Placenta xxx (2011) 1–5

Contents lists available at ScienceDirect

Placenta

journal homepage: www.elsevier.com/locate/placenta

Wharton's Jelly stem cells: Future clinical applications

R.R. Taghizadeh*, K.J. Cetrulo, C.L. Cetrulo

AuxoCell Laboratories, Inc., 245 First Street, 18th Floor, Cambridge, MA 02142, USA

ARTICLE INFO

Article history:
Accepted 16 June 2011

Keywords:
Mesenchymal stem cells
Umbilical cord blood
Umbilical cord tissue
Wharton's Jelly stem cells
Stem cells
Mesenchymal stromal cells

ABSTRACT

This review focuses on the therapeutic potential of stem cells harvested from the Wharton's Jelly of the human umbilical cord. Recently, investigators have found that a potent stem cell population exists within the Wharton's Jelly. In this review, the authors define a new subset of stem cells, termed perinatal stem cells, and compare them to other sources of stem cells. Furthermore, cryopreservation of Wharton's Jelly stem cells is described for potential use in future cell based therapies and/or regenerative medicine applications. Current evidence of the application of mesenchymal stem cells from various sources in both pre-clinical and clinical trials is reviewed in the context of potential indications of use for Wharton's Jelly derived mesenchymal stem cells.

1. Introduction

The term perinatal encompasses the time from the 20th week of gestation to the neonatal period (the first 28 days of life). The tissue that sustains natal development is typically discarded as medical waste post-delivery. As such, harvesting stem cells from these tissues represent a safe, non-invasive means for attaining therapeutically beneficial stem cells. These include amnion/amniotic fluid, umbilical cord blood, placental tissue, umbilical cord vein, and the Wharton's Jelly contained within the umbilical cord sometimes referred to as umbilical cord tissue [1–9].

Perinatal stem cells are not embryonic stem cells (ESCs), nor are they somatic (adult) stem cells (ASCs); they represent a bridge between embryonic and adult stem cells. Human embryonic stem cells are derived from the inner cell mass of the developing blastocyst in the initial post-fertilization cell divisions [10,11]. They possess the ability to produce all the cells from all germ layers, including adult tissue-specific (somatic) stem cells and differentiated cells. Adult stem cells, on the other hand, are multipotent tissue-specific stem cells that maintain cell turnover units within the tissue [8,12]. Furthermore, adult stem cells may possess the ability to trans-differentiate to other cell types from other tissues. Perinatal stem cells possess characteristics of both embryonic stem cells and adult stem cells as they possess pluripotency properties, as well as multipotent tissue maintenance [13].

Despite these inherent characteristics, both embryonic and adult stem cells have significant drawbacks. For one, human embryonic stem cells will form teratomas when transplanted [14]. As a result, embryonic stem cells cannot directly be transplanted into human patients and, therefore, must be manipulated *in vitro* and differentiated along tissue-specific lineages to form, for instance, adult stem cells or differentiated progeny, prior to transplantation. Furthermore, the derivation of human embryonic stem cells inherently destroys the development and potential of a human life. Not surprisingly, as a result, the use of human embryonic stem cells, in both research and clinical settings, possess a tremendous ethical cloud [15]. Adult stem cells, on the other hand, do not encompass similar ethical challenges (when proper IRB approval and/or patient consent are obtained). However, adult stem cells have relative limited proliferative potential (i.e. multipotent), are extremely rare *in vivo* and, generally speaking, are difficult to expand *ex vivo* [8]. Furthermore, the procurement of adult stem cells from patients is invasive and represents significant risk and discomfort to the patient [8].

These described limitations can be overcome with the utilization of various sources of an additional stem cell paradigm, termed here as perinatal stem cells. For one, perinatal stem cells, unlike embryonic stem cells, do not form tumors when transplanted. Additionally, perinatal stem cells may possess greater pluripotency capability than adult stem cells, as there is evidence that these cells can produce cells from all three germ layers [13,16–19]. This property will ultimately allow for greater tissue differentiation capacity. Since stem cells from perinatal tissue are procured from tissue that would otherwise be discarded as medical waste, there is little risk to the mother or newborn. Of the perinatal stem cell sources, Wharton's Jelly has great potential to emerge as a useful stem cell source to treat various diseases in the clinics. The

* Corresponding author. Tel./fax: +1 617 359 2612.
E-mail address: rouzbeh.taghizadeh@auxocell.com (R.R. Taghizadeh).

