

Adult stem cells therapy for urine incontinence in women

Terapia z wykorzystaniem dojrzałych komórek macierzystych w leczeniu nietrzymania moczu u kobiet

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Abstract

The past few years brought high development in obtaining and culturing autologous adult stem cells. In this paper we review publications of experimental investigations and clinical trials of the muscle-derived cells and the application in the treatment of stress urinary incontinence among women.

Mesenchymal stem cells (MSCs) can be obtained from bone marrow but it is associated with a painful biopsy procedure. Collection of muscle-derived stem cells (MDSCs) is less harmful because the skeletal muscle biopsy is performed with a small caliber needle in local anesthesia.

The stem-based therapy could be the next step in the treatment of urinary incontinence. There are still many elements of therapy such as effectiveness or long-term side effects which need to be researched.

Key words: **adult stem cells / biopsy needle / urinary incontinence /**

Streszczenie

Ostatnie lata przyniosły znaczący rozwój w pozyskiwaniu i hodowli autologicznych komórek macierzystych. W poniższej publikacji zawarliśmy przegląd doniesień naukowych oraz badań klinicznych dotyczących macierzystych komórek mięśniowych i ich wykorzystania w leczeniu nietrzymania moczu u kobiet.

Dotychczasowa konieczność wykorzystania bolesnej procedury biopsji szpiku kostnego w celu otrzymania komórek macierzystych typu mesenchymalnego, niosła z sobą ograniczenia. Możliwość pozyskania dojrzałych komórek macierzystych mięśniowych na drodze cienko igłowej biopsji mięśnia- w znacznym stopniu ułatwia jej powszechne zastosowanie.

Terapia nietrzymania moczu w oparciu o komórki macierzyste jawi się jako kolejny krok w ewolucji leczenia nietrzymania moczu. Do wyjaśnienia pozostają jej długoterminowa skuteczność oraz ewentualne skutki uboczne.

Słowa kluczowe: **dojrzałe komórki macierzyste / biopsja igłowa /
/ nietrzymanie moczu /**

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Introduction

Urinary incontinence is a disorder observed in 200 million people worldwide. It affects mainly women, as a result of minor pelvic anatomy and sociologic factors [1]. Frequency of urinary incontinence increases with age and is also associated with the decrease of sexual hormones [2]. Escalation of symptoms is associated with the number of factors including natural delivery, heavy birth weight, vaginal procedures, high body mass index (BMI), and chronic diseases [3].

In developed countries, special attention should be paid to the costs of symptomatic therapies, i.e., uses of absorbent pads or pant liners, and surgery. It is estimated that patients with urinary incontinence in the United States (USA), 75% of whom are women, require annual expenditures of \$16 billion dollars (£9.987 billion) toward symptomatic therapies. In the United Kingdom, about £424 million (\$679 million) is spent toward this aim annually. Taking into consideration an aging trend and an increase of mean life expectancy in the developing countries, the problem of urinary incontinence is increasing [4].

Due to a range of causes leading to urinary incontinence, the following types have been established: stress urinary incontinence (SUI), urge urinary incontinence (UII), mixed urinary incontinence (MUI), and overflow incontinence. Each type requires specific therapeutic management [5]. Gynecological examination with cough test, pat test, and urodynamic test are all the diagnostic standards. A combination of all these tests enables a definitive diagnosis and a plan for proper therapeutic management for each patient [6].

The most common form of urinary incontinence, causing the highest number of diagnostic and therapeutic doubts, is stress urinary incontinence. According to the International Continence Society's (ICS's) definition, stress urinary incontinence is caused by excessive urethral mobility or intrinsic sphincter deficiency (ISD) [7].

Stress urinary incontinence therapy in women – standard management

Surgical therapy is standard management in SUI. Classic surgical techniques, i.e., Burch's method (performed with laparoscopic access) or with use of various structural profile tapes as well as a range of access methods (Tension free Vaginal Tape, Trans Obturator Tape procedures) are used depending on their indications [8]. Minimally invasive procedures, such as tissue fixation system (TFS) and TVT Secure (Gynacare®), are also novel methods [9].

Attempts using Teflon®, collagen, or silicone in paraurethral injections are also made. All the above methods are associated with some complications [10]. In case of classic colposuspension, bleeding from vascular plexuses may bring some difficulties, as it requires

advanced operational abilities of the medical personnel [11]. Erosion, observed in 3% to 10% of patients, inflammation, and, in consequence, formation of so-called "rigid urethra" are the most serious complications of tapes. Use of artificial materials for injections is associated with local allergic reactions and difficulties in urine flow. As optimal management does not exist, alternative methods are continuously sought.

