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Stem cell applications for pathologies of the urinary bladder

MousaNA*et al*. Stem cells for urinary bladder pathologies

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**Abstract**

New stem cell based therapies are undergoing intense research and are widely investigated in clinical fields including the urinary system. The urinary bladder performs critical complex functions that rely on its highly coordinated anatomical composition and multiplex of regulatory mechanisms. Bladder pathologies resulting in severe dysfunction are common clinical encounter and often cause significant impairment of patient’s quality of life. Current surgical and medical interventions to correct urinary dysfunction or to replace an absent or defective bladder are sub-optimal and are associated with notable complications. As a result, stem cell based therapies for the urinary bladder are hoped to offer new venues that could make up for limitations of existing therapies. In this article, we review research efforts that describe the use of different types of stem cells in bladder reconstruction, urinary incontinence and retention disorders. In particular, stress urinary incontinence has been a popular target for stem cell based therapies in reported clinical trials. Furthermore, we discuss the relevance of the cancer stem cell hypothesis to the development of bladder cancer. A key subject that should not be overlooked is the safety and quality of stem cell based therapies introduced to human subjects either in a research or a clinical context.

**Key words:** Stem cells; Urinary bladder; Urethra; Incontinence; Bladder reconstruction; Cancer stem cells; Safety

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**Core tip:** This article reviews the current status of the stem cell based therapies in the field of treating urinary tract pathologies. We provide a particular focus on bladder reconstruction models that were based on stem cellular therapies. Stress incontinence and voiding dysfunction represent common clinical problems that could benefit from advancing the stem cell field. A brief highlight is given to bladder cancer stem cells and therapeutic value that could arise from controlling their behavior. We also note the pressing need for more robust regulations and quality guidelines to prevent the transfer of research findings prematurely into routine clinical practice.

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**Introduction**

The bladder and urethra compose the lower urinary tract. Pathologies of this part of the urinary system are common and clinically significant. Due to the bladder role as a versatile and dynamic reservoir of urine, it has unique structural and regulatory mechanisms that need to be thoroughly considered when cell and tissues engineering based therapies are thought of. Its complex mucosal-muscular-neural elements work together efficiently to produce a synchronized effortless urinary filling and emptying process. For instance, the bladder mucosa hosts a particular type of epithelium,“ the transitional epithelium or urothelium”, with cellular, intercellular and architectural features that make it expansible, durable, and resilient to life long persistent irritation by urine[[1](#_ENREF_1)]. Additionally, the detrusor muscle that surrounds the mucosa has a peculiar pattern of fibre arrangement to enable instant and complete emptying of the bladder once it is contracted. It can adjust to chronic outflow obstruction by hypertrophy and hyperplasia[[2](#_ENREF_2)]. The neurological control of the bladder and its urethral sphincter is compound by autonomic, sensory and motor neural interaction[[3](#_ENREF_3)]. Multiple other hormonal and metabolic factors contribute to the integrity of the bladder function. Disturbance in one or more of these elements results in dysfunction in the vital process of urination which could significantly impair a patient’s quality of life and may, in severe cases, cause disability or even death. Surgical removal of the bladder due to cancer or end stage benign disease necessitates the replacement of its function using alternative mechanisms, of which, stem cell based engineered tissue is proposed as a possible future strategy.

**Considerations in the Applicability of Stem Cell Therapy**

Stem cells are classically defined by their self-renewal based long-term survival and their flexible fate, being able to differentiate into other cell types. However, as simple as the concept seems to be, the accurate identification, isolation and transplantation of stem cells could be more intricate than originally thought. For example, the functional distinction between stem cells and their progenitor cells remains ambiguous[[4](#_ENREF_4),[5](#_ENREF_5)]. This should be taken in consideration when planning stem cell therapies; a successful therapy may require other progenitor cells to turn on differentiation and proliferation signals of the parent stem cell. As well, the transition of the stem cell between quiescent and active stages is a dynamic process that challenges the accurate prediction of stem cell behaviour in different environments[[6](#_ENREF_6)]. The control of stem cell niche and microenvironment appears to be fundamental in the long-term success of stem cell therapy. Therefore, studying the stem cell niche could identify important biochemical and mechanical cues that need to be provided in addition to the cellular component to bridge the variability gap between the donor and host stem cells’ niche to allow their effective engraftment and proper function *in vivo*[[7](#_ENREF_7)].

**Bladder Adult Stem cells**

The ability to use someone’s cells to repair another area of one’s own body could indeed reduce risks of infectivity and immunogenicity. For that, adult stem cells maintained its key position among other stem cell types[[8](#_ENREF_8)]. Tumorigenicity is also of much less concern. However, adult stem cells could be of less potential for long term survival due to inherent cell aging concerns[[9](#_ENREF_9)]. Various adult stem cell types have been used in treating bladder dysfunction including muscle; bone marrow and adipose tissue derived stem cells[[10](#_ENREF_10)]. In addition, urine stem cells have been isolated and differentiated into specialized cells to offer a readily accessible stem cell source for various applications[[11](#_ENREF_11)]. Although a general depot of adult stem cells exists in fat and bone marrow, however, lineage specific stem cells are believed to dominate in specific organs such as skin and cornea[[12](#_ENREF_12),[13](#_ENREF_13)].

The urothelial adult stem cells are thought to be slow cycling *in vivo* (3-6 months), clonogenic, highly proliferative and located in protected sites. These cells are commonly identified by their localization in the basal layer of the urothelium, and by being label-retaining cells with high expression of β-4 integrin[[14](#_ENREF_14),[15](#_ENREF_15)]. The identification and isolation of these cells are important for tissue engineering of urothelium-lined organs including bladder, urethra and ureters.

