Review Article

Urine-Derived Stem Cells: The Present and the Future

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Stem cell research provides promising strategies in improving healthcare for human beings [1, 2]. Tremendous research has been conducted on two types of stem cells: the pluripotent stem cells (PSCs) and the somatic stem cells. The most commonly investigated PSCs include embryonic stem cells (ESCs) and induced pluripotent stem cells (iPSCs). ESCs are usually derived from the inner cell mass of early embryos which could be proliferated for a long term and differentiated into cell types of all three germ layers in vitro and in vivo [3]. Based on the research of ESCs, mammalian somatic cells were reprogrammed to iPSCs by enforced expression of OCT3/4, SOX2, KLF4, and c-MYC [4] or an alternative set of OCT3/4, SOX2, LIN28, and NANOG [5]. The iPSCs, which bypass the ethical issue of ESCs resulting from destroying early embryos, could generate patient-specific cell types of various lineages in vitro. However, some obstacles including long-term manipulation, low reprogramming and differentiation efficiency, and tumorigenicity have prevented iPSCs from a broad range of clinical application. Meanwhile, the somatic stem cells could also be propagated to a large number of differentiated cell types, without the risk of tumor formation, thus enabling them to be closer to the clinical application compared with PSCs.

For the somatic stem cells, bone marrow-derived stem cells (BMSCs) and adipose-derived stem cells (ADSCs) are investigated for a long period and have been applied in various experimental studies and preclinical trials [6–9]. Although both BMSCs and ADSCs have multiple potential to differentiate into various cell types, they are obtained through invasive procedures and could bring damage to the patients, especially to the pediatric patients and those with abnormal hemorrhagic diseases. Therefore, within the latest decade, urine-derived stem cells (UDSCs) are emerging as a promising cell resource for their noninvasive obtaining procedure, potent proliferation ability, multiple application in cell therapy, and tissue engineering [10–15] and serve as original cells for reprogramming into disease-specific iPSCs [16]. Based on these applications, UDSCs are currently playing an important role in adult stem cell biology.

1. Multiple Transdifferentiation of UDSCs Applied in Cell Therapy and Tissue Engineering

The autologous somatic stem cells have special advantages for the future clinical application, since they usually do not induce immune rejection. In addition, the ability to expand
to a large amount and to be induced into various cell lineages is also the basis for the somatic stem cells to explore their application in cell-based therapies and regenerative medicine. UDSCs could be expanded to yield a large population, and their plasticity has also been fully confirmed through investigation [17].

First, urine-derived cells were cultured from newborn children and displayed limited proliferation potential [18]. Urine cells with high proliferation ability were successfully cultured in a study of urological tissue reconstruction in 2008 [10]. In total, 55 urine samples from 15 volunteers and 8 patients were collected and cultured. Three types of cells with different morphologies were propagated representing fully differentiated, differentiating, and progenitor-like cells, respectively. Only the small cell type with a spindle appearance was named as UDSCs, as they could be consecutively proliferated for up to 20 passages, reaching accumulated population doubling (PD) rate of more than 60 [10, 15]. These cells most likely came from the parietal cell-podocyte interface of the renal glomerulus and expressed the corresponding markers [11, 12, 15]. Once isolated, the UDSCs could be consecutively expanded in culture in vitro and give rise to a variety of cell types via induction of lineage-specific differentiation under appropriate experimental conditions. Since its identification, up to now, UDSCs have been induced into ectodermal, mesodermal, and endodermal lineages. Ectodermal neural lineage was obtained through culturing UDSCs in neural induction medium supplemented with basic fibroblast growth factor [15, 19, 20]. Approximately 40% of the induced cells expressed several neural markers such as nestin, S100, NEF200, and GFAP, as well as exhibiting neurogenic extensions and processes, both in vitro and in vivo [15, 19]. Human urine cells from volunteers and Wilson’s disease patient could also be induced into neural lineage through the overexpression of Ascl1, Brn2, NeuroD, c-Myc, and Myt1l, characterized by expressing multiple neuronal markers and generating action potentials [20]. The neural lineage differentiation of UDSCs needs to be further investigated in future research. Endodermal lineage was obtained through culturing UDSCs in endothelial basal medium supplemented with vascular endothelial growth factor (VEGF). The induced cells developed a cobblestone-like morphology and expressed urothelial-specific markers such as uroplakin-III, uroplakin-Ia, CK7, and AE1/AE3 [15]. UDSCs have also been induced into multiple mesodermal lineage including osteogenic cells [21–23] and muscle cells [24–26]. After seeding on composite PLGA/CS scaffolds which were incorporated with calcium silicate, UDSCs demonstrated therapeutic potential in bone tissue regeneration in vivo through activation of the Wnt/β-catenin signaling pathway [22]. The UDSCs overexpressing VEGF could enhance the survival of grafted cells and promote myogenic differentiation, as well as improving the innervations, which could help to develop cell therapeutic strategy to correct stress urinary incontinence [26].

