ALMA MATER STUDIORUM
UNIVERSITÀ DI BOLOGNA

ARCHIVIO ISTITUZIONALE
DELLA RICERCA

Alma Mater Studiorum Università di Bologna
Archivio istituzionale della ricerca

Stem cells from foetal adnexa and fluid in domestic animals: An update on their features and clinical application

This is the final peer-reviewed author's accepted manuscript (postprint) of the following publication:

Published Version:
Iacono, E., Rossi, B., Merlo, B. (2015). Stem cells from foetal adnexa and fluid in domestic animals: An update on their features and clinical application. REPRODUCTION IN DOMESTIC ANIMALS, 50(3), 353-364 [10.1111/rda.12499].

Availability:
This version is available at: https://hdl.handle.net/11585/525148 since: 2019-07-15

Published:
DOI: http://doi.org/10.1111/rda.12499

Terms of use:

Some rights reserved. The terms and conditions for the reuse of this version of the manuscript are specified in the publishing policy. For all terms of use and more information see the publisher's website.

This item was downloaded from IRIS Università di Bologna (https://cris.unibo.it/).
When citing, please refer to the published version.

(Article begins on next page)

23 July 2024

This is the peer reviewed version of the following article:

Stem cells from foetal adnexa and fluid in domestic animals: an update on their features and clinical application.

which has been published in final form at 10.1111/rda.12499.

This article may be used for non-commercial purposes in accordance with Wiley Terms and Conditions for Use of Self-Archived Versions.

Department of Veterinary Medical Sciences, University of Bologna, Ozzano Emilia (Bo), Italy

Stem cells from foetal adnexa and fluid in domestic animals: an update on their features and clinical application.

Running Title: MSCs from foetal adnexa in domestic animals

Eleonora Iacono*, DVM, PhD, Professor

Barbara Rossi, Bsc, PhD student

Barbara Merlo, DVM, PhD, ECAR Diplomate, Professor

*Corresponding Author: Eleonora Iacono, Department of Veterinary Medical Sciences, University of Bologna, via Tolara di Sopra 50, 40064, Ozzano Emilia (Bo), Italy.

Phone number: +39-051-2097567

e-mail: eleonora.iacono2@unibo.it

Summary

Over the past decade stem cell research has emerged as an area of major interest for its potential in regenerative medicine applications. This is in constant need of new cell sources to conceive regenerative medicine approaches for diseases that are still without therapy. Scientists drew the attention toward alternative sources such as foetal adnexa and fluid, since these sources possess many advantages: first of all, cells can be extracted from discarded foetal material and it is noninvasive for the patient and inexpensive, secondly abundant stem cells can be obtained and finally, these stem cell sources are free from ethical considerations. Cells derived from foetal adnexa and fluid preserve some of the characteristics of the primitive embryonic layers from which they originate. Many studies have demonstrated the differentiation potential in vitro and in vivo toward mesenchymal and non-mesenchymal cell types; in addition the immune-modulatory properties make these cells a good candidate for allo- and xenotransplantation. Naturally occurring diseases in domestic animals can be more ideal as disease model of human genetic and acquired diseases and could help to define the potential therapeutic use efficiency and safety of stem cells therapies. This review offers an update on the state of the art of characterization of domestic animals MSCs derived from foetal adnexa and fluid and on the latest findings in pre-clinical or clinical setting of the stem cell populations isolated from these sources.

Key words: Amniotic fluid, Amniotic Membrane, Umbilical cord blood, Wharton's Jelly, Mesenchymal Stem Cells

Introduction

A stem cell is an undifferentiated cell capable of differentiating into specialized cells and to originate other stem cells. In mammals, there are three types of stem cells (SCs): embryonic stem cells (ESCs), foetal stem cells, and adult stem cells. ESCs derive from the inner cell mass (ICM) of a blastocyst. They are pluripotent but highly tumorigenic after transplantation. Foetal stem cells are distinguished into two types: foetal proper stem cells (from the tissue of the fetus), and extra-embryonic foetal stem cells (from extra-embryonic membranes). Adult SCs are multipotent cells, designate to tissue repairing, and they are present in almost all adult tissues.

Mesenchymal stem cells (MSCs) are a population of multipotent stem cells that meet the following criteria: 1) plastic-adherence when maintained in standard culture conditions; 2) expression of CD105, CD73 and CD90, and lack of CD45, CD34, CD14 or CD11b, CD79α or CD19 and HLA-DR surface molecules; 3) differentiation to osteoblasts, adipocytes and chondroblasts in vitro (Dominici et al. 2006).