**Clinical Applications of Stem Cells in Bladder Pathologies**

***Stem cells for urinary bladder replacement***

Following cystectomy for benign or malignant bladder pathologies, bladder replacement or reconstruction is a critical step for maintaining patient’s life. Whether ureterosigmoidostomy, ileal conduit or orthotopic neobladder are used for reconstructing a new urinary reservoir, significant morbidity and mortality often occur due to the incorporation of intestinal segment into the urinary tract. This could result in recurrent urinary tract infection, metabolic and electrolyte disturbance, mucous retention and anastomotic site cancer. Moreover, the patient is left to deal with either an external draining bag through an opening on the skin called stoma or self catheterization with no external bag, both of which could interfere with body image and daily activities. Current in situ surgical bladder constructs are also unable to contract and squeeze the urine through the urethra since it lacks the muscle layer and the patient needs to adapt to techniques to push urine out such as contracting the abdominal muscles[[16](#_ENREF_16)-18]. Therefore, seeking new therapies to provide optimal bladder reconstruction is of utmost clinical importance.

Ideally, a perfect bladder reconstruct should be made of low immunogenic or autologous tissue that contains all anatomical layers of the bladder wall (mucosa, submucosa and muscle layer). It should be designed to mimic the detrusor muscle mechanics and to provide significant dispensability. It has also to provide similar urothelial mucosal barrier and ultimately should be incorporating functioning neuronal elements. Accordingly, advanced and complex tissue engineering and regenerative models are required. Thus far, there has been no such comprehensive efficient model; however preliminary studies are ongoing worldwide to achieve such goals.

Recently, tissue engineering using cell seeded scaffolds has been investigated in urinary bladder bioengineering studies[[19](#_ENREF_19)]. This method includes the seeding of a scaffold with autologous bladder muscle and epithelial cells. The use of autologous cells, however, may not be available as in cases of cancer[[20](#_ENREF_20)] or benign end-stage bladder diseases[[21](#_ENREF_21)]. Alternatively, stem cells can be derived from other sources including adipose tissue, bone marrow or amniotic fluid cells. They can be seeded on scaffolds and transplanted for *in vivo* differentiation. However, current data shows that such differentiation occurs only in a small percentage of the delivered cells[[22](#_ENREF_22)]. Another method is to differentiate stem cells into urothelial and smooth muscle cells *in vitro*. Stem cells have shown a good potential for urothelial differentiation. This was achieved by using either conditioned medium[[23](#_ENREF_23),[24](#_ENREF_24)] with or without growth factors such as all-transretinoic acid[[25](#_ENREF_25)]. Both direct culture (seeding stem cells with urothelial cells) and indirect co-culture (using trans-well system) have been attempted with variable outcome, though cell-to-cell contact in direct culture appears to be an enhancing factor for stem cell differentiation[[26](#_ENREF_26),[27](#_ENREF_27)]. Stem cell differentiation into smooth muscle cells is more feasible and can be achieved either by chemical induction[[28](#_ENREF_28)] or co-culture with smooth muscle cells[[22](#_ENREF_22)].

 Induced pluripotent stem cells (iPS) reprogrammed from adult tissue such as skin fibroblasts, urinary tract stromal cells and urine-derived cells have been also used and were subsequently differentiated into urothelial and smooth muscles[[29-31](#_ENREF_29)]. However, urinary tract-derived iPs cells are believed to be of superior differentiation properties into other sources, which should emphasize the epigenetic differences between individual iPs cell lines and stress on the importance of organ-specific iPs cells for tissue-specific studies[[29-31](#_ENREF_29)].

***Stem cells for voiding dysfunction***

Voiding dysfunction (VD) can affect the patient’s quality of life and interfere with social activities. It can be manifested clinically as various disorders of urine storage or emptying. Current therapies of VD are generally inadequate and often fail to target the actual pathophysiology of the disease. Stem cell therapies have been also investigated in this field and were shown to cause some positive response either due to differentiation or more likely due to indirect paracrine effect associated with the release of growth factors and cytokines. The later mechanism could lead to modulation of local and systemic inflammatory responses and mobilization, stimulation and differentiation of native stem cells in addition to the enhancement of vascularization of regenerating tissues and reduction of fibrosis[[32](#_ENREF_32)]. A variety of stem cell types have been explored for the treatment of VD, including bone marrow, skeletal muscle and adipose derived stem cells[[33-35](#_ENREF_33)]. Adipose stem cells was most popular due to their easy harvest, high yield of stem cells and easier smooth muscle differentiation compared to other types[[35](#_ENREF_35)]. They could improve VD in animal models of bladder overactivity or hypoactivity associated with different etiologies such as diabetes mellitus, post radiation and hyperlipidemia[[32](#_ENREF_32),[35](#_ENREF_35)]. Nevertheless, more studies are needed to determine the efficiency of such therapy, the form of transplanted cells (unmodified or differentiated cells), the method of cell delivery (systemic or local injection into the bladder) and the appropriate cellular dose (number of cells per injection and frequency of treatment).

## Stem cells for stress incontinence

Stress urinary incontinence (SUI) is a widespread disorder, particularly in women[[36](#_ENREF_36)], due to inherent predisposing anatomical and physiological factors specific to the female urethra. In addition, SUI is commonly initiated or aggravated by female specific physiological stages such as pregnancy, vaginal birth and menopause or pathological conditions such as uterine fibroid tumors[[37](#_ENREF_37)]. The current treatment of SUI relies on pelvic floor exercise in mild cases and surgical interventions in severe cases. The surgical methods aim to provide support to the urethra using various artificial tapes or autologous tissue slings. Each one of these methods has its adverse effects and post-operative complications[[38](#_ENREF_38),[39](#_ENREF_39)]. More recently, the injection of bulking agents around or through the urethra to treat SUI has gained some popularity. Many agents have been used with variable success rates and complications were reported. Collagen, fat, Teflon, silicon or carbon coated beads are common examples of various agents employed[[40](#_ENREF_40)]. A recent systematic review in the Cochrane database showed that current data is insufficient to prove benefit of these therapies especially that saline injection was of similar effectiveness to bulking agents, with some reports of serious side effects associated with some of these agents[[41](#_ENREF_41)]. Consequently, stem cell therapies appeared as the next generation of therapies in SUI, which gained recent attention. Animal studies showed potential benefit in treating SUI. The animal model of SUI is usually induced by cutting the pudendal nerves. Muscle derived stem cells have been the most widely applied source and are believed to provide stem cells that are able to differentiate into committed striated muscle cells more than other stem cell sources. However, many other sources have been attempted with comparable success. To evaluate the success of stem cell therapy in SUI animal models, multiple outcome measures were used including leak point pressure, intra-bladder pressure, maximum bladder volume, urethral functional length, max urethral closure pressure, morphological examination of sphincter muscle and matrix[[42](#_ENREF_42)].

