other developmental stages such as 8-cell stages and morula. Patient-specific pluripotent cells can be generated from SCNT constructs, by replacing the oocyte genetic material with a somatic nucleus (Tachibana *et al.*, 2013).

One of the major scientific breakthroughs in stem cell biology in recent years has been the development of patient-specific pluripotent cells through overexpression of a set of four transcription factors (OCT4,

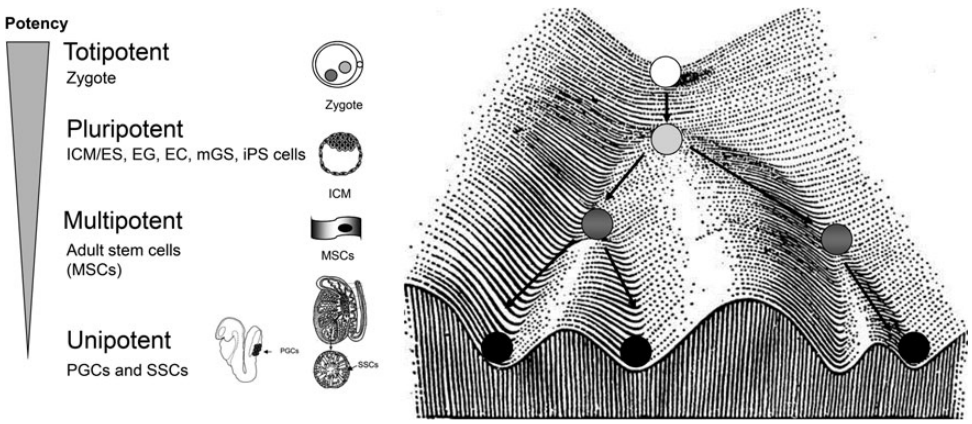

Figure 1 Schematic representation of the loss of potency as development and cell differentiation occurs. ICM, inner cell mass; ES, embryonic stem; MSC, mesenchymal stem cells; PGC, primordial stem cells; SSC, spermatogonia stem cell. Adapted from Eguizabal *et al.* (2013).

SOX2, cMYC and KLF4) in somatic cells (Takahashi and Yamanaka, 2006; Takahashi *et al.*, 2007). These cells are called iPSCs.

Adult mesenchymal stem cells can be derived from bone marrow, cord blood and adipose tissue, among others. They are widely used in therapeutic applications although their mode of action is not completely understood. In the context of this paper, mesenchymal stem cell therapy is considered in endometrial and vaginal atrophy, and erectile dysfunction.

SSCs constitute a small population of progenitor cells which are located in the basal compartment of the seminiferous tubules, surrounded by Sertoli cells. SSC derive from gonocytes in the embryonic developing gonads, and resume their mitotic activity post-natally. Their role is to produce cells of the spermatogenic lineage, in order to provide sperm production throughout life in men.

In the mammalian female, the number of primary oocytes is set at birth and decreases with time until the pool is functionally exhausted at menopause. In 2004, however, mice studies challenged this long-held tenet of reproductive biology, suggesting instead that post-natal ovaries possess the capacity of producing germ cells, and thus new oocytes throughout life (Johnson *et al.*, 2004). Although a few studies have followed, the existence of an ovarian stem cell pool in the adult human ovary remains under debate.

Naïve and primed pluripotent stem cells

Despite the common origin of mouse and human ESC from the blastocyst ICM closer scrutiny of these cells revealed striking differences. In 2007, a new type of pluripotent stem cell (PSC) was isolated from the post-implantation epiblast, the mouse epiblast stem cells (mEpiSCs) (Brons *et al.*, 2007; Tesar *et al.*, 2007) that showed remarkable similarities with hESCs (Nichols and Smith, 2009). The term 'naïve' was introduced to describe the pluripotent nature of mESC, and the term 'primed' for the mEpiSC and hESC. There are important differences between these states of pluripotency. First of all, unlike mEpiSCs, mESCs can give rise to chimeras with germ-line transmission potential. Second, although both types of cell share expression of key pluripotency factors, these factors are wired to different upstream signaling modules. Recently, a landmark publication described culture conditions that permit direct derivation of naïve hESCs from the blastocyst (Gafni *et al.*, 2013), and this was followed by a series of other papers describing the procurement of naïve hESC (Takashima *et al.*, 2014; Theunissen *et al.*, 2014; Ware *et al.*, 2014). The understanding of the naïve state of pluripotency is of primordial importance for the differentiation toward gametes (Hayashi *et al.*, 2011, 2012).