0143-4004/$ – see front matter © 2011 Elsevier Ltd. All rights reserved.
doi:10.1016/j.placenta.2011.06.010

potential clinical applications and indications for Wharton's Jelly stem cells are numerous [1,16–18,20]. This review will focus on the stem cells derived from the Wharton's Jelly of the human umbilical cord and their potential use in therapeutic applications.

2. What is Wharton's Jelly?

Wharton's Jelly is the primitive mucous, connective tissue of the umbilical cord lying between the amniotic epithelium and the umbilical vessels. First observed by Thomas Wharton in 1656, this gelatinous substance is comprised of proteoglycans and various isoforms of collagen. The main role of the Wharton's Jelly is to prevent the compression, torsion, and bending of the umbilical vessels which provide the bi-directions flow of oxygen, glucose and amino acids to the developing fetus, while also depleting the fetus and placenta of carbon dioxide and other waste products [16,18,21]. Cells found in Wharton's Jelly are a primitive mesenchymal stem cell (MSC), likely trapped in the connective tissue matrix as they migrated to the AGM (aorta-gonad-mesonephros) region through the developing cord, during embryogenesis (prior to E10.5) [21].

During early embryogenesis, hematopoiesis takes place in the yolk sac and later in the AGM region. Colonies of early hematopoietic cells and mesenchymal cells migrate through the early umbilical cord to the placenta between embryonic day 4 and 12 of embryogenesis [21]. There is a second migration from the placenta again through the early umbilical cord to the fetal liver and then finally to the fetal bone marrow where hematopoietic stem cells (HSCs) and mesenchymal stem cells engraft and predominantly reside for the duration of life. Included in these migrating hematopoietic colonies are early precursors of HSCs, as well as primitive mesenchymal stromal (stem) cells. Researchers have postulated that during this migration to and from the placenta through the umbilical cord, mesenchymal stromal cells become embedded in the Wharton's Jelly early in embryogenesis and remain there for the duration of gestation [21].

The formation of these perinatal stem cells at such an early embryonic state allows them to retain a resemblance to embryonic stem cells (ESCs), while still maintaining the properties of somatic mesenchymal stem cells found in bone marrow, as defined by the International Society for Cellular Therapy (ISCT) [22]. For the purposes of this article, Wharton's Jelly Stem Cells (WJSCs) are defined as native stem cell populations residing within the *in situ* umbilical cord extracellular matrices and umbilical cord mesenchymal stem cells (UC-MSCs) are defined as *in vitro* cell populations derived from Wharton's Jelly Stem Cells (WJSCs).

The problem of how to define UC-MSCs is further exacerbated because 'within the scientific literature, the acronym MSC has been used to represent (bone) marrow stromal cells, mesenchymal stem cells, and multipotent mesenchymal stromal cells' [22]. Further complicating the matter, researchers have used a variety of methods for isolation, culture, and characterization. The use of various methods leads to considerable ambiguity when study comparisons are attempted. In 2006, in order to pursue standardization, the Mesenchymal and Tissue Stem Cell Committee of the ISCT proposed minimal criteria for defining MSCs. To begin with, they proposed that these cells be designated as multipotent mesenchymal stromal cells (MSC). They further proposed three criteria for defining these cells: adherence to plastic culture ware, specific surface antigen expression, and multipotent differentiation potential. In the case of adherence to plastic, the adherence must be maintained under standard cell culture conditions using tissue culture flasks. Flow cytometric analysis of MSCs should demonstrate surface antigen expression of CD105, CD73, and CD90. Furthermore, these cells should show minimal expression of CD45, CD34, CD14 or CD11b, CD79a or CD19, and HLA class II proteins. Finally, these cells must demonstrate the ability to differentiate along osteogenic, adipogenic and chondrogenic lineages under standard *in vitro* differentiating conditions [22]. UC-MSC possess plastic adherence, can be differentiated along chondrogenic, adipogenic, and osteogenic lineages and possess expression of the following cell markers: CD105+ (endoglin, SH2), CD73+ (SH3), CD90+ (Thy-1), HLA-A,B,C+ (MHC class I), CD34, CD45-, HLA-DR- (MHC class II) [16–18].