Adipose derived stem cells have been injected in rat models of stress incontinence using intravenous or trans-urethral routes, and showed significant improvement in terms of increased elastin content and voiding function measured by cystometry[[43](#_ENREF_43)]. Adding nerve growth factor and poly (lactic-co-glycolic) acid to the adipose stem cells when injected in the rat urethral sphincter improved stem cell proliferation *in vivo* and that was increasing in a dose dependant pattern. Such factors appear to improve stem cell survival and functional performance of the urethra compared to using adipose stem cells alone[[44](#_ENREF_44)]. Furthermore, human amniotic fluid stem cells seem to be of potential benefit and favourable safety profile in restoring normal urethral function in the animal models of SUI due to their low immunogenicity and tumorigenicity[[45](#_ENREF_45)]. A triple stem cell therapy approach used human amniotic stem cells that were processed to the stage of early differentiation into three lineages *in vitro* (myogenic, neurogenic and endothelial). This approach was able to improve SUI signs in the animal model compared to using only one or two types of differentiated cells[[46](#_ENREF_46)]. A combination of gene therapy strategy by inducing urine derived stem cells to over express VEGF showed improvement of the sphincter composition especially the nerve fibres, muscle cells and vascularisation[[47](#_ENREF_47)].

The reconstruction of stem cell tissue engineered based slings to support the urethra was also investigated. A silk scaffold covered with bone marrow derived mesenchymal stem cell sheet has been implanted as a sling to support the rat urethra showing a better matrix deposition compared to using a silk sling alone[[48](#_ENREF_48)]. Likewise, adipose tissue derived stem cells and silk fibroin microspheres were combined together and they were able to retain improvement in SUI for longer duration than the silk fibroin microspheres alone[[49](#_ENREF_49)]. Following animal studies, a number of clinical trials have been attempted; examples are shown in Table 1.

***Urinary sources and applications of iPS***

iPS can be generated from somatic cells by inducing the expression of different transcription factors, originally Oct4, Sox2, Klf4 and Myc (OSKM). The induced pleuripotent stem (iPS) cells have been considered as the stem cell that could offer several advantages over the adult stem cells while maintaining the plasticity of embryonic stem cells[[50](#_ENREF_50)]. However, close examination of their possible adverse outcomes, such as the reported immunogenicity, should be undertaken prior to wide application[[51](#_ENREF_51)]. The urinary bladder and prostate cells were also reprogrammed successfully to iPS cells, offering a new method of the urinary system disease modeling[[29](#_ENREF_29)]. Likewise, urine cells were reprogrammed into iPS cells that were differentiated successfully into cardiac cells *in vitro*[[52](#_ENREF_52)]. Moreover, a model of personalized medicine was introduced by reprogramming urine cells of a haemophilia A patient into liver cells that express the traits of the disease[[53](#_ENREF_53)].Conversely, the urinary bladder cells were obtained from iPS cells *in vitro*[[54](#_ENREF_54)]. Therefore, theoretically, the use of urine cells to produce urinary bladder wall cells for bladder reconstruction purposes would be attractive as they are originating in the same system[[55](#_ENREF_55)].

***Bladder cancer stem cells***

On the other end of the stem cell paradigm, adult cancer stem cells (CSC) are implicated in cancer development, progression and recurrence after conventional therapy. CSC are practically identified and confirmed by their ability to initiate tumours in immunocompromised mice and by their *in vitro* clonogenicity. The cancer stem cell theory was a build up on older clinical observations that related cancer to stemness[[56](#_ENREF_56)]. Starting with blood cancers that showed strong representation of the cancer stem cell model[[57](#_ENREF_57)], most solid cancers were studied for their CSC population[[58-62](#_ENREF_58)]. Several groups were able to isolate cells with CSC features from urinary bladder cancers. These cells were identified by several markers as being CD44+, negative for EMA, high expression of 67LR, low expression of CD66C, strong association with ALDH1A1 expression or as a side population based on the efflux of the Hoechst dye[[63](#_ENREF_63),[64](#_ENREF_64)]. It appears that CD44+ is one of the reproducible markers and its expression on bladder CSC was commonly supported by several groups[[65](#_ENREF_65)].

The value of such findings is to tailor specific target based therapies. Unlike traditional bladder cancer therapies, a cancer stem cell targeted therapy is postulated to be able to eradicate the resistant cancer initiating cells and minimize risk of recurrence. It is also important to understand bladder carcinogenesis and recognize how risk factors initiate cancer. For instance, activation of the transcription factor stat3 led to transformation of CK14+ urothelial stem cells into invasive cancer cells in a stat3 transgenic mouse model[[66](#_ENREF_66)]. Also, arsenic could transform normal prostate stem cells into cancer stem cells; this may explain its known detrimental effect on the urogenital system[[67](#_ENREF_67)].

One of the promising molecular targets include the CD47 which is an integrin associated protein known for its “do not eat me” effect on the cancer cell, leading to less recognition by cell mediated immunity especially macrophages. The CD47 was found to be significantly more expressed in CSC compared to normal cells[[68](#_ENREF_68)]. A CD47 antibody labeled was fluorescence was also used to distinguish human bladder cancer cells from normal cells with a specificity of more than 90%[[69](#_ENREF_69)]. A number of clinical trials have started to use the antibody against CD47 as a model of immunotherapy in leukemia and solid tumors[[70](#_ENREF_70)]. The bladder CSC niche and the microenvironment are also believed to significantly protect CSC from immunity and therapeutic factors and to maintain its carcinogenic behavior. Therefore, understanding the composition of this niche could highlight other potential targets for new therapies[[71](#_ENREF_71)].