In vitro gamete production

From pluripotent cells to primordial germ cells

In humans, development of gametes goes through a fetal stage primordial germ cells (PGCs). PGCs can be first identified at the end of the third week of gestation in the wall of the yolk sac. After migration, human PGCs colonize the gonadal tissue at 5–6 weeks of gestation. In order to restore the developmental potential in the next life cycle, PGCs undergo global epigenetic reprogramming through chromatin remodeling, erasure of genomic imprints and extensive DNA demethylation (Hajkova *et al.*, 2008). The X chromosome is also reactivated in female germ cells (Seki *et al.*, 2005). Finally, following reprogramming, PGCs enter a phase of global re-methylation, resulting in a highly methylated sperm or partially methylated oocyte genome (Smith *et al.*, 2012).

In the last few years it has become clear that PSCs in general can differentiate into PGCs. Putative PGCs were derived *in vitro* from mESC (Hubner *et al.*, 2003; Toyooka *et al.*, 2003; Geijsen *et al.*, 2004; Eguizabal *et al.*, 2009) and iPSC (Imamura *et al.*, 2014). In the human species several authors could show an increase of later stage PGC markers (Clark *et al.*, 2004) by embryoid body (Bucay *et al.*, 2009), monolayers (Tilgner *et al.*, 2008, 2010) or co-culture with fetal gonads (Park *et al.*, 2009).

In 2011, mouse PGCs were generated *in vitro* using a novel approach (Hayashi *et al.*, 2011). It was demonstrated that the majority of the EpiSC have lost their competency toward a PGC fate (Hayashi and Surani, 2009). To circumvent this hurdle, naïve mESCs derived from the preimplantation epiblast were differentiated toward an intermediate epiblast-like stage in the presence of activin A, resembling more the early pluripotent epiblast cells that give rise to PGCs *in vivo* (Hayashi *et al.*, 2011). The PGCs formed using this novel approach could eventually lead to functional oocytes and sperm capable of generating new offspring.

Spermatozoa from ESC/iPS

Obtaining PGCs from pluripotent cells is the first step in the differentiation toward post-meiotic spermatozoa. However, further differentiation has proven challenging *in vitro*. The strategies employed to favor it include supplementing the differentiation medium with the growth factors bone morphogenetic protein (BMP)-4, -7, -8b (Geijsen *et al.*, 2004; Tilgner *et al.*, 2008), N2B27, activin, Fibroblast growth factor 2 (FGF2) (Hayashi *et al.*, 2011), retinoid acid (Nayernia *et al.*, 2006; Eguizabal *et al.*, 2009, 2011; Cai *et al.*, 2013; Peng *et al.*, 2013), R115866 (Eguizabal *et al.*, 2011), and hormones such as Insulin and Testosterone (Easley *et al.*, 2012). Moreover, the overexpression of specific male germ cell genes such as deleted in azoospermia (DAZ), deleted in azoospermia like (DAZL), BOULE (Kee *et al.*, 2009; Panula *et al.*, 2011),VASA (Tilgner *et al.*, 2008, 2010), Blimp1, PR Domain Containing 14 (PRDM14), and transcription factor Ap-2 gamma (TFAP2C) (Nakaki *et al.*, 2013) have been used to guide differentiation toward spermatozoa.

A relevant step is also to recreate *in vitro* the testicular niche for a better differentiation of pluripotent cells, where male pre-meiotic germ cells can be differentiated along with Sertoli and Leydig cells (Bucay *et al.*, 2009; Eguizabal *et al.*, 2011).

As seen before, correct epigenetic reprogramming is essential for the development of functional gametes in general. However, although the male germ cells obtained *in vitro* follow a normal methylation pattern for some imprinted genes (H19, Insuline Growth Factor 2 (IGF2), Small Nuclear Ribonucleoprotein Polypeptide N (SNRPN)) (Tilgner *et al.*, 2008; Kee *et al.*, 2009; Park *et al.*, 2009; Eguizabal *et al.*, 2011; Panula *et al.*, 2011; Medrano *et al.*, 2012) in human, the methylation pattern and offspring were abnormal (Nayernia *et al.*, 2006) when using a totally *in vitro* protocol for the generation of mouse male germ cells. Healthy offspring with normal methylation patterns of imprinted genes are conversely obtained when gametogenesis is resumed in *in vivo* conditions (Hayashi *et al.*, 2011; Nakaki *et al.*, 2013).