Since these properties, MSCs offer a great chance for cell-based therapies and tissue engineering applications. The effective management of companion animals, such as dog, cat or horse, requires sophisticated new treatments and preventive strategies. This, combined with their role as models for human diseases, led to an increase in the number of research focused on isolation and clinical use of MSCs in these animals.

Bone marrow (BM) is the common source of autologous MSCs for clinical applications in veterinary medicine. Alternatively, adipose tissue-derived MSCs can be used, since they have a higher proliferation potential. Anyway, for both sources, an invasive procedure is required and there is a large variability in the cell yield related to the donor (Colleoni et al. 2009).

Placental tissues and foetal fluids represent a source of cells for regenerative medicine, and are readily available and easily procured without invasive procedures. MSCs from foetal

fluids and adnexa are defined as an intermediate between ESCs and adult SCs, due to the preservation of some characteristics typical of the primitive native layers. Among foetal adnexal tissues, the major sources of MSCs are amnion, amniotic fluid, umbilical cord and umbilical cord blood. These cells can be obtained in large numbers without risks for donors and cryopreserved for future use in regenerative veterinary medicine.

The markers required for defining a MSC were determined for human cells, and the lack of specific antibodies for different animal species lead to a great variability of approaches and results for molecular characterization of animal MSCs. The aim of this review is to provide an overview of the advances in characterization and clinical applications of domestic animal MSCs derived from foetal adnexa and fluid.

Developmental biology of foetal adnexa and amniotic fluid

Extra-embryonic or foetal membranes are structures that develop from the zygote, do not form part of the embryo itself, and are of functional importance only in embryonic life. At the end of cleavage, the blastocysts, enclosed within the zona pellucida, consists of an ICM and a trophoblastic layer. During the second week of development, hatched blastocyst leaves its confined space and elongates. In particular this process involves the trophoblast, and, partially, the inner endodermal lining. The head and tail of growing embryo, progressively, push deeper in to the trophoblast, which become lined by a layer of somatic mesoderm forming the extra-embryonic somatopleure (McGeady et al. 2006). The outer somatopleure membrane forms chorion, while the inner somatopleure membrane, attached to the embryos at umbilicus, forms amnion. Due to its early development, amnion could be a reservoir of pluripotent stem cells, derived from the contiguity of embryonic epiblast, even at full term pregnancy. Amnion is a thin, tough, membranous and fluid-filled sac which surrounds and protects the embryo and then the foetus (McGeady et al. 2006). When first formed, it is in

contact with the embryo's body, but, after few weeks of pregnancy, fluid begins to accumulate within it. This fluid increases in quantity relatively rapidly up to mid-pregnancy and then gradually decreases. In early pregnancy, the composition of amniotic fluid and plasma are similar and may be considered a dialysate of the maternal or foetal extracellular fluid. Later in gestation, when the foetus urethra becomes patent, foetal urine passes in amniotic cavity. Amniotic fluid (AF) contains less than 2% of solids, consisting of urea and other extractives, inorganic salts, a small amount of protein, a trace of sugar and multiple cell types from embryonic and extra-embryonic tissues (Gosden 1983).

With the progress of gestation, the embryo sinks into the amniotic membrane (AM), which wraps the mesoblastic axis in which allantois and umbilical vessels developed, giving rise to the umbilical cord (UC) (Barone 1994). Based on structural and functional studies, at least four distinctive zones are now recognized in the UC: surface amniotic epithelium, subamniotic stroma, perivascular stroma, vessels. All together these structures are called umbilical cord matrix (UCM). The connective tissue included between subamnion and perivascular regions consists of mucous, mesenchymal connective tissue, and it is called Wharton's jelly (WJ). It contains a very low number of cells and a high amount of extracellular matrix components, such as collagen, hyaluronic acid and proteoglycans (Sobolewski et al. 1997). These substances make this tissue highly hydrated and resistant to extension and compression evoked by foetal movements and uterine contraction. Stromal cells of WJ, previously described as myofibroblast (Takechi et al. 1993), have properties of potentially multipotent SCs (Mitchell et al. 2003).