***Safety and quality***

It is obvious that the use of stem cells in clinical investigations and therapeutics of bladder and urethral pathologies will be of potential importance in the near future. Nonetheless, the rapidly gained popularity of these interventions should be cautiously perceived and publicized in scientific and non-scientific media. Premature application of such therapies in human subjects without matching scientific evidence as off label treatment in research studies or their unregulated use in clinical practice should be truly concerning. In underdeveloped settings, such unregulated practices are quite common. Stem cell therapy tourism is also expanding and could expose patients to considerable health hazards[[72](#_ENREF_72)]. Factors such as sterility and viral screening of the introduced cell therapy, the lack of contaminants and toxic molecules, cell viability and cell karyotyping should be all included in stem cell clinical studies and should be reported clearly by researchers before being accepted in formal scientific publications[[73](#_ENREF_73)]. Tumorigenicity of stem cell therapies is another factor that should not be overlooked and available tumorigenicity assays should be routinely used[[74](#_ENREF_74)]. Awareness about current good manufacturing practices in stem cell therapy should be widely raised[[75](#_ENREF_75)]. Nonetheless, this would not be adequate in the absence of strict regulations enforced on using such therapies in human especially in underdeveloped settings where baseline research ethics and quality control are not well developed or monitored. Wide international collaboration is, therefore, strongly needed to come up with a global consensus and standards that allow the safe use of stem cell therapies in human subjects without ignoring the cost and training involved to implement these standards in poor settings.

**Conclusion**

Stem cell therapies will be in the clinical setting in the near future. At present, their use in bladder dysfunction and bladder reconstruction is under intense investigation. Various sources of stem cells have been attempted with comparable success. More complex tissue engineered models that mimic the original bladder anatomy are awaited. Bladder cancer stem cells are investigated as a potential target for cancer therapies.

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**References**

1 **Hicks RM**. The function of the golgi complex in transitional epithelium. Synthesis of the thick cell membrane. *J Cell Biol* 1966; **30**: 623-643 [PMID: 5971009 DOI: 10.1083/jcb.30.3.623]

2 **Andersson KE**, Arner A. Urinary bladder contraction and relaxation: physiology and pathophysiology. *Physiol Rev* 2004; **84**: 935-986 [PMID: 15269341 DOI: 10.1152/physrev.00038.2003]

3 **Fowler CJ**, Griffiths D, de Groat WC. The neural control of micturition. *Nat Rev Neurosci* 2008; **9**: 453-466 [PMID: 18490916 DOI: 10.1038/nrn2401]

4 **Garza LA**, Yang CC, Zhao T, Blatt HB, Lee M, He H, Stanton DC, Carrasco L, Spiegel JH, Tobias JW, Cotsarelis G. Bald scalp in men with androgenetic alopecia retains hair follicle stem cells but lacks CD200-rich and CD34-positive hair follicle progenitor cells. *J Clin Invest* 2011; **121**: 613-622 [PMID: 21206086 DOI: 10.1172/jci44478]

5 **Hsu YC**, Li L, Fuchs E. Transit-amplifying cells orchestrate stem cell activity and tissue regeneration. *Cell* 2014; **157**: 935-949 [PMID: 24813615 DOI: 10.1016/j.cell.2014.02.057]

6 **Li L**, Bhatia R. Stem cell quiescence. *Clin Cancer Res* 2011; **17**: 4936-4941 [PMID: 21593194 DOI: 10.1158/1078-0432.ccr-10-1499]

7 **Li L**, Neaves WB. Normal stem cells and cancer stem cells: the niche matters. *Cancer Res* 2006; **66**: 4553-4557 [PMID: 16651403 DOI: 10.1158/0008-5472.can-05-3986]

8 **Young HE**, Black AC. Adult stem cells. *Anat Rec A Discov Mol Cell Evol Biol* 2004; **276**: 75-102 [PMID: 14699636 DOI: 10.1002/ar.a.10134]

9 **Sethe S**, Scutt A, Stolzing A. Aging of mesenchymal stem cells. *Ageing Res Rev* 2006; **5**: 91-116 [PMID: 16310414 DOI: 10.1016/j.arr.2005.10.001]

10 **Kim JH**, Lee HJ, Song YS. Treatment of bladder dysfunction using stem cell or tissue engineering technique. *Korean J Urol* 2014; **55**: 228-238 [PMID: 24741410 DOI: 10.4111/kju.2014.55.4.228]

11 **Lang R**, Liu G, Shi Y, Bharadwaj S, Leng X, Zhou X, Liu H, Atala A, Zhang Y. Self-renewal and differentiation capacity of urine-derived stem cells after urine preservation for 24 hours. *PLoS One* 2013; **8**: e53980 [PMID: 23349776 DOI: 10.1371/journal.pone.0053980]

12 **Kim DS**, Cho HJ, Choi HR, Kwon SB, Park KC. Isolation of human epidermal stem cells by adherence and the reconstruction of skin equivalents. *Cell Mol Life Sci* 2004; **61**: 2774-2781 [PMID: 15549181 DOI: 10.1007/s00018-004-4288-4]

13 **Cotsarelis G**, Cheng SZ, Dong G, Sun TT, Lavker RM. Existence of slow-cycling limbal epithelial basal cells that can be preferentially stimulated to proliferate: implications on epithelial stem cells. *Cell* 1989; **57**: 201-209 [PMID: 2702690 DOI: 10.1016/0092-8674(89)90958-6]

14 **Kurzrock EA**, Lieu DK, Degraffenried LA, Chan CW, Isseroff RR. Label-retaining cells of the bladder: candidate urothelial stem cells. *Am J Physiol Renal Physiol* 2008; **294**: F1415-F1421 [PMID: 18367656 DOI: 10.1152/ajprenal.00533.2007]

15 **Zhang H**, Lin G, Qiu X, Ning H, Banie L, Lue TF, Lin CS. Label retaining and stem cell marker expression in the developing rat urinary bladder. *Urology* 2012; **79**: 746.e1-746.e6 [PMID: 22197204 DOI: 10.1016/j.urology.2011.10.051]