Tissue engineering is a promising field offering the possibility of providing scaffolds in order to structurally and functionally restore the altered pathological tissues. Although the complex structure and functions of the bladder have made this process challenging, great advancements have been achieved in the last few years using a scaffold and various stem cells to construct bladder and other urological tissues. Obtaining large amount of patients’ cells is the primary issue in constructing autologous tissues. The ideal cells should be propagated through noninvasive manipulation and possess high proliferation ability. As a noninvasive and easy-to-expand cell resource, UDSCs have been applied in urological tissue engineering including bladder tissue engineering and urethral reconstruction [10, 15, 27–34]. The recent and previous publications have demonstrated that the autologous UDSCs could be differentiated into urothelial cells and smooth muscle cells. Besides, it could also be applied in urethral reconstruction with the advantages of less inflammation and fibrosis compared with the control group in urethral defect models [10, 27–29]. UDSCs have also been applied in bladder tissue engineering after being transdifferentiated into bladder-associated cell types such as smooth muscle cells and urothelial cells. Even more, they demonstrated markers such as tight junction including ZO-1, E-cadherin, and cingulin, which indicated that a protective ultrastructure barrier has formed and could probably protect the engineered bladder tissues from urine [15, 32]. Although there are still tremendous issues such as vascularization and neurotization which need to be settled before clinical application, these efforts have provided great potential for the use of UDSCs. In addition, more applications of UDSCs on nonurological tissue engineering need to be explored and investigated in the future.

3. UDSCs Served as Original Cells for Reprogramming into Disease-Specific iPSCs

Although animal models are commonly applied to investigate disease mechanisms, these in vivo research models have a few limitations which could be potentially overcome through ex vivo human cellular models such as iPSCs. Modeling various human diseases “in a culture dish” is a fundamental application of human disease-specific iPSCs for its genetic background of the targeted disease [16, 35–37].

Two steps including derivation of iPSCs from a patient’s somatic cells and subsequent differentiation into disease-related cell types are important in modeling human diseases. Typically, parental somatic cells such as fibroblast and blood cells are harvested invasively from patients through biopsy or blood extraction. For some special patients such as children or those with abnormal hemorrhagic diseases, UDSCs have special advantages as they could be obtained noninvasively and cultured easily. Thus, UDSCs have been selected as alternative starting cells to generate iPSCs for both genders and all ages [38–40]. UDSC-derived disease-specific iPSCs have already been established in cardiac diseases [16], endocrine diseases [41, 42], abnormal hemorrhagic diseases resulting from various causes [43–45], aneuploidy diseases such as Down syndrome [46], neural diseases [47, 48], muscular disorders [49, 50], fibrodyplasia ossificans progressiva [51, 52], systemic lupus erythematosus [53], cryptorchidism [54], hypercholesterolemia [55], paroxysmal kinesigenic dyskinesia [56],

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and so on (Table 1). After successful reprogramming and characterization, differentiation experiments are essential, since most of disease phenotypes are usually observed in lineage-committed cells after in vitro differentiation rather than being observed in the iPSCs. In vitro, stepwise-directed differentiation is usually conducted according to the in vivo developmental pathway of the targeted cell type and often spans multiple weeks. Marker expression is detected during the consecutive developmental stage of differentiation both at mRNA level and at protein level. Even more, functional assays such as electrophysiology are also needed to study the pathophysiology of the targeted cells.

Based on these abovementioned research strategies, UDSC-derived iPSCs and the subsequent functional experiments have been applied in several disease modeling techniques. For the hemorrhagic disease category, iPSCs were generated successfully from 7 hemophilia A patients. The differentiated hepatocytes from these iPSCs failed to produce FVIII, which recapitulated the FVIII deficiency of hemophilia A. Thus, this cell model provided an effective way for modeling hemophilia A in vitro for further gene and cell therapy studies [45].