Interestingly, UC-MSCs not only possess MSC properties but they exhibit properties to those attributed to ESCs. Specifically, UC-MSCs express human ESC markers Tra-1-60, Tra-1-81, SSEA-1 (stage-specific embryonic antigen-1), SSEA-4, alkaline phosphatase and even form embryoid bodies *in vitro* [23]. Additionally, UC-MSCs express the pluripotency markers Oct-4, Sox-2, and Nanog [13,16,24,25], albeit at relatively lower levels than ESCs [19,23]. Both qRT-PCR and microarray data have confirmed the relative lower expression of pluripotency markers in UC-MSCs, compared to human ESCs, irrespective of early- or late-passaged cells [19,23]. These markers are up-regulated in undifferentiated human embryonic stem cells and have been shown to maintain pluripotency in self-renewing human ESC populations [26]. Although lower relative expression of pluripotent markers would suggest that UC-MSCs are not as pluripotent as ESCs, it would suggest, however, that UC-MSCs are highly multipotent. In fact, microarray studies confirm that UC-MSCs express markers of all three primordial germ layers independent of passage length [19]. Furthermore, the regulation of these pluripotent genes has been shown to induce pluripotency in somatic cells (otherwise known as iPS; induced pluripotent stem cell) [26,27]. Not surprising, Oct-4, Sox-2 and Nanog are not expressed by adult stem cell sources, including bone marrow-derived MSCs [13], although forced expression of Oct-4 and Nanog in human BM-MSCs has been reported to improve their stemness [28].

Since WJSCs are trapped within the Wharton's Jelly between day 4 and 12 of embryonic development and reside there for the duration of gestation, they can be procured after birth of the newborn. The ability to procure these cells, in ethically unchallenged ways, is quite significant. Furthermore, these cells that formed during the earliest ontogenic time period result in significant differences in expansion potential compared to (adult) bone marrow mesenchymal stem cells (BM-MSCs). As previously reported, the number and potency of BM-MSCs specifically decreases with age, as indicated by lower *in vitro* CFU-F and proliferative potential, lower telomerase activity, longer population doubling times, and shorter times to senescence [29–32]. UC-MSCs maintain the same multipotent differentiation potential with relatively higher CFU-F and proliferative potential, higher telomerase activity, shorter population doubling times, and longer times to senescence, without loss of stem cell potency. Thus, UC-MSCs appear to be more primitive mesenchymal stem cells than those found in bone marrow and represent an earlier stage mesenchymal-like stem cell than those derived from adult fat or bone marrow [13,18,20].

In approximately 99% of all deliveries, these potentially therapeutic cells are discarded as medical waste. In the United States alone, every one of the approximately 4,000,000 annual births represents an opportunity to collect these cells. The same WJSCs which are trapped in early embryonic development during the migration to and from the placenta through the early umbilical cord can be easily collected and harvested from the Wharton's Jelly of the umbilical cord at the time of delivery. This ease of collection has obvious advantages over the collection of adult stem cells from fat and bone marrow, for which the donor has to undergo an invasive surgical procedure. This factor, coupled with the great expansion capabilities of UC-MSCs, enables this cell source to represent a virtually inexhaustible source of stem cells for both autologous and allogeneic cellular therapies and regenerative medicine products [24,33].

Please cite this article in press as: Taghizadeh RR, et al., Wharton's Jelly stem cells: Future clinical applications, Placenta (2011), doi:10.1016/j.placenta.2011.06.010

3. Umbilical Cord-derived Mesenchymal Stem Cells (UC-MSCs) as a universal source

In the field of cellular therapy, there are two models typically pursued. One is an autologous model which utilizes a patient's own cells for the therapy; the other is an allogeneic model which utilizes cells from another donor for the therapies. Most often these models are determined based on the capabilities of the cell source. In order for an allogeneic model to be considered, the cell source must be immunologically privileged, suggesting that the cells do not immunologically cross-react and, therefore, do not have to be human leukocyte antigens (HLA) matched for transplantation or they are HLA-matched to the recipient.

UC-MSCs, like bone marrow mesenchymal stem cells, are immunologically privileged. MSCs invoke only minimal immune reactivity, and, furthermore, may possess anti-inflammatory and immuno-modulatory effects [30,34–37]. UC-MSCs express MHC class I antigens and express low levels of MHC class II antigens, relatively less than BM-MSCs. As several studies currently suggest, UC-MSCs, like BM-MSCs, do not require tissue matching, thus, allowing for an allogeneic cell therapy source, as any donor can give cells to any other person without rejection or need of immuno-suppressant drugs [37]. This characteristic suggests that UC-MSCs can be used as a 'universal' or 'off-the-shelf' stem cell product.