Hormonal balance in a treated patient is of crucial importance before surgery. Many studies confirm an increased frequency of urinary incontinence in women of perimenopausal age. Estrogen deficiency causes decreased blood flow through the submucosal vascular plexus which increases local atrophy. For these reasons, use of local estrogenic preparations in the preoperational period is recommended [12].

Use of Stem Cells in Stress Urinary Incontinence Therapy

Recent years have brought significant development of genetic engineering techniques and, as a result, the ability to apply therapeutic use of myoblastic and fibroblastic autologous stem cells. The ability to repair lower urinary tract defects with correctly functioning host tissue, without risk of denervation or revascularization, appears to be the ideal solution [13]. In the future, it will enable an expansion of qualifications for women with SUI, who, for various reasons, cannot undergo surgical treatment.

Tissue stem cells are obtained from the striated muscles, adipose tissue, and bone marrow. These cells have the ability to differentiate into properly functioning smooth and striated muscle tissue [14]. With administration into the paraurethral region, they contribute to the increase of urethral occlusion pressure and the restoration of normal urinary continence.

Use of Various Cell Populations

Bone marrow is a source of hematopoietic stem cells and Bone marrow is a source of hematopoietic stem cells and mesenchymal stem cells (MSCs). MSC have an ability to differentiate, both in vitro and in vivo, into adipocytes, osteocytes, chondrocytes, and muscular cells, as well as non-mesenchymal cells (endodermic or nervous cells) [15]. Unfortunately, their collection technique is associated with the painful bone marrow biopsy and, as a result, will have some limitations [16].

The above-referenced encourages scientists to use so-called "mature" stem cells that can be collected from striated muscle (muscles of arm or thigh) with biopsy under local anesthesia in therapeutic SUI models and enables the gain of muscle-derived stem cells (MDSCs) and adipose-derived stem cells (ADSCs). Based on the promising results seen in MDSC therapy for other conditions, such as myocardial infarction as described by Oshima, among others, studies focusing on treatment of urinary incontinence are now being done [17]. Some of the obstacles to overcome include survival of cells following a transplant, their ability to integrate with the urethral sphincter muscle, and their innervation.

Qu and associates from Pittsburgh, Pennsylvania, USA, has evaluated ways to improve the survival of transplanted myoblasts in the treatment of Duchenne muscular dystrophy. It has been observed that myoblasts expressing an inhibitor of the inflammatory cytokine, IL-1, are associated with a higher rate of survival [18].

According to Yokoyama et al., MDSCs present some advantages in SUI treatment. Collecting them from and using them in the same patient practically eliminates the risk of allergic or immunologic reaction that could occur if using injections with collagen [19].

The next important element is the ability of the above-mentioned cells to form junctions with postmitotic multinucleated myotubes, which differ from the fibroblasts that do not present the tendency for migration and, consequently, urine retention [20]. Their most important trait, however, is their ability to form myotubes and myofibers with reinnervation that enables them to build into the structure of the damaged muscle as a physiologically functional unit [21]. This finding was confirmed in studies on rat models by the Chermansky et al., which observed junctions between MDSCs in striated muscles layer 4 weeks after injection. Evaluation of leak point pressure (LPP) value in the group with MDSCs was significantly higher than the control group of rats with denervation [22].

MDCs (muscle-derived cells) and fibroblasts were used in research by Yokoyama on rat model. Although both MDCs and fibroblasts injection increased the LPP in a SUI, only MDCs significantly improved urethral muscle strip contractility [23]. Chancellor and associates evaluated lidocaine's influence on MDCs in the in vitro and in vivo conditions [24]. Earlier studies suggested neurotoxicity of this substance. Negative influences on corneal epithelial cells, urothelial cells, pneumocytes, and chondrocytes were also described. The study's results, therefore, eliminated the use of lidocaine as local anaesthesia. Studies on the rat model did not confirm the above theses and LPP values after therapy with MDCs were not affected by use of analgesic substance.

Adipose-derived stem cells are also used in SUI therapy. In 2006, Zeng showed an increase in LPP pressure and improved urethra function on the rat model after use of ADSCs [25]. Becker and Jakse point out that, despite significant developments in tissue engineering, there is still lack of scientific evidence relating to the possibility of differentiating autologous stem cells into urethral mucosa (urothelium), neither in vivo nor in vitro [26].

The unexplained risk of carcinogenesis associated with the ability for self-multiplication remains a crucial element of stem cell therapy. Mature stem cells are endowed with unpredictable plasticity and, as progenitors, may differentiate into mature cells of different tissue types [27, 28, 29]. Bone marrow-derived stromal cells (BM-SCs) characterize with particularly high plasticity. Their ability to differentiate into the non-haematopoietic cells/lineages was shown in the in vivo and in vitro studies [30, 31, 32].