For the neurological disease category, urine samples were collected from 10 individuals with Down syndrome comprising 5 females and 5 males. The iPSCs were established and named as T21-iPSCs which were more sensitive to proteotox stress than euploid iPSCs. This study also indicated that T21-iPSCs could be differentiated into glutamatergic neurons which could fire action potential similar to euploid iPSCs. T21-iPSCs could also be induced into cardiomyocytes which exhibited spontaneous contractions and were sensitive to the beta adrenergic agonist isoproterenol [46]. Since both neurological disorders and congenital heart defects were the two most common complications of Down syndrome, these researches could probably be applied in human cell-based high-throughput drug screening in translational preclinical studies aimed at improving the life quality of patients with Down syndrome. Meanwhile, UDSCs have also been applied in the research of rare diseases. The long QT syndrome is a genetically inherited cardiac disease that can cause potentially fatal cardiac arrhythmia. Research showed that hiPSC derived from theHERG A561P-mutated urine cells (A561P-UhiPS CMs) can be differentiated into functional cardiomyocytes. Compared with the control healthy UhiPS-CMs, the A561P mutation caused a trafficking defect which led to delayed rectifier K⁺ current [16]. Fibrodysplasia ossificans progressiva (FOP) is an extremely rare connective tissue disease without effective treatment currently. It is characterized by progressive heterotopic ossification of soft tissues, and the molecular mechanisms underlying the pathology of FOP need to be investigated, as well as the identification of new therapeutic drugs through a proper research model [51]. The FOP-iPSC lines containing ALK2 mutation displayed decreased differentiation efficiency into bone-forming progenitors and reduced expression of VEGF receptor 2 in differentiated endothelial cells. The ALK2 kinase inhibitor could also partly inhibit the increase in mineralization of FOP-hiPSC-derived pericytes [52]. All these achievements had enabled the FOP-iPSCs as an alternative research model to evaluate the bioactivity of ALK2 inhibitors and other therapeutic drug candidates. Cryptorchidism is a common congenital birth defects, and infertility is an important complication with no proper treatment in adulthood [57]. Cryptorchidism-specific iPSCs have been established and differentiated into VASA-positive germ cell lineage which produced a potential model for investigating the mechanisms and treatments to infertility [54]. The autosomal dominant hypercholesterolemia is caused by mutated proprotein convertase subtilisin/kexin type 9 (PCSK9), which is a critical modulator of cholesterol homeostasis. PCSK9-iPSC lines were successfully established from urine cells and could be differentiated into hepatocyte-like cells. This study also indicated that the induced hepatocyte-like cells displayed altered PCSK9 secretion and LDL uptake, which mimic the pathophysiology of hypercholesterolemia and could be applied in drug screening [55]. Except for these abovementioned disease modeling techniques with functional experiments, further functional experiments need to be conducted on other UDSC-derived iPSCs after being induced into target cell types [41–44, 48–50, 53].

4. Current Challenges and Future Perspectives

UDSCs have been applied as a novel noninvasive cell source possessing a broad feasibility in cell therapies and tissue regeneration especially for urinary tissue engineering and also serving as original cells for disease modeling through reprogramming. However, since the biological characteristics of UDSCs have not been fully investigated yet, further basic research and practical animal studies are needed before they could be applied to the clinical therapeutics.