WJSCs also work as a cell source for an autologous model. Presently, collection and cryogenic preservation of WJSCs along side matching umbilical cord blood (UCBs) units in private banking storage are being conducted. In this model, both the cord blood unit and the WJSC unit are processed using minimally manipulated procedures. Studies in mice have shown that a co-transplant of a single cord blood unit and a WJSC unit from either a related or an unrelated donor increases the engraftment efficiency of the infused cord blood HSCs, although the mechanism of action is currently unclear [20,38]. As such, preservation of matching WJSCs along side private umbilical cord blood units can significantly increase the chances of having a successful transplant if that cord blood unit is needed for transplantation. WJSCs are currently being banked along side public and private cord blood units. AuxoCell Laboratories, Inc. and the New Jersey Public Cord Blood Bank are working together to bank WJSCs units that match public cord blood units listed on the National Marrow Donor Program's (NMDP) registry, as well as private units. Currently, however, WJSC units are not listed on the NMDP's registry.

By currently building an inventory of WJSC units, this partnership anticipates a future where WJSCs will be used to potentially enhance every cord blood transplantation. Currently, investigative trials in animals are being conducted and will need to be translated into humans to further prove the safety and efficacy of these cells for this indication. Moreover, this publicly banked inventory will greatly increase the therapeutic potential of the existing worldwide public cord blood inventory as low potential therapeutic units (based on total nucleated cell count) can now be enhanced and administered as a clinically sufficient therapeutic dose for transplantation. As interest in banking of WJSCs continues to increase, various methods to process umbilical cord have been developed and implemented to bank WJSCs, in conjunction with umbilical cord blood stem cells. These advances have allowed the procurement, processing and cryopreservation of WJSCs to be conducted in existing infrastructure and, importantly, in a similar timeframe as current cord blood banking (unpublished data).

At the time of delivery, after the donor's umbilical cord blood has been collected, the umbilical cord, in its entirety, is clipped, and placed in the included collection jar. The collected cord blood and cord are placed in the kit and shipped to the processing center within 48 h, where the umbilical cord blood is processed using a FDA-certified automated method; the umbilical cord is processed using a separate technique, which involves a series of processing and separation steps. The final homogenous cell product is cryopreserved in a 25 mL cryopreservation bag, similar to the one used for the associated cord blood unit. WJSCs are then cryopreserved at a controlled rate, and then transferred to liquid nitrogen for long-term storage, once the units have passed all quality controls and are found to be free of pathogens and contaminants.

Samples are taken from each unit and characterized for expression of cell surface proteins using flow cytometry. WJSCs express CD105 (Endoglin Receptor), CD73 glycoprotein, CD90 (Thy-1), CD44 (homing-associated cell adhesion molecule; H-CAM), CD29 (Integrin β1), HLA-ABC, HLA-DR and lack expression for CD34 and CD45. Further characterizations may include CFU-F (colony forming unit–fibroblast), expansion potential and multipotential differentiation along osteogenic, chondrogenic, and adipogenic lineages [16,18]. Unit sterility (i.e. lack of bacterial contaminants) and the indicated characterization is sufficient for release of the unit for transplantation.

In the United States, AuxoCell Laboratories, Inc.'s proprietary processing technology is licensed by ViaCord for private banking. Additionally, AuxoCell has partnered with select private cord blood banks internationally, and continues to develop partnerships with both private and public cord blood banks worldwide.

4. Regenerative medicine applications of MSCs

Additional properties of MSCs make them useful stem cell candidates for use in various cell based therapies, beyond umbilical cord blood hematopoietic engraftment. For instance, UC-MSCs share the natural homing capabilities of BM-MSCs. For MSCs, an injury serves as a homing beacon, as they home to sites of inflammation and to locally effect the inflammatory/immune mediated tissue damage with subsequent ability to support tissue healing. They shift the spectrum of local cytokines from pro-inflammatory to anti-inflammatory [39,40]. Studies are currently ongoing to take full advantage of these unique properties for specific indications. The immunosuppressive ability of these cells has the potential to treat many disorders including graft-versus-host disease (GvHD) [41–44], diabetes [45,46], Crohn's disease [44], heart disease [39,47,48], and solid tumor cancers [40,49].

5. MSCs for the treatment of heart disease

There are many different heart conditions for which stem cell treatments are potentially valuable. The rationale to use MSCs to treat heart conditions is based on the ability of MSCs to home to areas of injury/inflammation and/or on their ability to down regulate the immune response and support the tissue repair process. Stem cells may be shown to reduce the amount of scar tissue and increase the pumping strength of the heart in myocardial infarcted patients.