Clinical Trials

Results of studies on the animal model encouraged experimentation with human.

The first reports of these studies were published in 2004 and 2007. The application of MDSCs in the therapy of stress urinary incontinence was presented in the original study that appeared in *The Lancet* in June 2007. The purpose of the publication was to compare the method in question with other treatment strategies. However, due to certain doubts as to the actual existence of such a trial, the editors of the journal decided to retract the article [33].

A study on the use of "pure" MDSC cells in SUI therapy involving eight women was published in 2008. Carr et al. used biopsy needles for collection of samples [34]. MDSCs isolated in the laboratory were administered to patients under local anaesthesia via one of three methods as follows: three women

obtained transurethral injection with 8 mm needles in the 3rd and 9th hours, two patients were injected transurethrally with 10 mm needles in 4 places (the 3rd, 6th, 9th, and 12th hours); and the remaining three were injected in 4 places but 25G needles were used. The patients in the third group received additional injections three months after the first procedure, again in the 4 locations described above.

Mean observation time was 17 months. Among five women available for follow-up study, one patient presented with totally controlled urination. Two more patients reported significant improvement and received further injections 4 and 8 months after their first procedures. No adverse reactions to the treatment were described.

Results of the above studies are very encouraging. Multicenter studies, the use of various cell types, and verification of their administration procedures enable medical scientists to form objective conclusions on the efficacy of using stem cell therapy for stress urinary incontinence in the future.

Stem cell-based therapy is not merely a "wish of the future." This type of therapy is currently being used as a promising alternative treatment for many diseases, including myocardial infarction and Duchenne muscular dystrophy. Due to legal and ethical challenges, the use of embryonic stem cells is not widely accepted. Therefore, the discovery of mature tissue stem cells has helped researchers avoid many reservations [35].

However, several unexplained elements of therapy exist, including its effectiveness, long-term side effects, and the stem cells' ability for carcinogenesis.

Yoo and associates from Wake Forest, North Carolina USA, presented a report on current tendencies in the use of cell therapy and previous achievements in explant culture, also in urogynecology, at the 38th Meeting of the International Continence Society (ICS) in Cairo, Egypt, in 2008. There, he declared completed transplantation of the urethra, vagina, ureter, and the bladder. Yoo's research team is currently working on the development of kidney, blood vessel, and heart valve tissues with the use of biological scaffolds.

There are several on-going clinical trials and investigations, including the work of our gynecology department that will help further define the future role of cell-based therapies for urinary incontinence.

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References

1. Griebling T. Urinary incontinence in the elderly. *Clin Geriatr Med.* 2009, 25, 445-457.
2. Cody J, Richardson K, Moehrer B, [et al.]. Oestrogen therapy for urinary incontinence in post-menopausal women. *Cochrane Database Syst Rev.* 2009, CD001405.
3. Subak L, Wing R, West D, [et al.]. Weight loss to treat urinary incontinence in overweight and obese women. *N Engl Med.* 2009, 360, 481-490.
4. Subak LL, Brubaker L, Chai T, [et al.]. High Costs of Urinary Incontinence Among Women Electing Surgery to Treat Stress Incontinence. *Obstet Gynecol.* 2008, 111, 899-907.