16 **Turedi S**, Incealtin O, Hos G. Complications associated with ureterosigmoidostomy--colon carcinoma and ascendens infection resulting in nephrectomy: a case report. *Acta Chir Belg* ; **109**: 531-533 [PMID: 19803273]

17 **Salom EM**, Mendez LE, Schey D, Lambrou N, Kassira N, Gómez-Marn O, Averette H, Peñalver M. Continent ileocolonic urinary reservoir (Miami pouch): the University of Miami experience over 15 years. *Am J Obstet Gynecol* 2004; **190**: 994-1003 [PMID: 15118628 DOI: 10.1016/j.ajog.2004.01.023]

18 **Park J**, Ahn H. Radical cystectomy and orthotopic bladder substitution using ileum. *Korean J Urol* 2011; **52**: 233-240 [PMID: 21556208 DOI: 10.4111/kju.2011.52.4.233]

19 **Orabi H**, Bouhout S, Morissette A, Rousseau A, Chabaud S, Bolduc S. Tissue engineering of urinary bladder and urethra: advances from bench to patients. *ScientificWorldJournal* 2013; **2013**: 154564 [PMID: 24453796 DOI: 10.1155/2013/154564]

20 **Donnenberg VS**, Zimmerlin L, Rubin JP, Donnenberg AD. Regenerative therapy after cancer: what are the risks? *Tissue Eng Part B Rev* 2010; **16**: 567-575 [PMID: 20726819 DOI: 10.1089/ten.TEB.2010.0352]

21 **Subramaniam R**, Hinley J, Stahlschmidt J, Southgate J. Tissue engineering potential of urothelial cells from diseased bladders. *J Urol* 2011; **186**: 2014-2020 [PMID: 21944117 DOI: 10.1016/j.juro.2011.07.031]

22 **Zhang R**, Jack GS, Rao N, Zuk P, Ignarro LJ, Wu B, Rodríguez LV. Nuclear fusion-independent smooth muscle differentiation of human adipose-derived stem cells induced by a smooth muscle environment. *Stem Cells* 2012; **30**: 481-490 [PMID: 22213158 DOI: 10.1002/stem.1023]

23 **Tian H**, Bharadwaj S, Liu Y, Ma PX, Atala A, Zhang Y. Differentiation of human bone marrow mesenchymal stem cells into bladder cells: potential for urological tissue engineering. *Tissue Eng Part A* 2010; **16**: 1769-1779 [PMID: 20020816 DOI: 10.1089/ten.tea.2009.0625]

24 **Shi JG**, Fu WJ, Wang XX, Xu YD, Li G, Hong BF, Hu K, Cui FZ, Wang Y, Zhang X. Transdifferentiation of human adipose-derived stem cells into urothelial cells: potential for urinary tract tissue engineering. *Cell Tissue Res* 2012; Epub ahead of print [PMID: 22290635]

25 **Gandhi D**, Molotkov A, Batourina E, Schneider K, Dan H, Reiley M, Laufer E, Metzger D, Liang F, Liao Y, Sun TT, Aronow B, Rosen R, Mauney J, Adam R, Rosselot C, Van Batavia J, McMahon A, McMahon J, Guo JJ, Mendelsohn C. Retinoid signaling in progenitors controls specification and regeneration of the urothelium. *Dev Cell* 2013; **26**: 469-482 [PMID: 23993789 DOI: 10.1016/j.devcel.2013.07.017]

26 **Liu J**, Huang J, Lin T, Zhang C, Yin X. Cell-to-cell contact induces human adipose tissue-derived stromal cells to differentiate into urothelium-like cells in vitro. *Biochem Biophys Res Commun* 2009; **390**: 931-936 [PMID: 19852942 DOI: 10.1016/j.bbrc.2009.10.080]

27 **Zhang M**, Peng Y, Zhou Z, Zhou J, Wang Z, Lu M. Differentiation of human adipose-derived stem cells co-cultured with urothelium cell line toward a urothelium-like phenotype in a nude murine model. *Urology* 2013; **81**: 465.e15-465.e22 [PMID: 23374843 DOI: 10.1016/j.urology.2012.10.030]

28 **Jack GS**, Zhang R, Lee M, Xu Y, Wu BM, Rodríguez LV. Urinary bladder smooth muscle engineered from adipose stem cells and a three dimensional synthetic composite. *Biomaterials* 2009; **30**: 3259-3270 [PMID: 19345408 DOI: 10.1016/j.biomaterials.2009.02.035]

29 **Moad M**, Pal D, Hepburn AC, Williamson SC, Wilson L, Lako M, Armstrong L, Hayward SW, Franco OE, Cates JM, Fordham SE, Przyborski S, Carr-Wilkinson J, Robson CN, Heer R. A novel model of urinary tract differentiation, tissue regeneration, and disease: reprogramming human prostate and bladder cells into induced pluripotent stem cells. *Eur Urol* 2013; **64**: 753-761 [PMID: 23582880 DOI: 10.1016/j.eururo.2013.03.054]

30 **Xue Y**, Cai X, Wang L, Liao B, Zhang H, Shan Y, Chen Q, Zhou T, Li X, Hou J, Chen S, Luo R, Qin D, Pei D, Pan G. Generating a non-integrating human induced pluripotent stem cell bank from urine-derived cells. *PLoS One* 2013; **8**: e70573 [PMID: 23940595 DOI: 10.1371/journal.pone.0070573]

31 **Osborn SL**, Kurzrock EA. Production of urothelium from pluripotent stem cells for regenerative applications. *Curr Urol Rep* 2015; **16**: 466 [PMID: 25404180 DOI: 10.1007/s11934-014-0466-6]

32 **Kim JH**, Lee SR, Song YS, Lee HJ. Stem cell therapy in bladder dysfunction: where are we? And where do we have to go? *Biomed Res Int* 2013; **2013**: 930713 [PMID: 24151627 DOI: 10.1155/2013/930713]

33 **Song YS**, Lee HJ, Doo SH, Lee SJ, Lim I, Chang KT, Kim SU. Mesenchymal stem cells overexpressing hepatocyte growth factor (HGF) inhibit collagen deposit and improve bladder function in rat model of bladder outlet obstruction. *Cell Transplant* 2012; **21**: 1641-1650 [PMID: 22506988 DOI: 10.3727/096368912X637488]