There are still a few issues yet to be settled. The first issue lies on the cell diversity which was displayed as line-to-line variations on both UDSCs and reprogrammed iPSCs resulting mainly from genetic background. When the experimental cells were compared with the control cells derived from another individual with different genetic background, these diversities could probably complicate the data interpretation and bring other problems such as experimental reproducibility. This issue could probably be settled through setting the experimental disease-causing mutation group and the control group originated from the same cell resource, which means that the experimental group is created by specific gene mutation on the control group through targeted genome editing technology. In such circumstance, both groups have the same genetic background except for the targeted mutation which could help to elucidate the disease-causing mechanism of the mutation. The second issue lies on the establishment of a clinical-grade cell resource of both UDSCs and reprogrammed iPSCs. This issue could probably be settled through recent technological innovations such as using integration-free reprogramming technology and xeno-free culture conditions. Great efforts have been made in making a clinical-grade cell under the guidelines of Good Manufacturing Practice and would probably benefit the patients in the near future. The third issue lies on the epigenetic memories of reprogrammed iPSCs. Which type of the original cell is chosen to reprogram usually depends on the
| Disease | Genetic etiology/mutation sites | Reprogramming factors | Reprogramming strategy | Major findings | Refs |
|---------|--------------------------------|-----------------------|------------------------|----------------|------|
| Type 2 long QT syndrome | KCNH2/A561P.c7:150 648 800G>C | OSKMLN + SV40LT | Episomal vectors | A561P-UhiPSC lines were established. A561P-UhiPSCs could be differentiated into functional cardiomyocyte cells. A561P KCNH2 mutation caused a trafficking defect of the HERG channel. HERG A561P mutation increased the susceptibility to arrhythmia. | [16] |
| Multiple endocrine neoplasia type 1 syndrome | Men1/exon 9 | OSK + miR-302-367 | Episomal plasmids | MEN1-iPSC lines of male and female were established. No functional experiments were achieved. | [41, 42] |
| Novel heterozygous PAI-1 mutation | PAI-1/exon 4 | OSKM | Sendai virus | No functional experiments were achieved. HA-iPSC lines were established. HA-iPSCs could be differentiated into hepatocyte-like cells in vitro. HA-iPSC-derived hepatocyte-like cells displayed FVIII deficiency. T21-iPSC lines were established. T21-iPSCs were more sensitive to proteotoxic stress than euploid iPSCs. T21-iPSCs could be differentiated into glutamatergic neurons and cardiomyocytes. Neurons from T21-iPSCs could fire AP similar to euploid iPSCs. SCI-iPSC lines were established. SCI-iPSCs could be differentiated into A2B5+ NPCs. A2B5+ NPCs could give rise to neurons and astrocytes after implantation. | [43, 44, 45] |
| Hemophilia A | FVIII/intron 22 inversion | OSK + SV40LT | Episomal vectors | | |
| Down syndrome | Trisomy 21 | OSKM | Episomal vectors | | |
| Spinal cord injury | No verification/no verification | OSKM | Sendai virus | A2B5+ NPCs could give rise to neurons and astrocytes after implantation. | [47] |
| Attention-deficit hyperactivity disorder | No verification/no verification | OSKM | Sendai virus | No functional experiments were achieved. | [48] |
| Dilated cardiomyopathy | No verification/no verification | OSKM | Sendai virus | DCM-iPSC lines were established. | [49] |
cell accessibility, reprogramming efficiency, and the expected progression pattern of the specific disease. Several researchers have demonstrated that iPSCs reprogrammed from different original somatic cells including UDSCs exhibited distinct transcriptional and epigenetic patterns, as well as various in vitro differentiation potentials [58, 59]. Research also indicated that the epigenetic memory of the original cells resulted from incomplete reprogramming and the biased in vitro differentiation could influence the applications in disease modeling and treatment [60]. This issue could probably be settled through improving reprogramming strategies and achieving a complete pluripotency. In addition, further research is needed to elucidate the mechanisms that regulate pluripotency and to improve directed differentiation efficiency to produce the mature target cell type. Upon all these abovementioned issues, polygenic disease-specific iPSCs to recapitulate more complex diseases are facing even greater challenges.

5. Conclusions

In conclusion, UDSCs are a novel noninvasively obtained cell source with high proliferation ability and multiple