An important proof of functionality of male germ cells from pluripotent cells is to transplant them into the testis of sterile individuals, or

performing ICSI, as demonstrated in the mouse (Geijsen *et al.*, 2004; Hayashi *et al.*, 2011; Nakaki *et al.*, 2013).

Recently, human iPSCs (hiPSC) from azoospermic and normospermic males were transplanted into sterile mouse testis, partially colonizing the testicular niche and showing signs of early stage spermatogenesis (Ramathal *et al.*, 2014), raising the question of whether an initial *in vitro* differentiation step is needed at all. It has been recently shown that transplanting *in vitro* produced PGC-like cells into the mouse testis could be used to obtain functional mouse sperm, capable of producing healthy and fertile offspring (Hayashi *et al.*, 2011). From the recent scientific literature, it seems that so far there is still the need of a natural testicular niche in order to obtain mature functional spermatozoa, and that *in vitro* production of spermatozoa is currently not possible.

Oocytes from ESC/iPS

Several authors demonstrated that mESCs are able to form follicle-like structures with oocyte-like cells, but which were not able to progress into meiosis (Hubner *et al.*, 2003; Lacham-Kaplan *et al.*, 2006). Research conducted using human pluripotent stem cells (hPSCs) to direct differentiation into the female germ line has so lagged far behind. As mentioned above, this outcome was recently achieved in the mouse model by Hayashi *et al.* (2012) by making use of PGCs obtained from female EpiLCs, which were subsequently aggregated with somatic cells from embryonic ovaries and transplanted under the ovarian bursa. PGC-derived immature oocytes were obtained and were subsequently matured and fertilized *in vitro*, and embryos were transplanted into foster mothers. While this *in vitro* approach was less efficient than the use of *in vivo* derived PGCs, healthy and fertile offspring were obtained.

Still, PGC themselves were often not completely 'normal' as, for example, the second-generation PGCs often produced fragile and misshapen oocytes, sometimes lacking supporting granulosa cells. After fertilization of these artificial oocytes, abnormal pronucleus formation was often observed, which could be responsible for the lower rates of post-implantation development compared with controls. These defects could arise from faulty epigenetic reprogramming during the *in vitro* formation of PGCs (Cyranoski, 2013).

So far, there are no reports of studies in which this same approach was applied for the generation of human oocytes, therefore *in vitro* production of human oocytes from pluripotent cells is still a distant prospect.

Spermatozoa from SSCs

The process of sperm production, i.e. spermatogenesis, is a tightly regulated process that takes place in the seminiferous tubules within the testis. This process starts with SSCs, that either self-renew to keep the stem cell pool intact or differentiate to ensure the continuous, lifelong production of $>40 \times 10^6$ haploid sperm cells per day from puberty onwards in the normospermic man. Unfortunately, SSCs cannot be distinguished from their committed daughter cells due to the lack of SSC specific morphological or molecular markers. The only way to determine the presence of SSCs is by means of (xeno)transplantation. If transplantation is conducted within the same species, the transplanted SSCs will colonize the testis through a series of self-renewal divisions followed by differentiation to functional spermatids (Parreira *et al.*, 1998).

Autotransplantation of SSCs has therefore been suggested as a clinical solution for restoring fertility in male survivors of prepubertal cancer who have become sterile due to gonadotoxic treatment. SSC cell therapy will then include cryopreservation of a testis biopsy before gonadotoxic treatment, *in vitro* propagation of SSCs from the biopsy and autotransplantation once the patient has recovered. Besides SSC autotransplantation, alternative options for autotransplantation are to generate sperm from immature germ cells *in vitro* or in a tissue graft. These alternative methods are still experimental. Furthermore, it is not clear if in these methods sustained spermatogenesis will occur from SSCs or whether sperm arises only from a single wave from committed spermatogonia.

Several steps of the SSC cell therapy have been studied extensively in the mouse. The technique of SSC transplantation was first described in 1994 (Brinster and Zimmermann, 1994) and in 1996 (Avarbock *et al.*, 1996) it was shown that frozen SSCs are also capable of restoring spermatogenesis and fertility upon transplantation in infertile mice. Thereafter, autotransplantation has successfully been established in several other species including rat, bull, goat, sheep, dog and recently non-human primates (Hermann *et al.*, 2012). In all these models the concept of transplantation was investigated by taking cells from the testis of allogenic animals and transplanting them to an allogenic recipient that was treated with the chemotherapeutic compound busulfan or with local irradiation to empty the SSC niche in the seminiferous epithelium. Besides demonstrating the effectiveness of the transplantation, these results also demonstrate that chemotherapeutic treatment or irradiation does not seem to affect the ability of the testis to support spermatogenesis, even in the prepubertal phase of life. The efficiency of the SSC transplantation is highly associated with the number of stem cells injected, while the number of SSCs in the testis is limited. Hence, *in vitro* propagation of SSCs prior to transplantation will be essential to clinically apply this technology. Long-term propagation of SSCs *in vitro* was developed for the first time in 2003 in a mouse model (Kanatsu-Shinohara *et al.*, 2003). Thereafter, long-term propagation of SSCs has been established for other animal model systems including rat, hamster, bovine, and recently for humans (Sadri-Ardekani *et al.*, 2009, 2011).