According to the National Heart, Lung and Blood Institute, 1.1 million people suffer heart attacks in United States annually. Coronary heart disease, which causes heart attacks and angina, is a leading cause of death in the United States with nearly 450,000 related fatalities in 2005, according to the American Heart Association. As such, cellular therapies to treat cardiac disease are aggressively being pursued, and cardiac stem cell therapies could be commonplace within several years.

BM-MSCs have been shown to benefit patients early after myocardial infarction by exhibiting lower incidence of arrhythmias [47,48,50]. A recent study using a mesenchymal stem cell therapy presented by Osiris Therapeutics, Inc. showed that an intravenous injection of bone marrow-derived mesenchymal stem cells

Please cite this article in press as: Taghizadeh RR, et al., Wharton's Jelly stem cells: Future clinical applications, Placenta (2011), doi:10.1016/j.placenta.2011.06.010

repaired heart damage in patients who had experienced heart attacks within 10 days [47,48]. The trial now has moved to a phase II study in 50 hospitals in the United States. However, one of the limitations of BM-MSCs cell therapies is the difficulty to expand early-passage BM-MSCs to sufficient numbers and doses required to have a therapeutic benefit in the patient [24,33]. This is due primarily to increased senescence of BM-MSCs when expanded *in vitro* (unpublished data).

6. MSCs for the treatment of cancer

One of the other exciting research areas that utilize MSCs for cellular therapies is in the field of cancer. Two recent studies examine ability of MSCs to home to tumors to treat metastatic cancer [40,49]. Progress of this kind would be a major breakthrough in the treatment of solid tumors. The first study from the London Research Institute reported that MSCs engineered to produce and deliver tumor necrosis factor-related apoptosis-inducing ligand (TRAIL) home to and kill cancer cells in a lung metastatic cancer model. This study was the first to show a significant reduction in metastatic tumor burden with frequent eradication of metastases using induced TRAIL expressing MSCs. The researchers concluded that this method 'would have a wide potential therapeutic role, including the treatment of both primary tumors and their metastases, possibly as an adjuvant therapy in clearing micro-metastatic disease following primary tumor resection' [49]. TRAIL has also been used with umbilical cord blood [51].

A second study takes advantage of MSC homing capacities by combinatorial treatment with intraperitoneal (IP) injections of 5-fluorouracil (5-FU) and targeted interferon beta (IFN-β) gene therapy in UC-MSCs to treat metastatic human breast cancer in SCID mouse lung cancer [40]. UC-MSCs were found in the lung and not in other observed tissue, although this is not surprising as there is overwhelming evidence that intravenous injected cells initially home to the lung [52]. Although both treatments alone, significantly resulted in reductions in lung tumor area, the combined treatment of IFN-β transduced UC-MSCs and 5-FU resulted in greater lung tumor reduction, compared to each treatment alone [40]. Although only two cancer studies utilizing MSCs are highlighted here, the use of MSCs to combat tumors continues to progress toward therapeutic utilization.

7. Conclusion

Although there are no current clinical trials ongoing with WJSCs or UC-MSCs, several pre-clinical trials have been conducted to suggest the possible clinical benefits of this cell source. Several indications have been investigated in animals including hematopoietic reconstitution [20,38], Parkinson's [24], diabetes [45,46], Macular Degeneration [53] and spinal cord injuries [54]. Before WJSCs can be safely translated into human trials, further investigation and characterization in animals must be completed to ensure safety and efficacy. WJSCs are immuno-privileged, immunosuppressive, have a multipotent/pluripotent differentiation capacity and are readily available as a cell source; WJSCs may be an important cell therapy source for specific indications in the near future to treat several diseases and improve the quality of life in many patients.

Conflict of interest

RRT and KJC are employees of AuxoCell Laboratories, Inc. CLC is on the Board of AuxoCell Laboratories, Inc. and has worked as a consultant for AuxoCell Laboratories, Inc.