| Disease                              | Genetic etiology/mutation sites | Reprogramming factors | Reprogramming strategy | Major findings                                                                 | Refs   |
|--------------------------------------|---------------------------------|-----------------------|------------------------|---------------------------------------------------------------------------------|--------|
| Muscular dystrophy                   | Dystrophin/exon deletion        | OSKM                  | Sendai virus           | No functional experiments were achieved. MD-iPSC lines were established. MD-iPSCs lack the expression of dystrophin. FOP-iPSC lines were established. Differentiation efficiency into bone-forming progenitors was decreased. Expression of VEGF receptor 2 in differentiated endothelial cells was reduced. Mineralization of pericytes from FOP-hiPSCs was increased. | [50]   |
| Fibrodysplasia ossificans progressiva | ALK2/R206H                      | OSKM/OSK + mirR-302-367 | Sendai virus/episomal vectors | FOP-iPSC lines were established. Di ff erentiation efficiency into bone-forming progenitors was decreased. Expression of VEGF receptor 2 in differentiated endothelial cells was reduced. Mineralization of pericytes from FOP-hiPSCs was increased. | [51, 52] |
| Systemic lupus erythematosus         | No verification/no verification | OSKM                  | Lentivirus             | SLE-iPSC lines were established. No functional experiments were achieved. Cryp-iPSC lines were established. Cryp-iPSCs could be differentiated into VASA+ germ cell. PCSK9-iPSC lines were established. PCSK9-iPSCs could be differentiated into hepatocyte-like cells. PCSK9 secretion and LDL uptake were altered. PKD-iPSC lines were established. PKD-iPSCs could be differentiated into functional glutamatergic, dopaminergic, and motor neurons. The expression of PRRT2 was decreased in PKD-iPSCs. | [53]   |
| Cryptorchid                          | INSL3/c.A178>G, ZNF214/c.A197>G, c.T383>A and c.T754>G, ZNF215/c.T108>A, c.A400>G, c.A780>T, and c.C788>T | OSKM                  | Lentivirus             | No functional experiments were achieved. Cryp-iPSC lines were established. Cryp-iPSCs could be differentiated into VASA+ germ cell. PCSK9-iPSC lines were established. PCSK9-iPSCs could be differentiated into hepatocyte-like cells. PCSK9 secretion and LDL uptake were altered. PKD-iPSC lines were established. PKD-iPSCs could be differentiated into functional glutamatergic, dopaminergic, and motor neurons. The expression of PRRT2 was decreased in PKD-iPSCs. | [54]   |
| Hypercholesterolemia                 | PCSK9/S127R and R104C/V114A     | OSKMLN + SV40LT       | Episomal vectors       | No functional experiments were achieved. Cryp-iPSC lines were established. Cryp-iPSCs could be differentiated into VASA+ germ cell. PCSK9-iPSC lines were established. PCSK9-iPSCs could be differentiated into hepatocyte-like cells. PCSK9 secretion and LDL uptake were altered. PKD-iPSC lines were established. PKD-iPSCs could be differentiated into functional glutamatergic, dopaminergic, and motor neurons. The expression of PRRT2 was decreased in PKD-iPSCs. | [55]   |
| Paroxysmal kinesigenic dyskinesia    | PRRT2/c.649dupC                 | OSKM                  | Retroviruses           | FOP-iPSC lines were established. Di ff erentiation efficiency into bone-forming progenitors was decreased. Expression of VEGF receptor 2 in differentiated endothelial cells was reduced. Mineralization of pericytes from FOP-hiPSCs was increased. | [56]   |

The abbreviations represent a combination of reprogramming factors: O: OCT3/4; S: SOX2; K: KLF4; M: c-MYC; L: LIN28; N: NANOG.
differentiation potential and were being reprogrammed to model diseases. However, the broad and powerful application of UDSCs is yet to achieve through further investigation, both on regenerative medicine and on disease modeling after being reprogrammed.

Conflicts of Interest

The authors declare that they have no conflict of interests.