34 **Nitta M**, Tamaki T, Tono K, Okada Y, Masuda M, Akatsuka A, Hoshi A, Usui Y, Terachi T. Reconstitution of experimental neurogenic bladder dysfunction using skeletal muscle-derived multipotent stem cells. *Transplantation* 2010; **89**: 1043-1049 [PMID: 20150836 DOI: 10.1097/TP.0b013e3181d45a7f]

35 **Orabi HGC**, Rousseau A, Fradette J, Bolduc S. Adipose Derived Stem Cells for treatment of Lower Genitourinary Dysfunction. *J Stem Cell Res Ther* 2014; **4**: 190 [DOI: 10.4172/2157-7633.1000190]

36 **Rogers RG**. Clinical practice. Urinary stress incontinence in women. *N Engl J Med* 2008; **358**: 1029-1036 [PMID: 18322284 DOI: 10.1056/NEJMcp0707023]

37 **Waetjen LE**, Liao S, Johnson WO, Sampselle CM, Sternfield B, Harlow SD, Gold EB. Factors associated with prevalent and incident urinary incontinence in a cohort of midlife women: a longitudinal analysis of data: study of women's health across the nation. *Am J Epidemiol* 2007; **165**: 309-318 [PMID: 17132698 DOI: 10.1093/aje/kwk018]

38 **Hou JC**, Alhalabi F, Lemack GE, Zimmern PE. Outcome of transvaginal mesh and tape removed for pain only. *J Urol* 2014; **192**: 856-860 [PMID: 24735934 DOI: 10.1016/j.juro.2014.04.006]

39 **Khan ZA**, Nambiar A, Morley R, Chapple CR, Emery SJ, Lucas MG. Long-term follow-up of a multicentre randomised controlled trial comparing tension-free vaginal tape, xenograft and autologous fascial slings for the treatment of stress urinary incontinence in women. *BJU Int* 2014; Epub ahead of print [PMID: 24961647 DOI: 10.1111/bju.12851]

40 **Kerr LA**. Bulking agents in the treatment of stress urinary incontinence: history, outcomes, patient populations, and reimbursement profile. *Rev Urol* 2005; **7** Suppl 1: S3-S11 [PMID: 16985874]

41 **Kirchin V**, Page T, Keegan PE, Atiemo K, Cody JD, McClinton S. Urethral injection therapy for urinary incontinence in women. *Cochrane Database Syst Rev* 2012; **2**: CD003881 [PMID: 22336797 DOI: 10.1002/14651858.CD003881.pub3]

42 **Wu G**, Song Y, Zheng X, Jiang Z. Adipose-derived stromal cell transplantation for treatment of stress urinary incontinence. *Tissue Cell* 2011; **43**: 246-253 [PMID: 21704350 DOI: 10.1016/j.tice.2011.04.003]

43 **Lin G**, Wang G, Banie L, Ning H, Shindel AW, Fandel TM, Lue TF, Lin CS. Treatment of stress urinary incontinence with adipose tissue-derived stem cells. *Cytotherapy* 2010; **12**: 88-95 [PMID: 19878076 DOI: 10.3109/14653240903350265]

44 **Zhao W**, Zhang C, Jin C, Zhang Z, Kong D, Xu W, Xiu Y. Periurethral injection of autologous adipose-derived stem cells with controlled-release nerve growth factor for the treatment of stress urinary incontinence in a rat model. *Eur Urol* 2011; **59**: 155-163 [PMID: 21050657 DOI: 10.1016/j.eururo.2010.10.038]

45 **Kim BS**, Chun SY, Lee JK, Lim HJ, Bae JS, Chung HY, Atala A, Soker S, Yoo JJ, Kwon TG. Human amniotic fluid stem cell injection therapy for urethral sphincter regeneration in an animal model. *BMC Med* 2012; **10**: 94 [PMID: 22906045 DOI: 10.1186/1741-7015-10-94]

46 **Chun SY**, Kwon JB, Chae SY, Lee JK, Bae JS, Kim BS, Kim HT, Yoo ES, Lim JO, Yoo JJ, Kim WJ, Kim BW, Kwon TG. Combined injection of three different lineages of early-differentiating human amniotic fluid-derived cells restores urethral sphincter function in urinary incontinence. *BJU Int* 2014; **114**: 770-783 [PMID: 24841807 DOI: 10.1111/bju.12815]

47 **Liu G**, Wang X, Sun X, Deng C, Atala A, Zhang Y. The effect of urine-derived stem cells expressing VEGF loaded in collagen hydrogels on myogenesis and innervation following after subcutaneous implantation in nude mice. *Biomaterials* 2013; **34**: 8617-8629 [PMID: 23932297 DOI: 10.1016/j.biomaterials.2013.07.077]

48 **Zou XH**, Zhi YL, Chen X, Jin HM, Wang LL, Jiang YZ, Yin Z, Ouyang HW. Mesenchymal stem cell seeded knitted silk sling for the treatment of stress urinary incontinence. *Biomaterials* 2010; **31**: 4872-4879 [PMID: 20303586 DOI: 10.1016/j.biomaterials.2010.02.056]

49 **Shi LB**, Cai HX, Chen LK, Wu Y, Zhu SA, Gong XN, Xia YX, Ouyang HW, Zou XH. Tissue engineered bulking agent with adipose-derived stem cells and silk fibroin microspheres for the treatment of intrinsic urethral sphincter deficiency. *Biomaterials* 2014; **35**: 1519-1530 [PMID: 24275524 DOI: 10.1016/j.biomaterials.2013.11.025]

50 **Takahashi K**, Yamanaka S. Induction of pluripotent stem cells from mouse embryonic and adult fibroblast cultures by defined factors. *Cell* 2006; **126**: 663-676 [PMID: 16904174 DOI: 10.1016/j.cell.2006.07.024]

51 **Zhao T**, Zhang ZN, Rong Z, Xu Y. Immunogenicity of induced pluripotent stem cells. *Nature* 2011; **474**: 212-215 [PMID: 21572395]