Amniotic membrane

After mechanically separating amnion from allantois, as in human medicine, it is possible to harvest two cellular types, amniotic epithelial (AECs) or mesenchymal cells (AMSCs). AECs

have a typical polygonal morphology while AMSCs are fibroblast-like. Both AMSCs and AECs from different species have been investigated for proliferative potential, differentiation ability, and phenotypically characterized.

AECs showed similar patterns to AMSCs in the sheep (Gloria et al. 2010; Mauro et al. 2010; Mattioli et al. 2012), but the latter were superior in promoting interbody fusion in induced anterior cervical discectomy after allogenic implantation combined with bone grafts (Goldschlager et al. 2011). AECs phenotype, methylation status, immunomodulatory and stemness properties are affected by gestational age (Barboni et al. 2014). In the horse (Lange-Consiglio et al. 2012) and in the bovine (Corradetti et al., 2013), AECs showed a lower number of doublings compared to AMSCs but the same phenotype and plasticity at differentiation. Feline AECs were fully characterized and were able to differentiate into adipogenic, chondrogenic, osteogenic and neurogenic lineages (Rutigliano et al. 2013). Porcine AECs were successfully used for construction of tissue-engineered cornea (Luo et al. 2013).

Focusing on MSCs, they express pluripotency and mesenchymal markers, have a multi-lineage differentiation potential and possess a privileged immunogenic status related to the low expression or even absent levels of MHC I and MHC II (data are summarized in Table 1). AMSCs were capable of inhibiting peripheral blood mononuclear cell proliferation after allogenic stimulation either when co-cultured in cell-to-cell contact or when separated by a transwell membrane (Lange-Consiglio et al. 2013b). Furthermore, it was demonstrated that AMSCs possess nonteratogenicity 4 weeks post injection into immunodeficient mice (Vidane et al. 2014).

Comparing results from different species, among pluripotency markers, a difference was found for NANOG expression. While in the horse (Violini et al. 2012) and buffalo (Dev et al. 2012; Mann et al. 2013) AMSCs expressed NANOG at RT-PCR, canine AMSCs were

151 negative (Filioli Uranio et al. 2011, 2014). Anyway, Park et al. (2012) could quantified it in
152 canine AMSCs using qPCR.
153 Mesenchymal markers were expressed in all species while haematopoietic markers were not
154 found. In the horse, CD34 was not expressed in early stages of culture, but began to be
155 expressed at later passages (P5) (Lange-Consiglio et al. 2013a). By the way, the consensus
156 about absence of CD34 expression on MSCs has been challenged. The CD34 expression
157 status in haematopoietic and MSCs appears to depend on the environment, and can change
158 from positive to negative, and vice versa (Lin et al. 2012).
159 Karyotype of cultured AMSCs was confirmed to be stable in the dog (Filioli Uranio et al.
160 2011, 2014), buffalo (Dev et al. 2012c; Mann et al. 2013) and horse (Seo et al. 2013). In the
161 dog, a high telomerase activity was found (Filioli Uranio et al. 2011). Telomerase activity and
162 telomere maintenance characterize all cells with self-renewal capability, such as ESCs, MSCs,
163 and most cancer cells (Colosimo et al. 2013a).
164
165 **Amniotic fluid**

166 In the veterinary field, as in the human, AF contains a heterogeneous population of cells:
167 cuboidal epitheloid (E-cells), round (R-cells) and spindle-shaped fibroblastic (F-cells) cells.
168 E-cells probably derive from foetal skin and urine and they are lost during the first passages
169 of culture; R-cells are supposed to originate from foetal membranes and trophoblasts; F-cells
170 are generated from mesenchymal tissues and are supposed to be the MSC population of the
171 AF (Klemmt et al. 2011). In domestic animals, the prevalent cell population that persist
172 during culture is the F-type, except for one study on porcine AFMSCs (Sartore et al. 2005)
173 and one on bovine AFMSCs (Rossi et al. 2014), where both F-cells and R-cells persist
174 through passages. Other groups, that studied bovine AF cell populations (Corradetti et al.
175 2013; Gao et al. 2014; da Cunha et al. 2014), found R-cells only at the very first passages,
176 maybe due to the addition in the culture medium of growth factors like EGF and bFGF.