SSC therapy in humans is not clinically applied yet. Nevertheless, cryopreservation of testicular biopsies is now offered at various centers. Translational studies on some aspects of the therapy are still required before bringing SSC therapy to the clinic (Struijk *et al.*, 2013).

Oocytes from OSC

It has long been thought that the female oogonia in the developing ovaries either degenerate or differentiate into primary oocytes, resulting in the absence of renewable germ cells in mammalian females. In 2004, however, mice studies challenged this long-lasting dogma of a fixed ovarian reserve, suggesting that mouse ovaries contain mitotically active germ cells, and thus are able to produce new oocytes throughout life (Johnson *et al.*, 2005). In 2012, researchers claimed that ovaries of reproductive-aged women, similar to adult mice, contain some mitotically active germ cells that can be propagated *in vitro*, and after re-aggregation with dispersed adult ovarian tissue, some follicle-like structures containing oocytes were observed (White *et al.*, 2012).

However, the theory of the existence of an OSC pool remains controversial. An alternative explanation indicates that OCS are dedifferentiated cells which have the capacity to develop as germ cells under certain conditions (Lei and Spradling, 2013), or that OSC are in fact very small embryonic-like stem cells in the ovary (Bhartiya *et al.*, 2013). Using an endogenous genetic approach, Zhang (Zhang *et al.*, 2012) clearly demonstrated that germline cells in post-natal female mice do

not enter mitosis nor contribute to *de novo* folliculogenesis. While some researchers postulated that *de novo* oocyte production originates from circulating bone-marrow-derived germ cells (Johnson *et al.*, 2005), this claim was refuted, with no proof of bone marrow cells, or any other circulating cells, contributing to the *de novo* production of oocytes (Eggan *et al.*, 2006).

Although commercial enterprises are actively studying the possibility of using putative OSC to treat some kinds of infertility, there is currently no consensus on their existence, origin and functionality.

Stem cells and the reproductive tract

Stem cells and the endometrium

In humans, regeneration of the endometrium occurs monthly during reproductive life and beyond under appropriate ovarian steroid priming. Cyclic replenishment of the cellular compartments of the endometrial functionality by adult stem cells (ASC) is essential for the preparation of this organ for its main function, i.e. allowing implantation and pregnancy to proceed (Cha *et al.*, 2013).

ASC present a MSC phenotype (Chan *et al.*, 2004; Schwab and Gargett, 2007; Schwab *et al.*, 2008; Cervello *et al.*, 2010) and they functionally contribute to human endometrial regeneration *in vitro* and *in vivo* (Cervello *et al.*, 2010, 2011; Matsuda and Shi, 2010).

In addition to the existence of an ESC niche located at the perivascular region of the basal layer, the contribution of the bone marrow as an exogenous source of endometrial ASC was first described in 2004 (Taylor, 2004). Bone marrow-derived stem cells (BMDSCs) have been shown to contribute as an exogenous source to tissue repair and regeneration of different organs and tissues (Pittenger *et al.*, 1999). In the human, BMDSCs are also a source of non-hematopoietic cells in the different endometrial cellular compartments (stroma, glandular epithelium, and luminal epithelium). They contribute mainly to the formation of endometrial stromal compartment cells and to a much lesser extent to the glandular and luminal epithelium (Du and Taylor, 2007; Mints *et al.*, 2008; Ikoma *et al.*, 2009; Cervello *et al.*, 2012).

Asherman's Syndrome (AS) consists of a destruction of the endometrium caused by repeated or aggressive curettages, or endometritis. It produces an obliteration of the uterine cavity with intrauterine adhesions and absence of functional endometrium in many areas. Women with this condition or with atrophic endometrium ($\leq$4 mm) often struggle with infertility, menstrual irregularities including amenorrhea, hypo menorrhea, and recurrent pregnancy loss (Yu *et al.*, 2008). It has been recently proposed that BMDSC infusion might improve endometrial regeneration in a murine model of AS (Alawadhi *et al.*, 2014; Jing *et al.*, 2014; Kilic *et al.*, 2014; Zhao *et al.*, 2015). However, peripheral blood stem cell transplant (PBSCT) did not result in engraftment of donor stem cells in the recipient uterus in a macaque model, or human (Wolff *et al.*, 2013). Although endometrial stem/progenitor cells hold great promise for new treatments for infertility associated disorders (Deane *et al.*, 2013), currently no effective treatment for these endometrial pathologies exists.