52 **Zhou YF**, Yao JL, Yang XJ. GW25-e0242 Urine may be a preferred source to generate induced pluripotent stem cell-derived cardiomyocytes for cardiac regenerative medicine. *J Am Coll Cardiol* 2014; **64** [DOI: 10.1016/j.jacc.2014.06.301]

53 **Jia B**, Chen S, Zhao Z, Liu P, Cai J, Qin D, Du J, Wu C, Chen Q, Cai X, Zhang H, Yu Y, Pei D, Zhong M, Pan G. Modeling of hemophilia A using patient-specific induced pluripotent stem cells derived from urine cells. *Life Sci* 2014; **108**: 22-29 [PMID: 24834837 DOI: 10.1016/j.lfs.2014.05.004]

54 **Osborn SL**, Thangappan R, Luria A, Lee JH, Nolta J, Kurzrock EA. Induction of human embryonic and induced pluripotent stem cells into urothelium. *Stem Cells Transl Med* 2014; **3**: 610-619 [PMID: 24657961 DOI: 10.5966/sctm.2013-0131]

55 **Qin D**, Long T, Deng J, Zhang Y. Urine-derived stem cells for potential use in bladder repair. *Stem Cell Res Ther* 2014; **5**: 69 [PMID: 25157812 DOI: 10.1186/scrt458]

56 **Wicha MS**, Liu S, Dontu G. Cancer stem cells: an old idea--a paradigm shift. *Cancer Res* 2006; **66**: 1883-190; discussion 1883-190; [PMID: 16488983 DOI: 10.1158/0008-5472.can-05-3153]

57 **Chen K**, Huang YH, Chen JL. Understanding and targeting cancer stem cells: therapeutic implications and challenges. *Acta Pharmacol Sin* 2013; **34**: 732-740 [PMID: 23685952 DOI: 10.1038/aps.2013.27]

58 **Singh SK**, Clarke ID, Terasaki M, Bonn VE, Hawkins C, Squire J, Dirks PB. Identification of a cancer stem cell in human brain tumors. *Cancer Res* 2003; **63**: 5821-5828 [PMID: 14522905]

59 **Prince ME**, Sivanandan R, Kaczorowski A, Wolf GT, Kaplan MJ, Dalerba P, Weissman IL, Clarke MF, Ailles LE. Identification of a subpopulation of cells with cancer stem cell properties in head and neck squamous cell carcinoma. *Proc Natl Acad Sci USA* 2007; **104**: 973-978 [PMID: 17210912 DOI: 10.1073/pnas.0610117104]

60 **Eramo A**, Lotti F, Sette G, Pilozzi E, Biffoni M, Di Virgilio A, Conticello C, Ruco L, Peschle C, De Maria R. Identification and expansion of the tumorigenic lung cancer stem cell population. *Cell Death Differ* 2008; **15**: 504-514 [PMID: 18049477 DOI: 10.1038/sj.cdd.4402283]

61 **Todaro M**, Alea MP, Di Stefano AB, Cammareri P, Vermeulen L, Iovino F, Tripodo C, Russo A, Gulotta G, Medema JP, Stassi G. Colon cancer stem cells dictate tumor growth and resist cell death by production of interleukin-4. *Cell Stem Cell* 2007; **1**: 389-402 [PMID: 18371377 DOI: 10.1016/j.stem.2007.08.001]

62 **Chiba T**, Kita K, Zheng YW, Yokosuka O, Saisho H, Iwama A, Nakauchi H, Taniguchi H. Side population purified from hepatocellular carcinoma cells harbors cancer stem cell-like properties. *Hepatology* 2006; **44**: 240-251 [PMID: 16799977 DOI: 10.1002/hep.21227]

63 **Chan KS**, Espinosa I, Chao M, Wong D, Ailles L, Diehn M, Gill H, Presti J, Chang HY, van de Rijn M, Shortliffe L, Weissman IL. Identification, molecular characterization, clinical prognosis, and therapeutic targeting of human bladder tumor-initiating cells. *Proc Natl Acad Sci USA* 2009; **106**: 14016-14021 [PMID: 19666525 DOI: 10.1073/pnas.0906549106]

64 **Chan KS**, Volkmer JP, Weissman I. Cancer stem cells in bladder cancer: a revisited and evolving concept. *Curr Opin Urol* 2010; **20**: 393-397 [PMID: 20657288 DOI: 10.1097/MOU.0b013e32833cc9df]

65 **Ho PL**, Kurtova A, Chan KS. Normal and neoplastic urothelial stem cells: getting to the root of the problem. *Nat Rev Urol* 2012; **9**: 583-594 [PMID: 22890301 DOI: 10.1038/nrurol.2012.142]

66 **Ho PL**, Lay EJ, Jian W, Parra D, Chan KS. Stat3 activation in urothelial stem cells leads to direct progression to invasive bladder cancer. *Cancer Res* 2012; **72**: 3135-3142 [PMID: 22532166 DOI: 10.1158/0008-5472.can-11-3195]

67 **Tokar EJ**, Diwan BA, Waalkes MP. Arsenic exposure transforms human epithelial stem/progenitor cells into a cancer stem-like phenotype. *Environ Health Perspect* 2010; **118**: 108-115 [PMID: 20056578 DOI: 10.1289/ehp.0901059]

68 **Majeti R**, Chao MP, Alizadeh AA, Pang WW, Jaiswal S, Gibbs KD, van Rooijen N, Weissman IL. CD47 is an adverse prognostic factor and therapeutic antibody target on human acute myeloid leukemia stem cells. *Cell* 2009; **138**: 286-299 [PMID: 19632179 DOI: 10.1016/j.cell.2009.05.045]

69 **Pan Y**, Volkmer JP, Mach KE, Rouse RV, Liu JJ, Sahoo D, Chang TC, Metzner TJ, Kang L, van de Rijn M, Skinner EC, Gambhir SS, Weissman IL, Liao JC. Endoscopic molecular imaging of human bladder cancer using a CD47 antibody. *Sci Transl Med* 2014; **6**: 260ra148 [PMID: 25355698 DOI: 10.1126/scitranslmed.3009457]