AFMSCs generally express MSC markers and mesodermal markers like Vimentin and α-Smooth Muscle Actin (αSMA) and are negative for haematopoietic markers (Table 2). In the horse, expression of CD73 is controversy. Positivity was found only for AD2 clone, while clone 5F/B9 did not cross-react (Gulati et al. 2013). Anyway, for AD2 clone cross-reactivity was not found in equine (Iacono et al. 2012a) and also in feline (Iacono et al. 2012b) and bovine AFMSCs (Rossi et al. 2014). Like AMSCs, also AFMSCs express pluripotency markers and only in one study in the bovine (Rossi et al. 2014) expression of OCT4, NANOG and SOX2 was not found.

Different authors verified that AFMSCs show telomerase activity and that telomere length is not affected by culture conditions (Filioli Uranio et al. 2011; Colosimo et al. 2013a), such as karyotype (Filioli Uranio et al. 2011; Dev et al. 2012a; Dev et al. 2012b; Colosimo et al. 2013b; Weber et al. 2013; da Cunha et al. 2014).

Umbilical cord blood (UCB)

In human medicine, the use of UCB as a source of MSCs can be traced back to 2000: in this year Erices et al. showed that UCB cells were able to give rise to two types of adherent cells, one of which expressed antigens typical of MSCs. Also in veterinary medicine cells with characteristics similar to human UCBMSCSs have been isolated in different species (Table 3). Usually, an isolation rate of fibroblast-like, spindle-shaped cells, similar to human, between 50 and 70% is reported (Koch et al. 2007; Iacono et al. 2012a; Mohanty et al. 2014), except for feline UCB where cells at P0 comprised a heterogeneous population, including fibroblast-like, spindle-shaped, and small round morphologies (Jin et al. 2008). Reported data may be due to the limited number of MSCs in UCB as well as suboptimal isolation and expansion conditions. In horses was demonstrated that samples cultured at 5% O_2 proliferated rapidly to the desired cell number. The effect of mild hypoxia may be attributable in part to the

induction of signaling pathways but also to a decrease in survival of contaminating leukocytes in UCB, thus increasing the isolation rate up to 80% (Schuh et al. 2009). Furthermore, particularly in equine species, the UCB recovered volume is often low, because of a wide spread belief that the mare continues to pass blood to the foal through the umbilicus immediately post foaling (Rossdale 1958). However, using Doppler ultrasound, Doarn (1985) showed that blood flow through the cord ceases within few minutes following delivery. All authors reported that UCBMSCs cells from domestic animals sources were positive for CD44 (Table 3), an antigen expressed by a variety of cells types, not part of the ISCT MSC definition (Dominici et al. 2006). Given its constant expression and considering the difficulty in UCBMSCs isolation, sorting of CD44+ cells prior to plastic adherence isolation could lead to more homogenous populations in domestic animals (Paebst et al. 2014). A variability of the other MSCs marker expression exists particularly for CD73. Similarly to AFMSCs, in the horse only Mohanty et al. (2014) showed that UCBMSCs expressed CD73, both with RT-PCR and flow cytometry. Authors explained their finding with the use of different CD73 clones (5F/B9), compared with a previous study reporting its negative expression (De Schauwer et al. 2012; 10fl clone). However, Mohanty's research group stated that equine AFMSCs reacted with AD2 clone, but not with 5F/B9 clone (Gulati et al. 2013), showing opposing results for the same species. Anyway, differences in the culture medium, such as the content of FBS, the timeframe of plastic-adherent culture and the cell harvesting technique could influence the cell characteristics (Paebst et al. 2014).

UCBMSCs from different species possess an intermediate phenotype that more closely resembles ES cells: Tra1-60, Tra1-81 and Oct4 are present (Reed et al. 2008; Seo et al. 2009; Raoufi et al. 2011; Mohanty et al. 2014), but conflicting data are reported for Nanog and Sox2 (Reed et al. 2008; Mohanty et al. 2014), particularly in the horse.