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References

[1] S. Morrison, “Advancing stem cell science and translation,” Stem Cell Reports, vol. 6, no. 6, pp. 785–786, 2016.
[2] A. Atala, “Advancing the translation of stem cells to medicine,” Stem Cells Translational Medicine, vol. 6, no. 1, pp. 1-2, 2017.
[3] J. A. Thomson, J. Itskovitz-Eldor, S. S. Shapiro et al., “Embryonic stem cell lines derived from human blastocysts,” Science, vol. 282, no. 5391, pp. 1145–1147, 1998.
[4] K. Takahashi, K. Tanabe, M. Ohnuki et al., “Induction of pluripotent stem cells from adult human fibroblasts by defined factors,” Cell, vol. 131, no. 5, pp. 861–872, 2007.
[5] J. Yu, M. A. Vodyanik, K. Smuga-Otto et al., “Induced pluripotent stem cell lines derived from human somatic cells,” Science, vol. 318, no. 5858, pp. 1917–1920, 2007.
[6] S. Liu, J. Zhou, X. Zhang et al., “Strategies to optimize adult stem cell therapy for tissue regeneration,” International Journal of Molecular Sciences, vol. 6, p. 17, 2016.
[7] G. I. Im, “Bone marrow-derived stem/stromal cells and adipose tissue-derived stem/stromal cells: their comparative efficacies and synergistic effects,” Journal of Biomedical Materials Research Part A, vol. 105, no. 9, pp. 2640–2648, 2017.
[8] A. El-Badawy, S. M. Ahmed, and N. El-Badri, “Adipose-derived stem cell-based therapies in regenerative medicine,” Advances in Stem Cell Therapy, pp. 117–138, 2017.
[9] J. H. Arrizabalaga and M. U. Nollert, “Properties of porcine adipose-derived stem cells and their applications in preclinical models,” Adipocytes, vol. 6, no. 3, pp. 217–223, 2017.
[10] Y. Zhang, E. McNeill, H. Tian et al., “Urine derived cells are a potential source for urological tissue reconstruction,” Journal of Urology, vol. 180, no. 5, pp. 2262–2263, 2016.
[11] D. Zhang, G. Wei, P. Li, X. Zhou, and Y. Zhang, “Urine-derived stem cells: a novel and versatile progenitor source for cell-based therapy and regenerative medicine,” Genes & Diseases, vol. 1, no. 1, pp. 8–17, 2014.
[12] G. Liu, C. Deng, and Y. Zhang, “Urine-derived stem cells: biological characterization and potential clinical applications,” Stem Cells: Current Challenges and New Directions, pp. 19–28, 2013.
[13] B. Bussolati and G. Camussi, “Therapeutic use of human renal progenitor cells for kidney regeneration,” Nature Reviews Nephrology, vol. 11, no. 12, pp. 695–706, 2015.
[14] F. Oliveira Arcolino, A. Tort Piella, E. Papadimitriou et al., “Human urine as a noninvasive source of kidney cells,” Stem Cells International, vol. 2015, Article ID 362562, 7 pages, 2015.
[15] S. Bharadwaj, G. Liu, Y. Shi et al., “Multipotential differentiation of human urine-derived stem cells: potential for therapeutic applications in urology,” Stem Cells, vol. 31, no. 9, pp. 1840–1856, 2013.
[16] M. Jouni, K. Si-Tayeb, Z. Es-Salah-Lamoureux et al., “Toward personalized medicine: using cardiomycocytes differentiated from urine-derived pluripotent stem cells to recapitulate electrophysiological characteristics of type 2 long QT syndrome,” Journal of the American Heart Association, vol. 4, no. 9, article e002159, 2015.
[17] L. Shi, Y. Cui, J. Luan, X. Zhou, and J. Han, “Urine-derived induced pluripotent stem cells as a modeling tool to study rare human diseases,” Intractable & Rare Diseases Research, vol. 5, no. 3, pp. 192–201, 2016.
[18] G. R. Sutherland and A. D. Bain, “Culture of cells from the urine of newborn children,” Nature, vol. 239, no. 536, p. 231, 1972.
[19] J. J. Guan, X. Niu, F. X. Gong et al., “Biological characteristics of human-urine-derived stem cells: potential for cell-based therapy in neurology,” Tissue Engineering Part A, vol. 20, no. 13-14, pp. 1794–1806, 2014.
[20] S. Z. Zhang, L. X. Ma, W. J. Qian, H. F. Li, and Z. F. Wang, “Modeling neurological disease by rapid conversion of human urine cells into functional neurons,” Stem Cells International, vol. 2016, Article ID 2452985, 8 pages, 2016.
[21] J. Guan, J. Zhang, Z. Zhu et al., “Bone morphogenetic protein 2 gene transduction enhances the osteogenic potential of human urine-derived stem cells,” Stem Cell Research & Therapy, vol. 6, p. 5, 2015.
[22] J. Guan, J. Zhang, S. Guo et al., “Human urine-derived stem cells can be induced into osteogenic lineage by silicate bioceramics via activation of the Wnt/β-catenin signaling pathway,” Biomaterials, vol. 55, pp. 1–11, 2015.
[23] J. Guan, J. Zhang, H. Li et al., “Human urine derived stem cells in combination with β-TCP can be applied for bone regeneration,” PLoS One, vol. 10, no. 5, article e0125253, 2015.
[24] E. Y. Kim, P. Page, L. M. Dellefave-Castillo, E. M. McNally, and E. J. Wyatt, “Direct reprogramming of urine-derived cells with inducible Myod for modeling human muscle disease,” Skeletal Muscle, vol. 6, p. 32, 2016.
[25] W. Chen, M. Xie, B. Yang et al., “Skeletal myogenic differentiation of human urine-derived cells as a potential source for skeletal muscle regeneration,” Journal of Tissue Engineering and Regenerative Medicine, vol. 11, no. 2, pp. 334–341, 2017.
[26] G. Liu, X. Wang, X. Sun, C. Deng, A. Atala, and Y. Zhang, “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, vol. 34, no. 34, pp. 8617–8629, 2013.
[27] S. Ramsay, C. Ringouette-Goulet, A. Langlois, and S. Bolduc, “Clinical challenges in tissue-engineered urethral reconstruction,” Translational Andrology and Urology, vol. 5, no. 2, pp. 267–270, 2016.
[28] Y. Liu, W. Ma, B. Liu et al., “Urethral reconstruction with autologous urine-derived stem cells seeded in three-
dimensional porous small intestinal submucosa in a rabbit model,” Stem Cell Research & Therapy, vol. 8, no. 1, p. 63, 2017.