References

[1] Boissel L, Betancur M, Klingemann H, Marchand J. In: Cetrulo KJ, Cetrulo CL, Jr., Cetrulo CL, editors. Umbilical cord mesenchymal stem cells. Hoboken, NJ: Wiley-Blackwell; 2009. p. 69–75.
[2] Breymann C. In: Cetrulo KJ, Cetrulo CL, Jr., Cetrulo CL, editors. Use of fetal cells in regenerative medicine. Hoboken, NJ: Wiley-Blackwell; 2009. p. 41–8.
[3] Cetrulo Jr CL, Starnes MJ. In: Cetrulo KJ, Cetrulo CL, Jr., Cetrulo CL, editors. Perinatal endothelial progenitor cells. Hoboken, NJ: Wiley-Blackwell; 2009. p. 95–9.
[4] Lutjemeier B, Troyer DL, Weiss ML. In: Cetrulo KJ, Cetrulo CL, Jr., Cetrulo CL, editors. Wharton's jelly-derived mesenchymal stromal cells. Hoboken, NJ: Wiley-Blackwell; 2009. p. 79–90.
[5] Marongiu F, Gramignoli R, Miki T, Ranade A, Ellis ECS, Dorko K, et al. In: Cetrulo KJ, Cetrulo CL, Jr., Cetrulo CL, editors. Amniotic epithelial stem cells in regenerative medicine. Hoboken, NJ: Wiley-Blackwell; 2009. p. 159–65.
[6] Park H-J, Zhang Y, Naggar J, Georgescu SP, Kong D, Galper JB. In: Cetrulo KJ, Cetrulo CL, Jr., Cetrulo CL, editors. Human umbilical vein endothelial cells and human dermal microvascular endothelial cells offer new insights into the relationship between lipid metabolism, angiogenesis, and abdominal aortic aneurysm. Hoboken, NJ: Wiley-Blackwell; 2009. p. 169–85.
[7] Rhodes KE, Mikkola HKA. In: Cetrulo KJ, Cetrulo CL, Jr., Cetrulo CL, editors. Hematopoietic stem cell development in the placenta. Hoboken, NJ: Wiley-Blackwell; 2009. p. 189–99.
[8] Taghizadeh RR, Sherley JL. In: Cetrulo KJ, Cetrulo CL, Jr., Cetrulo CL, editors. Expanding the therapeutic potential of umbilical cord blood hematopoietic stem cells. Hoboken, NJ: Wiley-Blackwell; 2009. p. 21–7.
[9] Tsai M- S. In: Cetrulo KJ, Cetrulo CL, Jr., Cetrulo CL, editors. Amniotic fluid derived stem cells. Hoboken, NJ: Wiley-Blackwell; 2009. p. 147–55.
[10] Jones JM, Thomson JA. Human embryonic stem cell technology. Seminars in Reproductive Medicine 2000;18(02):219–24.
[11] Odorico JS, Kaufman DS, Thomson JA. Multilineage differentiation from human embryonic stem cell lines. Stem Cells 2001;19(3):193–204.
[12] Potten C. Cell replacement in epidermis (keratopoiesis) via discrete units of proliferation. International Review of Cytology 1981;69:271–318.
[13] Carlin R, Davis D, Weiss M, Schultz B, Troyer D. Expression of early transcription factors Oct-4, Sox-2 and Nanog by porcine umbilical cord (PUC) matrix cells. Reproductive Biology and Endocrinology 2006;4(1):8.
[14] Mitjavila-Garcia MT, Simonin C, Peschanski M. Embryonic stem cells: meeting the needs for cell therapy. Advanced Drug Delivery Reviews 2005;57(13):1935–43.
[15] Hyun I. The bioethics of stem cell research and therapy. The Journal of Clinical Investigation 2010;120(1):71–5.
[16] Can A, Karahuseyinoglu S. Concise review: human umbilical cord Stroma with Regard to the source of fetus-derived stem cells. Stem Cells 2007;25(11):2886–95.
[17] Secco M, Zucconi E, Vieira NM, Foga¡a LLQ, Cerqueira A, Carvalho MDF, et al. Multipotent stem cells from umbilical cord: cord is richer than blood! Stem Cells 2008;26(1):146–50.
[18] Troyer DL, Weiss ML. Concise review: Wharton's jelly-derived cells are a primitive stromal cell population. Stem Cells 2008;26(3):591–9.
[19] Fong C-Y, Chak L-L, Biswas A, Tan J-H, Gauthaman K, Chan W-K, et al. Human Wharton's jelly stem cells have unique transcriptome profiles compared to human embryonic stem cells and other mesenchymal stem cells. Stem Cell Reviews and Reports 2011;7(1):1–16.
[20] Friedman R, Betancur M, Boissel L, Tuncer H, Cetrulo C, Klingemann H. Umbilical cord mesenchymal stem cells: adjuvants for human cell transplantation. Biology of Blood and Marrow Transplantation: Journal of the American Society for Blood and Marrow Transplantation 2007;13(12):1477–86.
[21] Wang X-Y, Lan Y, He W-Y, Zhang L, Yao H-Y, Hou C-M, et al. Identification of mesenchymal stem cells in aorta-gonad-mesonephros and yolk sac of human embryos. Blood 2008;111(4):2436–43.
[22] Dominici M, Le Blanc K, Mueller I, Slaper-Cortenbach I, Marini F, Krause D, et al. Minimal criteria for defining multipotent mesenchymal stromal cells. The International Society for cellular therapy position statement. Cytotherapy 2006;8(4):315–7.
[23] Fong CY, Richards M, Manasi N, Biswas A, Bongso A. Comparative growth behaviour and characterization of stem cells from human Wharton's jelly. Reproductive Biomedicine Online 2007;15(6):708–18.
[24] Weiss ML, Medicetty S, Bledsoe AR, Rachakatla RS, Choi M, Merchav S, et al. Human umbilical cord matrix stem cells: preliminary characterization and effect of transplantation in a Rodent model of Parkinson's disease. Stem Cells 2006;24(3):781–92.
[25] La Rocca G, Anzalone R, Corrao S, Magno F, Loria T, Lo Iacono M, et al. Isolation and characterization of Oct-4+/HLA-G+ mesenchymal stem cells from human umbilical cord matrix: differentiation potential and detection of new markers. Histochemistry and Cell Biology 2009;131(2):267–82.
[26] Zhao R, Daley GQ. From fibroblasts to iPS cells: induced pluripo tency by defined factors. Journal of Cellular Biochemistry 2008;105(4):949–55.
[27] Takahashi K, Tanabe K, Ohnuki M, Narita M, Ichisaka T, Tomoda K, et al. Induction of pluripotent stem cells from adult human fibroblasts by defined factors. Cell 2007;131(5):861–72.
[28] Liu TM, Wu YN, Guo XM, Hui JHP, Lee EH, Lim B. Effects of ectopic nanog and Oct4 overexpression on mesenchymal stem cells. Stem Cells and Development 2009;18(7):1013–22.