Umbilical Cord

UC is routinely discarded at parturition and its extracorporeal nature facilitates isolation by eliminating the invasive and discomfort extraction procedures as well as patient risks that attend adult stem cell isolation. Most significantly, the comparatively large volume of UC and ease of physical manipulation theoretically increase the number of stem cells that can be extracted, which make it possible to get substantial number of cells in several passages without need of long term culture and extensive expansion ex vivo. Probably due to the possible absence of this structure at birth, because of reduction of the water and substances content towards the end of pregnancy, different authors, particularly in veterinary medicine, named interchangeably WJ or UCM. Due to this reason, in Table 4, we reported data regarding WJ and UCM MSCs together. Usually, spindle-shaped cells have been isolated; however few authors described a heterogeneous cell population (fusiform or spindle form and small round cells with large and prominent nucleus), from porcine and caprine UC, characterized by a plateau or a stationary phase (Mitchell et al. 2003; Babaei et al. 2008). In these species, such as in canine (Filioli Uranio et al. 2011) and bovine (Cardoso et al. 2012), UC cells expressed high levels of telomerase activity, and this phenomenon could explain the long term of cell culture observed by the authors. Moreover, tissue culture procedure does not alter chromosomal organization (Filioli Uranio et al. 2011; Cardoso et al. 2012; da Cunha et al. 2014). As reported above, also cell isolated from UC are negative for hematopoietic markers and express a number of antigens associated with adult MSCs (Table 4). Conflicting data are reported regarding their expression of embryonic markers. Cells from porcine, bubaline and equine UC resulted positive for pluripotency genes OCT4, NANOG, SOX2 at different culture passages (Mitchell et al. 2003; Carlin et al. 2006; Hoynowsky et al. 2007; Singh et al. 2013; Sreekumar et al. 2014). In canine, all authors agree that during in vitro passages cells lost OCT4 expression. However, while Lee et al. (2013a) stated that expression of NANOG and SOX2 is negatively correlated with the number of passages but is always

present, most recently Filioli Uranio et al. (2014) showed that NANOG gene is present only in cells recovered at late gestational age. This discrepancy is likely due to different culture conditions and to different selection of cord tissue to be processed. The immature cells that retain the ability to proliferate were located close to the amniotic surface, whereas highly differentiated, non-proliferating fibroblasts were located in closer proximity to the umbilical vessels (Nanaev et al. 1997).

Clinical Applications

Clinical application of foetal MSCs in domestic animals had an increase in the last five years. The majority of the studies are on AFMSCs, mainly used in animal-model diseases (heart, diaphragm, trachea and tendon induced injuries in different species) (Table 5). Among tissue-engineering studies in the sheep, diaphragmatic repair with a mesenchymal amniocyte-based engineered tendon led to improved structural outcomes when compared with equivalent fetal myoblast-based and acellular graft (Kunisaki et al. 2006c). Investigating the ability of AFMSCs for phenotypic conversion to vascular cells and cardiomyocytes (CM) when autotransplanted in a porcine model acute ischemic myocardium (AMI), MSCs were able to transdifferentiate to cells of vascular cell lineages but not to CM. Thus, porcine AFMSCs may require further ex vivo re-programming to be suitable for therapeutic use in AMI (Sartore et al. 2005). On the other hand, intramuscular injection of porcine AFMSCs reduced scar size and preserved heart function after induced myocardial infarction in mice (Peng et al. 2014). Only in the horse AFMSCs were used, combined with PRP (plateled rich plasma), to treat spontaneous decubitus ulcers (Iacono et al. 2012c). When comparing different treatments (aloe gel, PRP alone, MSCs+PRP), mean regression of ulcer treated with MSCs+PRP was higher than others.

Despites animal UC cells molecular characterization, in vitro differentiation and their immunosuppressive effects on T-cells in vitro (Cardoso et al. 2012), there are sparse literatures documenting their therapeutic applications (Table 5). Equine UCBMSCs were successfully used as allogenic therapy in race-horses with spontaneous tendon lesions (Kang et al. 2013; Van Loon et al. 2014). Canine UCBMSCs and WJMSCs were mainly applied to treat induced spinal cord injuries or bone defects. Dog WJMSCs can replace adult MSCs in clinical bone engineering procedures (Kang et al. 2012) and promote functional recovery in spinal cord injured dogs after allogenic transplantation (Ryu et al. 2012). Caprine WJMSCs allogenic (Azari et al. 2011) and xenogenic transplantation (Pratheesh et al. 2014) in a rabbit model, could be successfully performed for treating induced skin wounds. In not-treated wounds, incomplete re-epithelialization and thick granulation tissue were observed, whereas, in treated wounds intact skin with complete re-epithelialization, no inflammation, and thin granulation tissue were seen. Furthermore, in both studies, histomorphological evaluation demonstrated better epithelization, lower neovascularisation, and denser collagen fibres in treated wounds.