5. Holroyd-Leduc J, Tannerbaum C, Thorpe K, [et al.]. What type of urinary incontinence does this woman have? *JAMA*. 2008, 299, 1446-1456.
6. Abed H, Rogers R. Urinary incontinence and pelvic organ prolapse: diagnosis and treatment for the primary care physician. *Med Clin North Am*. 2008, 92, 1273-1293.
7. Abrams P, Cardozo L, Fall M, [et al.]. The standardization of terminology of lower urinary tract function: report from the standardization sub-committee of the International Continence Society. *NeuroUrol Urodyn*. 2002, 21, 167-178.
8. Rowner E, Lebed B. Stress incontinence surgery: which operation when? *Curr Opin Urol*. 2009, 19, 362-367.
9. Crivellaro S, Smith J 3rd. Minimally Invasive Therapies for Female Stress Urinary Incontinence: the Current Status of Bioinjectables/New Devices (Adjustable Continence Therapy, Urethral Submucosal Collagen Denaturation by Radiofrequency) *Scientific World Journal*. 2009, 9, 466-478.
10. Kotb A, Campeau L, Corcos J. Urethral bulking agents: Techniques and outcomes *Curr Urol Rep*. 2009, 10, 396-400.
11. Starkman J, Scarpero H, Dmochowski R. Emerging periurethral bulking agents for female stress urinary incontinence: is new necessarily better? *Curr Urol Rep*. 2006, 7, 405-413.
12. Moehrer B, Hextall A, Jackson S. Oestrogens for urinary incontinence in women. *Cochrane Database Syst Rev*. 2003, CD001405.
13. You R, Yoo J, Atala A. Restoration of functional motor units in a rat model of sphincter injury by muscle precursor cell autografts. *Transplantation*. 2003, 76, 1053-1060.
14. Buján J, Pascual G, Corrales C, [et al.]. Muscle-derived stem cells in tissue engineering: defining cell properties suitable for construct design. *Histol Histopathol*. 2005, 20, 891-899.
15. Shukla D, Box G, Edwards R, [et al.]. Bone marrow stem cells for urologic tissue engineering. *World J Urol*. 2008, 26, 341-349.
16. Pittenger M, Mackay A, Beck S, [et al.]. Multilineage potential of adult human mesenchymal stem cells. *Science*. 1999, 284, 143-147.
17. Oshima H, Payne T, Urish K, [et al.]. Differential Myocardial Infarct Repair with Muscle Stem Cells Compared to Myoblasts. *Mol Ther*. 2005, 12, 1130-1141.
18. Qu Z, Balkir L, van Deutekom J, [et al.]. Development of Approaches to Improve Cell Survival in Myoblast Transfer Therapy. *J Cell Biol*. 1998, 142, 1257-1267.
19. Yokoyama T, Yoshimura N, Dhir R, [et al.]. Persistence and survival of autologous muscle derived cells versus bovine collagen as potential treatment of stress urinary incontinence. *J Urol*. 2001, 165, 271-276.
20. Kwon D, Kim Y, Pruchnic R, [et al.]. Periurethral cellular injection: comparison of muscle-derived progenitor cells and fibroblasts with regard to efficacy and tissue contractility in an animal model of stress urinary incontinence. *Urology*. 2006, 68, 449-454.
21. Huard J, Yokoyama T, Pruchnic R, [et al.]. Muscle-derived cell-mediated ex vivo gene therapy for urological dysfunction. *Gene Ther*. 2002, 9, 1617-1626.
22. Chermansky C, Tarin T, Kwon DD [et al.]. Intraurethral muscle-derived cell injections increase leak point pressure in a rat model of intrinsic sphincter deficiency. *Urology*. 2004, 63, 780-785.
23. Yokoyama T, Yoshimura N, Dhir R, [et al.]. Persistence and survival of autologous muscle derived cells versus bovine collagen as potential treatment of stress urinary incontinence. *J Urol*. 2001, 165, 271-276.
24. Kim D, Jankowski R, Pruchnic R, [et al.]. In Vitro and In Vivo Effect of Lidocaine on Rat Muscle-derived Cells for Treatment of Stress Urinary Incontinence. *Urology*. 2009, 73, 437-441.
25. Zeng X, Jack G, Zhang R. Treatment of SUI using adipose derived stem cells: Restoration of urethral function. *J Urol*. 2006, 175, 291(abstract).
26. Becker Ch, Jakse G. Stem Cells for Regeneration of Urological Structures. *Eur Urol*. 2007, 51, 1217-1228.
27. Sell S. Stem cell origin of cancer and differentiation therapy. *Crit Rev Oncol Hemato*. 2004, 51, 1-28.
28. Vogel G. Can old cells learn new tricks? *Science*. 2000, 287, 1418-1419.
29. Alison M, Poulosom R, Forbes S, [et al.]. An introduction to stem cells. *J Pathol*. 2002, 197, 419-423.
30. Jiang Y, Jahagirdar B, Reinhardt R, [et al.]. Pluripotency of mesenchymal stem cells derived from adult marrow. *Nature*. 2002, 418, 41-49.
31. Guo Y, Lubbert M, Engelhardt M. CD32-hematopoietic stem cells: current concepts and controversies. *Stem Cells*. 2003, 21, 15-20.
32. Ratajczak M, Kucia M, Reza R, [et al.]. Stem cell plasticity revisited: CXCR4-positive cells expressing mRNA for early muscle, liver and neural cells 'hide out' in the bone marrow. *Leukemia*. 2004, 18, 29-40.
33. Kleinert S, Horton R. Retraction- autologous myoblasts and fibroblasts versus collagen for treatment of stress urinary incontinence in women: a randomized controlled trial. *Lancet*. 2008, 372, 789-790.
34. Carr L, Steel D, Steel S, [et al.]. 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.
35. Ratajczak M, Machaliński B, Czajka R, [et al.]. Physiological and pathological consequences of a presence of germ line stem cells in adult tissues. *Ginekol Pol*. 2009, 80, 935-941. Polish.