[29] L. R. Versteegden, P. K. de Jonge, J. IntHout et al., “Tissue engineering of the urethra: a systematic review and meta-analysis of preclinical and clinical studies,” European Urology, vol. 72, no. 4, pp. 594–606, 2017.

[30] P. Gao, D. Jiang, W. Liu, H. Li, and Z. Li, “Urine-derived stem cells, a new source of seed cells for tissue engineering,” Current Stem Cell Research & Therapy, vol. 11, no. 7, pp. 547–553, 2016.

[31] B. Liu, F. Ding, Y. Liu, G. Xiong, T. Lin, and D. He, “Urine-derived stem cells: a novel source for tissue engineering and regenerative medicine,” Journal of Biomaterials and Tissue Engineering, vol. 6, no. 8, pp. 589–601, 2016.

[32] Y. Y. Chan, S. K. Sandlin, E. A. Kurzrock, and S. L. Osborn, “The current use of stem cells in bladder tissue regeneration and bioengineering,” Biomedicine, vol. 5, no. 1, 2017.

[33] A. Bodin, S. Bharadwaj, S. Wu, P. Gatenholm, A. Atala, and Y. Zhang, “Tissue-engineered conduit using urine-derived stem cells seeded bacterial cellulose polymer in urinary reconstruction and diversion,” Biomaterials, vol. 31, no. 34, pp. 8889–8901, 2010.

[34] Z. Tong, C. Cao, M. Rao, and J. Lu, “Potential cell source for cell- based therapy and tissue engineering applications: urine-derived stem cells,” Journal of Biomaterials and Tissue Engineering, vol. 5, no. 2, pp. 150–156, 2015.

[35] I. H. Park, N. Arora, H. Huo et al., “Disease-specific induced pluripotent stem cells,” Cell, vol. 134, no. 5, pp. 877–886, 2008.

[36] D. L. Mack, X. Guan, A. Wagoner, S. J. Walker, and M. K. Childers. “Disease-in-a-dish: the contribution of patient-specific induced pluripotent stem cell technology to regenerative rehabilitation,” American Journal of Physical Medicine & Rehabilitation, vol. 93, no. 11, Supplement 3, pp. S155–S168, 2014.

[37] Y. Avior, I. Sagi, and N. Benvenisty, “Pluripotent stem cells in disease modelling and drug discovery,” Nature Reviews Molecular Cell Biology, vol. 17, no. 3, pp. 170–182, 2016.

[38] T. Zhou, C. Benda, S. Dunzinger et al., “Generation of human induced pluripotent stem cells from urine samples,” Nature Protocols, vol. 7, no. 12, pp. 2080–2089, 2012.

[39] T. Zhou, C. Benda, S. Dunzinger et al., “Generation of induced pluripotent stem cells from urine,” Journal of American Society of Nephrology, vol. 22, no. 7, pp. 1221–1228, 2011.

[40] E. Mizutani, K. Torikai, S. Wakayama et al., “Generation of cloned mice and nuclear transfer embryonic stem cell lines from urine-derived cells,” Scientific Reports, vol. 6, article 23808, 2016.

[41] D. Guo, F. Wu, H. Liu et al., “Generation of non-integrated induced pluripotent stem cells from a 23-year-old male with multiple endocrine neoplasia type 1 syndrome,” Stem Cell Research, vol. 18, pp. 70–72, 2017.

[42] D. Guo, F. Wu, H. Liu et al., “Generation of non-integrated induced pluripotent stem cells from a 59-year-old female with multiple endocrine neoplasia type 1 syndrome,” Stem Cell Research, vol. 18, pp. 64–66, 2017.

[43] M. Z. Afzal, M. Gartz, E. A. Klyachko et al., “Generation of human iPSCs from urine derived cells of patient with a novel heterozygous PAI-1 mutation,” Stem Cell Research, vol. 18, pp. 41–44, 2017.