Although implantation of human AMSCs into animals resulted in successful and persistent engraftment in multiple organs and tissues (Lindenmair et al. 2012), their application in veterinary medicine are still limited (Table 5). In fact, except for a study to enhance cervical interbody fusion in an ovine model (Goldschlader et al. 2011) and histological and functional improvement after transplantation in a porcine model of chronic myocardial ischemia (Kimura et al. 2012), AMSCs were only applied in equine spontaneous tendon injuries. Allogenic transplantation resulted in a quick reduction in tendon size and ultrasonographic cross-sectional area (Lange-Consiglio et al. 2012) and the re-injury rate was lower compared to autologous BMMSCs transplantation (Lange-Consiglio et al. 2013c). The hypothesis of a paracrine mechanism of tendon repair when AMSCs are injected was confirmed in vivo; when horse tendon injuries were treated with conditioned medium, a lower rate of re-injuries

was observed compared to untreated animals (Lange-Consiglio et al. 2013b). AMSCs and their conditioned medium were recently considered candidates for equine uterine regenerative therapy, due to their ability to increase in vitro endometrial cell proliferation rate (Corradetti et al. 2014). So far, AMSCs are promising not only for cell-based therapies, but also for novel therapeutic biological cell-free products.

Conclusion

Altogether, these studies offer authoritative views on markers expression and therapeutic potential of MSCs from foetal tissues and fluids in domestic animals. These sources are easily available and relatively abundant, so MSCs may have an attraction compared to other established SCs in different clinical approaches. Anyway, more clinical applications are needed to fully understand their properties and to establish the future clinical use in the treatment of various diseases.

Acknowledgment

Barbara Rossi was partially funded by RFO to Professor Cesare Galli.

References

Azari O, Babaei H, Derakhshanfar A, Nematollahi-Mahani SN, Poursahebi R, Moshrefi M, 2011: Effects of transplanted mesenchymal stem cells isolated from Wharton's jelly of caprine umbilical cord on cutaneous wound healing; histopathological evaluation. Vet Res Commun **35**, 211-222.

Babaei H, Moshrefi M, Golchin M, Nematollahi-Mahani SN, 2008: Assess the pluripotency of caprine umbilical cord Wharton's jelly mesenchymal cells by RT-PCR analysis of early transcription factor Nanog. IJVS **3**, 57-65.

Barberini DJ, Freitas NP, Magnoni MS, Maia L, Listoni AJ, Heckler MC, Sudano MJ, Golim MA, da Cruz Landim-Alvarenga F, Amorim RM, 2014: Equine mesenchymal stem cells from bone marrow, adipose tissue and umbilical cord: immunophenotypic characterization and differentiation potential. Stem Cell Res Ther **21,** 25.

Barboni B, Russo V, Curini V, Martelli A, Berardinelli P, Mauro A, Mattioli M, Marchisio M, Bonassi Signoroni P, Parolini O, Colosimo A, 2014: Gestational stage affects amniotic epithelial cells phenotype, methylation status, immunomodulatory and stemness properties. Stem Cell Rev **10**, 725-741.

Barone R, 1994: Formazione degli annessi embrionali. In: Anatomia Comparata dei mammiferi domestici, Vol. 4. Edagricole, Bologna, pp.431-437.

Berardinelli P, Valbonetti L, Muttini A, Martelli A, Peli R, Zizzari V, Nardinocchi D, Vulpiani MP, Tetè S, Barboni B, Piattelli A, Mattioli M, 2013: Role of amniotic fluid mesenchymal cells engineered on MgHA/collagen-based scaffold allotransplanted on an experimental animal study of sinus augmentation. Clin Oral Investig **17**, 1661-1675.

Berg L, Koch T, Heerkens T, Bessonov K, Thomsen P, Betts D, 2009: Chondrogenic potential of mesenchymal stromal cells derived from equine bone marrow and umbilical cord blood. Vet Comp Orthop Traumatol **22**, 363-370.

Burk J, Ribitsch I, Gittel C, Juelke H, Kasper C, Staszyk C, Brehm W, 2013: Growth and differentiation characteristics of equine mesenchymal stromal cells derived from different sources. Vet J **195**, 98-106.

Byeon YE, Ryu HH, Park SS, Koyama Y, Kikuchi M, Kim WH, Kang KS, Kweon OK, 2010: Paracrine effect of canine allogenic umbilical cord blood-derived mesenchymal stromal cells mixed with beta-tricalcium phosphate on bone regeneration in ectopic implantations. Cytotherapy **12**, 626-636.