Please cite this article in press as: Taghizadeh RR, et al., Wharton's Jelly stem cells: Future clinical applications, Placenta (2011), doi:10.1016/j.placenta.2011.06.010

[29] Campagnoli C, Roberts IAG, Kumar S, Bennett PR, Bellantuono I, Fisk NM. Identification of mesenchymal stem/progenitor cells in human first-trimester fetal blood, liver, and bone marrow. Blood 2001;98(8):2396–402.
[30] Gotherstrom C, Ringden O, Westgren M, Tammik C, Le Blanc K. Immunomodulatory effects of human foetal liver-derived mesenchymal stem cells. Bone Marrow Transplant 2003;32(3):265–72.
[31] Gotherstrom C, West A, Liden J, Uzunel M, Lahesmaa R, Le Blanc K. Difference in gene expression between human fetal liver and adult bone marrow mesenchymal stem cells. Haematologica 2005;90(8):1017–26.
[32] Guillot PV, Gotherstrom C, Chan J, Kurata H, Fisk NM. Human first-trimester fetal MSC express pluripotency markers and Grow Faster and have longer telomeres than adult MSC. Stem Cells 2007;25(3):646–54.
[33] Schugar RC, Chirieleison SM, Wescoe KE, Schmidt BT, Askew Y, Nance JJ, et al. High harvest yield, high expansion, and phenotype stability of CD146 mesenchymal stromal cells from whole primitive human umbilical cord tissue. Journal of Biomedicine and Biotechnology; 2009. Article ID 789526:11.
[34] Le Blanc K. Immunomodulatory effects of fetal and adult mesenchymal stem cells. Cytotherapy 2003;5(6):485–9.
[35] Marigo I, Dazzi F. The immunomodulatory properties of mesenchymal stem cells. Seminars in Immunopathology; 2011:1–10.
[36] Wang M, Yang Y, Yang D, Luo F, Liang W, Guo S, et al. The immunomodulatory activity of human umbilical cord blood-derived mesenchymal stem cells in vitro. Immunology 2009;126(2):220–32.
[37] Weiss ML, Anderson C, Medicetty S, Seshareddy KB, Weiss RJ, VanderWerff I, et al. Immune properties of human umbilical cord Wharton's jelly-derived cells. Stem Cells 2008;26(11):2865–74.
[38] Taghizadeh RR, Pollok KE, Betancur M, Boissel L, Cetrulo KJ, Marino T, et al. Wharton's jelly derived mesenchymal stem cells: regenerative medicine beyond umbilical cord blood. Placenta; 2011 (accepted).
[39] Boomsma RA, Swaminathan PD, Geenen DL. Intravenously injected mesenchymal stem cells home to viable myocardium after coronary occlusion and preserve systolic function without altering infarct size. International Journal of Cardiology 2007;122(1):17–28.
[40] Rachakatla RS, Pyle MM, Ayuzawa R, Edwards SM, Marini FC, Weiss ML, et al. Combination treatment of human umbilical cord matrix stem cell-based interferon-beta gene therapy and 5-fluorouracil significantly reduces growth of metastatic human breast cancer in SCID mouse lungs. Cancer Investigation 2008;26(7):662–70.
[41] Bernardo ME, Pagliara D, Locatelli F. Mesenchymal stromal cell therapy: a revolution in regenerative medicine [quest]. Bone Marrow Transplant; 2011.
[42] Caimi PF, Reese J, Lee Z, Lazarus HM. Emerging therapeutic approaches for multipotent mesenchymal stromal cells. Current Opinion in Hematology 2010;17(6):505–13.