[44] M. Z. Afzal, M. Gartz, E. A. Klyachko et al., “Generation of human iPSCs from urine derived cells of a non-affected control subject,” Stem Cell Research, vol. 18, pp. 33–36, 2017.

[45] B. Jia, S. Chen, Z. Zhao et al., “Modeling of hemophilia A using patient-specific induced pluripotent stem cells derived from urine cells,” Life Sciences, vol. 108, no. 1, pp. 22–29, 2014.

[46] Y. M. Lee, B. L. Zampieri, J. J. Scott-McKean, M. W. Johnson, and A. C. S. Costa, “Generation of integration-free induced pluripotent stem cells from urine-derived cells isolated from individuals with Down syndrome,” Stem Cells Translational Medicine, vol. 6, no. 6, pp. 1465–1476, 2017.

[47] Y. Liu, Y. Zheng, S. Li et al., “Human neural progenitors derived from integration-free iPSCs for SCI therapy,” Stem Cell Research, vol. 19, pp. 55–64, 2017.

[48] J. Sochacki, S. Devalle, M. Reis, P. Mattos, and S. Rehen, “Generation of urine iPSC cell lines from patients with attention deficit hyperactivity disorder (ADHD) using a non-integrative method,” Stem Cell Research, vol. 17, no. 1, pp. 102–106, 2016.

[49] Y. H. Lin, X. M. Chen, J. W. Zhang, X. Q. He, W. J. Dai, and M. S. Chen, “Preclinical study on induction of pluripotent stem cells from urine of dilated cardiomyopathy patients,” European Review for Medical and Pharmacological Sciences, vol. 20, no. 8, pp. 1450–1457, 2016.

[50] M. Z. Afzal and J. L. Strande, “Generation of induced pluripotent stem cells from muscular dystrophy patients: efficient integration-free reprogramming of urine derived cells,” Journal of Visualized Experiments, no. 95, article 52032, 2015.

[51] J. Cai, V. V. Orlova, X. Cai et al., “Induced pluripotent stem cells to model human fibro dysplasia ossificans progressiva,” Stem Cell Reports, vol. 5, no. 6, pp. 963–970, 2015.

[52] L. Hildebrand, B. Rossbach, P. Kuhnen et al., “Generation of integration free induced pluripotent stem cells from fibro dysplasia ossificans progressiva (FOP) patients from urine samples,” Stem Cell Research, vol. 16, no. 1, pp. 54–58, 2016.

[53] Y. Chen, R. Luo, Y. Xu et al., “Generation of systemic lupus erythematosus-specific induced pluripotent stem cells from urine,” Rheumatology International, vol. 33, no. 8, pp. 2127–2134, 2013.

[54] J. Zhou, X. Wang, S. Zhang et al., “Generation and characterization of human cryptorchid-specific induced pluripotent stem cells from urine,” Stem Cells Development, vol. 22, no. 5, pp. 717–725, 2013.

[55] K. Si-Tayeb, S. Idrissi, B. Champon et al., “Urine-sample derived human induced pluripotent stem cells as a model to study PCSK9-mediated autosomal dominant hypercholesterolemia,” Disease Models & Mechanisms, vol. 9, no. 1, pp. 81–90, 2016.

[56] S. Z. Zhang, H. F. Li, L. X. Ma, W. J. Qian, Z. F. Wang, and Z. Y. Wu, “Urine-derived induced pluripotent stem cells as a modeling tool for paroxysmal kinesigenic dyskinesia,” Biology Open, vol. 4, no. 12, pp. 1744–1752, 2015.

[57] P. A. Lee and C. P. Houk, “Cryptorchidism,” Current Opinion in Endocrinology, Diabetes & Obesity, vol. 20, no. 3, pp. 210–216, 2013.

[58] J. M. Polo, S. Liu, M. E. Figueroa et al., “Cell type of origin influences the molecular and functional properties of mouse induced pluripotent stem cells,” Nature Biotechnology, vol. 28, no. 8, pp. 848–855, 2010.
[59] F. Rouhani, N. Kumasaka, M. C. de Brito, A. Bradley, L. Vallier, and D. Gaffney, “Genetic background drives transcriptional variation in human induced pluripotent stem cells,” *PLoS Genetics*, vol. 10, no. 6, article e1004432, 2014.

[60] K. Kim, A. Doi, B. Wen et al., "Epigenetic memory in induced pluripotent stem cells," *Nature*, vol. 467, no. 7313, pp. 285–290, 2010.