70 **CIRM**. Clinical Investigation of a Humanized Anti-CD47 Antibody in Targeting Cancer Stem Cells in Hematologic Malignancies and Solid Tumors. California Institute For Regenerative Medicine 2014. Available from: URL: http: //wwwcirmcagov/our-progress/awards/clinical-investigation-humanized-anti-cd47-antibody-targeting-cancer-stem-cells

71 **van der Horst G**, Bos L, van der Pluijm G. Epithelial plasticity, cancer stem cells, and the tumor-supportive stroma in bladder carcinoma. *Mol Cancer Res* 2012; **10**: 995-1009 [PMID: 22714124 DOI: 10.1158/1541-7786.mcr-12-0274]

72 **Master Z**, Resnik DB. Stem-cell tourism and scientific responsibility. Stem-cell researchers are in a unique position to curb the problem of stem-cell tourism. *EMBO Rep* 2011; **12**: 992-995 [PMID: 21799519 DOI: 10.1038/embor.2011.156]

73 **Prockop DJ**, Olson SD. Clinical trials with adult stem/progenitor cells for tissue repair: let's not overlook some essential precautions. *Blood* 2007; **109**: 3147-3151 [PMID: 17170129 DOI: 10.1182/blood-2006-03-013433]

74 **Knoepfler PS**. Deconstructing stem cell tumorigenicity: a roadmap to safe regenerative medicine. *Stem Cells* 2009; **27**: 1050-1056 [PMID: 19415771 DOI: 10.1002/stem.37]

75 **Giancola R**, Bonfini T, Iacone A. Cell therapy: cGMP facilities and manufacturing. *Muscles Ligaments Tendons J* 2012; **2**: 243-247 [PMID: 23738304]

76 **Stangel-Wojcikiewicz K**, Jarocha D, Piwowar M, Jach R, Uhl T, Basta A, Majka M. Autologous muscle-derived cells for the treatment of female stress urinary incontinence: a 2-year follow-up of a Polish investigation. *Neurourol Urodyn* 2014; **33**: 324-330 [PMID: 23606303 DOI: 10.1002/nau.22404]

77 **Gerullis H**, Eimer C, Georgas E, Homburger M, El-Baz AG, Wishahi M, Borós M, Ecke TH, Otto T. Muscle-derived cells for treatment of iatrogenic sphincter damage and urinary incontinence in men. *ScientificWorldJournal* 2012; **2012**: 898535 [PMID: 22919359 DOI: 10.1100/2012/898535]

78 **Gräs S**, Klarskov N, Lose G. Intraurethral injection of autologous minced skeletal muscle: a simple surgical treatment for stress urinary incontinence. *J Urol* 2014; **192**: 850-855 [PMID: 24735937 DOI: 10.1016/j.juro.2014.04.005]

79 **Carr LK**, Steele D, Steele S, Wagner D, Pruchnic R, Jankowski R, Erickson J, Huard J, Chancellor MB. 1-year follow-up of autologous muscle-derived stem cell injection pilot study to treat stress urinary incontinence. *Int Urogynecol J Pelvic Floor Dysfunct* 2008; **19**: 881-883 [PMID: 18204978 DOI: 10.1007/s00192-007-0553-z]

80 **Mitterberger M**, Pinggera GM, Marksteiner R, Margreiter E, Fussenegger M, Frauscher F, Ulmer H, Hering S, Bartsch G, Strasser H. Adult stem cell therapy of female stress urinary incontinence. *Eur Urol* 2008; **53**: 169-175 [PMID: 17683852 DOI: 10.1016/j.eururo.2007.07.026]

81 **Gotoh M**, Yamamoto T, Kato M, Majima T, Toriyama K, Kamei Y, Matsukawa Y, Hirakawa A, Funahashi Y. Regenerative treatment of male stress urinary incontinence by periurethral injection of autologous adipose-derived regenerative cells: 1-year outcomes in 11 patients. *Int J Urol* 2014; **21**: 294-300 [PMID: 24033774 DOI: 10.1111/iju.12266]

82 **Kuismanen K**, Sartoneva R, Haimi S, Mannerström B, Tomás E, Miettinen S, Nieminen K. Autologous adipose stem cells in treatment of female stress urinary incontinence: results of a pilot study. *Stem Cells Transl Med* 2014; **3**: 936-941 [PMID: 24985079 DOI: 10.5966/sctm.2013-0197]

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**Table 1 Clinical studies of stem cell therapy in treating stress urinary incontinence: Origin of stem cells and duration of *in vitro* expansion**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Origin of stem cells and duration of *in vitro* expansion** | **Number of women and route of injection** | **Follow up** | **Success rate (improvement and cure)** | **Ref.** |
| Autologous muscle derived SC, expanded 8-10 d | 16 women, transurethral approach | 2 yr | Up to 75% | [[76](#_ENREF_76)] |
| Muscle derived SC | 222 men, transurethral injection | 1 yr | Up to 54%  | [[77](#_ENREF_77)] |
| Minced autologous muscle cells, no *in vitro* expansion | 35 females | 1 yr | Up to 63%. Improvement.(Clinical, diary, and ICIQ-SF scores). | [[78](#_ENREF_78)] |
| Muscle derived SC, expansion duration NA | 8 females, transurethral injection | 1 year | Significant improvement in 5 women (pad-weight, bladder diary and QOL assessment) | [[79](#_ENREF_79)] |
| Muscle derived SC, expanded *in vitro* for 7 wk | 20 females | 2 years | Significant improvement(Clinical, QOL and cystometry).Therapy based on this method is now licensed in Europe. | [[80](#_ENREF_80)] |
| ASCs combined withbovine collagen | 5 females  | 1 yr | 2 out of 5 patients were satisfied with treatment with negative cough test | [[81](#_ENREF_81)] |
| Autologous ASCs with andwithout fat | 11 male patients with post-proststectomy incontinence  | 1 yr  | 60% improvement in urine leakage, frequency and amount of incontinence in 8 patients with one patients achieve total continence | [[82](#_ENREF_82)] |