Stem cells and vaginal reconstruction

Apart from infections, many vaginal pathologies and conditions affect its function, such as Mayer Rokitansky Kuster Hauser syndrome (MRHK), vaginal prolapse, vaginal fistula, cancer and other types of trauma/surgeries. The majority of treatments available implies surgery and the transplantation of synthetic/biological meshes; unfortunately these procedures can give rise to complications like fibrosis, incorrect vascularization and rejection of the graft (Umoh and Arya, 2012). The use of stem cells for vaginal recovery has been poorly investigated so far, but most work has been carried out with muscle-derived stem cells (MDSC). Recently, muscle has been identified as an alternative source of adult stem cells, different from satellite cells, possessing the capacity to differentiate into different cell lineages. MDSCs appear as promising candidates for the treatment of muscular, cardiac and urological disorders. In mouse models it was shown that MDSC (Oshima *et al.*, 2005; Usas and Huard, 2007) seeded on scaffolds are able to improve vaginal regeneration by reducing fibrosis and enhancing epithelial tissue formation (Ho *et al.*, 2009). Furthermore, MSC administration after vaginal distension in rats can lead to recovery of urinary continence (Dissaranan *et al.*, 2014), and increase the number of stem cells homing to the injured region (Cruz *et al.*, 2012).

No relevant studies in human are reported, and these therapies need to be considered completely experimental at the moment.

Stem cells and erectile dysfunction

Penile erection is a neurovascular response involving an increase in arterial inflow, relaxation of corpora smooth muscles, and a reduction in venous outflow. Erectile dysfunction can be caused by conditions such as diabetes, neuropathies, cavernous nerve injuries, neurotransmitter imbalances, vascular diseases, inflammation diseases (Peyronie disease) or surgery (Wespes *et al.*, 2002). Although no clinical trials have been completed, many preclinical studies have been conducted, mainly in mouse and rat models. The majority of the studies involved MSC, neural crest stem cells, ESC, endothelial progenitor cells and MDSC. The results obtained indicated that the stem cell injections brought benefits in terms of restored erectile function and penile physiology (Zhang *et al.*, 2012); interestingly, the improvement in erectile function seems to be due to the paracrine factors secreted by the injected cells (cytoprotective, anti-fibrotic and anti-apoptotic molecules), rather than direct grafting/differentiation (Albersen *et al.*, 2010). No information is available on the duration of the beneficial effects. The only preclinical study involving patients was carried out with intracavernosal injections of umbilical cord blood stem cells in seven patients with diabetic-erectile dysfunction: patients reported an improvement in penile erection and an increase in penile rigidity, especially when in combination with PDE5 inhibitors; the duration of the effect was variable but after a few months all patients experienced a regression (Bahk *et al.*, 2010).

Taken together, these observations show how stem cell treatment is a promising tool to restore normal erectile function, but these applications need further development to overcome the limitations shown in the preclinical studies.

Conclusion

After reviewing the existing literature, the authors of this paper conclude that, to date, there are no stem cell-based therapies available to the larger public and outside of clinical trials directed at ameliorating or solving reproductive medicine issues. Specifically, there are no proven stem cell based means to improve reproductive function, either by producing functional gametes *in vitro*, or stimulating the resident stem cell

population (were it confirmed as being present in our species) in the ovary to elicit *de novo* oocyte production.

A promising avenue of research includes the development of therapies from adult stem cells of limited potency in the treatment of reproductive tract alterations, such as erectile dysfunction or damaged endometrial lining. Again, the studies reported in the literature are mostly based on animal models, and the few reports involving patients are still preliminary.

The current state of the art in stem cell and reproductive medicine mandates that patients and physicians should be wary of unfounded claims of improvement of existing medical conditions; at the moment, stem cell treatment for reproductive diseases and alteration is not feasible.

Authors' roles

R.V., C.E., B.H., K.S., C.S., A.M.M.v.P., A.V., F.Z.: conception and design, literature search, manuscript writing and revision, final approval of the manuscript.

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Conflict of interest

None declared.

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