Cardoso TC, Ferrari HF, Garcia AF, Novais JB, Silva-Frade C, Ferrarezi MC, Andrade AL, Gameiro R, 2012a: Isolation and characterization of Wharton's jelly-derived multipotent mesenchymal stromal cells obtained from bovine umbilical cord and maintained in a defined serum-free three-dimensional system. BMC Biotechnol, **12**, 18.

Cardoso TC, Novais JB, Antello TF, Silva-Frade C, Ferrarezi MC, Ferrari HF, Gameiro R, Flores EF, 2012b: Susceptibility of neuron-like cells derived from bovine Wharton's jelly to bovine herpesvirus type 5 infections. BMC Vet Res **8**, 242.

Carlin R, Davis D, Weiss M, Schultz B, Troyer D, 2006: Expression of early transcription factors Oct-4, Sox-2 and Nanog by porcine umbilical cord (PUC) matrix cells. Reprod Biol Endocrinol **4**, 8.

Carrade DD, Owens SD, Galuppo LD, Vidal MA, Ferraro GL, Librach F, Buerchler S, Friedman MS, Walker NJ, Borjesson DL, 2011: Clinicopathologic findings following intra-articular injection of autologous and allogeneic placentally derived equine mesenchymal stem cells in horses. Cytotherapy **13**, 419-430.

Carrade DD, Lame MW, Kent MS, Clark KC, Walker NJ, Borjesson DL, 2012: Comparative Analysis of the Immunomodulatory Properties of Equine Adult-Derived Mesenchymal Stem Cells. Cell Med **4**, 1-11.

Chen J, Lu Z, Cheng D, Peng S, Wang H, 2011: Isolation and characterization of porcine amniotic fluid-derived multipotent stem cells. PLoS One, **6**, e19964.

Choi SA, Choi HS, Kim KJ, Lee DS, Lee JH, Park JY, Kim EY, Li X, Oh HY, Lee DS, Kim MK, 2013: Isolation of canine mesenchymal stem cells from amniotic fluid and differentiation into hepatocyte-like cells. In Vitro Cell Dev Biol Anim **49**, 42-51.

Co C, Vickaryous MK, Koch TG, 2014: Membrane culture and reduced oxygen tension enhances matrix formation from equine cord blood mesenchymal stromal cells in vitro. Osteoarthritis Cartilage **22**, 472-480.

Coli A, Nocchi F, Lamanna R, Iorio M, Lapi S, Urciuoli P, Scatena F, Giannessi E, Stornelli MR, Passeri S, 2011: Isolation and characterization of equine amnion mesenchymal stem cells. Cell Biol Int Rep **18**, e00011.

Colleoni S, Bottani E, Tessaro I, Mari G, Merlo B, Romagnoli N, Spadari A, Galli C, Lazzari G, 2009: Isolation, growth and differentiation of equine mesenchymal stem cells: effect of donor, source, amount of tissue and supplementation with basic fibroblast growth factor. Vet Res Commun **33**, 811-821.

Colosimo A, Curini V, Russo V, Mauro A, Bernabo N, Marchisio M, Alfonsi M, Muttini A, Mattioli M, Barboni B, 2013a: Characterization, GFP gene Nucleofection, and allotransplantation in injured tendons of ovine amniotic fluid-derived stem cells. Cell Transplant **22**, 99-117.

Colosimo A, Russo V, Mauro A, Curini V, Marchisio M, Bernabo N, Alfonsi M, Mattioli M, Barboni B, 2013b: Prolonged in vitro expansion partially affects phenotypic features and osteogenic potential of ovine amniotic fluid-derived mesenchymal stromal cells. Cytotherapy, **15**, 930-950.

Corradetti B, Lange-Consiglio A, Barucca M, Cremonesi F, Bizzaro D, 2011: Size-sieved subpopulations of mesenchymal stem cells from intervascular and perivascular equine umbilical cord matrix. Cell Prolif **44**, 330-342.

Corradetti B, Meucci A, Bizzaro D, Cremonesi F, Lange Consiglio A, 2013: Mesenchymal stem cells from amnion and amniotic fluid in the bovine. Reproduction **145**, 391-400.

Corradetti B, Correani A, Romaldini A, Marini MG, Bizzaro D, Perrini C, Cremonesi F, Lange-Consiglio A, 2014: Amniotic membrane-derived mesenchymal cells and their conditioned media: potential candidates for uterine regenerative therapy in the horse. PLoS One **9**, e111324.