[43] Gonzalo-Daganzo R, Regidor C, Martin-Donaire T, Rico MA, Bautista G, Krsnik I, et al. Results of a pilot study on the use of third-party donor mesenchymal stromal cells in cord blood transplantation in adults. Cytotherapy 2009;11(3):278–88.
[44] Tyndall A, Uccelli A. Multipotent mesenchymal stromal cells for autoimmune diseases: teaching new dogs old tricks. Bone Marrow Transplant 2009;43(11):821–8.
[45] Anzalone R, Lo Iacono M, Loria T, Di Stefano A, Giannuzzi P, Farina F, et al. Wharton's jelly mesenchymal stem cells as candidates for beta cells regeneration: extending the differentiative and immunomodulatory benefits of adult mesenchymal stem cells for the treatment of type 1 diabetes. Stem Cell Reviews and Reports 2011;7(2):342–63.
[46] Chao KC, Chao KF, Fu YS, Liu SH. Islet-like clusters derived from mesenchymal stem cells in Wharton's jelly of the human umbilical cord for transplantation to control type 1 diabetes. PLoS ONE 2008;3(1):e1451.
[47] Amado LC, Saliaris AP, Schuleri KH, St. John M, Xie J-S, Cattaneo S, et al. Cardiac repair with intramyocardial injection of allogeneic mesenchymal stem cells after myocardial infarction. Proceedings of the National Academy of Sciences of the United States of America 2005;102(32):11474–9.
[48] Hare JM, Traverse JH, Henry TD, Dib N, Strumpf RK, Schulman SP, et al. Double-blind, placebo-controlled, dose-escalation study of intravenous adult human mesenchymal stem cells (prochymal) after acute myocardial infarction. Journal of the American College of Cardiology 2009;54(24):2277–86.
[49] Loebinger MR, Eddaoudi A, Davies D, Janes SM. Mesenchymal stem cell delivery of TRAIL can eliminate metastatic cancer. Cancer Research 2009;69(10):4134–42.
[50] Kawada H, Fujita J, Kinjo K, Matsuzaki Y, Tsuma M, Miyatake H, et al. Nonhematopoietic mesenchymal stem cells can be mobilized and differentiate into cardiomyocytes after myocardial infarction. Blood 2004;104(12):3581–7.
[51] Kim SM, Lim JY, Park SI, Jeong CH, Oh JH, Jeong M, et al. Gene therapy using TRAIL-secreting human umbilical cord blood-derived mesenchymal stem cells against intracranial glioma. Cancer Research 2008;68(23):9614–23.
[52] Noort WA, Kruisselbrink AB, in't Anker PS, Kruger M, van Bezooijen RL, de Paus RA, et al. Mesenchymal stem cells promote engraftment of human umbilical cord blood derived CD34+ cells in NOD/SCID mice. Experimental Hematology 2002;30(8):870–8.
[53] Lund RD, Wang S, Lu B, Girman S, Holmes T, Sauv» Y, et al. Cells isolated from umbilical cord tissue rescue photoreceptors and visual functions in a rodent model of retinal disease. Stem Cells 2007;25(3):602–11.
[54] Yang C-C, Shih Y-H, Ko M-H, Hsu S-Y, Cheng H, Fu Y-S. Transplantation of human umbilical mesenchymal stem cells from Wharton's jelly after complete transection of the rat spinal cord. PLoS ONE 2008;3(10):e3336.

Please cite this article in press as: Taghizadeh RR, et al., Wharton's Jelly stem cells: Future clinical applications, Placenta (2011), doi:10.1016/j.placenta.2011.06.010