Cremonesi F, Violini S, Lange Consiglio A, Ramelli P, Ranzenigo G, Mariani P, 2008: Isolation, in vitro culture and characterization of foal umbilical cord stem cells at birth. Vet Res Commun **32**, S139-S142.

da Cunha ER, Martins CF, Silva CG, Bessler HC, Báo SN, 2014: Effects of prolonged in vitro culture and cryopreservation on viability, DNA Fragmentation, chromosome stability and ultrastructure of bovine cells from amniotic fluid and umbilical cord. Reprod Domest Anim **49**, 806-812.

De Schauwer C, Meyer E, Cornillie P, De Vliegher S, van de Walle GR, Hoogewijs M, Declercq H, Govaere J, Demeyere K, Cornelissen M, Van Soom A, 2011: Optimization of the isolation, culture, and characterization of equine umbilical cord blood mesenchymal stromal cells. Tissue Eng Part C Methods **17**, 1061-1070.

De Schauwer C, Piepers S, Van de Walle GR, Demeyere K, Hoogewijs MK, Govaere JL, Braeckmans K, Van Soom A, Meyer E, 2012: In search for cross-reactivity to immunophenotype equine mesenchymal stromal cells by multicolor flow cytometry. Cytometry A **81**, 312-23.

De Schauwer C, Van de Walle, GR, Piepers S, Hoogewijs MK, Govaere JL, Meyer E, Van Soom A, 2013: Successful isolation of equine mesenchymal stromal cells from cryopreserved umbilical cord blood-derived mononuclear cell fractions. Equine Vet J **45**, 518-522.

De Schauwer C, Goossens K, Piepers S, Hoogewijs MK, Govaere JL, Smits K, Meyer E, Van Soom A, Van de Walle GR, 2014: Characterization and profiling of immunomodulatory genes of equine mesenchymal stromal cells from non-invasive sources. Stem Cell Res Ther **5**, 6.

Dev K, Gautam SK, Giri SK, Kumar A, Yadav A, Verma V, Kumar P, Singh, B, 2012a: Isolation, culturing and characterization of feeder-independent amniotic fluid stem cells in buffalo (Bubalus bubalis). Res Vet Sci **93**, 743-748.

Dev K, Giri SK, Kumar A, Yadav A, Singh B, Gautam SK, 2012b: Derivation, characterization and differentiation of buffalo (Bubalus bubalis) amniotic fluid derived stem cells. Reprod Domest Anim **47**, 704-711.

Dev K, Giri SK, Kumar A, Yadav A, Singh B, Gautam SK, 2012c. Expression of transcriptional factor genes (Oct-4, Nanog, and Sox-2) and embryonic stem cell-like characters in placental membrane of Buffalo (Bubalus bubalis). J Membr Biol **245**, 177-183.

Di Tomo P, Pipino C, Lanuti P, Morabito C, Pierdomenico L, Sirolli V, Bonomini M, Miscia S, Mariggiò MA, Marchisio M, Barboni B, Pandolfi A, 2013: Calcium Sensing Receptor Expression in Ovine Amniotic Fluid Mesenchymal Stem Cells and the Potential Role of R-568 during Osteogenic Differentiation. PLoS One, **8**, e73816.

Doarn R. Umbilical blood flow and the effects of premature severance in the neonatal horse. Theriogenology 1985, **78**, 789-800.

Dominici M, Le Blanc K, Mueller I, Slaper-Cortenbach I, Marini F, Krause D, Deans R, Keating A, Prockop Dj, Horwitz E, 2006: Minimal criteria for defining multipotent mesenchymal stromal cells. The International Society for Cellular Therapy position statement. Cytotherapy **8**, 315-317.

Erices A, Conget P, Minguell JJ, 2000: Mesenchymal progenitor cells in human umbilical cord blood. Br J Haematol **109**, 235-242.

Fadel L, Viana BR, Feitosa ML, Ercolin AC, Roballo KC, Casals JB, Pieri NC, Meirelles FV, Martins Ddos S, Miglino MA, Ambrósio CE, 2011: Protocols for obtainment and isolation of two mesenchymal stem cell sources in sheep. Acta Cir Bras **26**, 267-273.

Fernandes RA, Wenceslau CV, Reginato AL, Kerkis I, Miglino MA, 2012: Derivation and characterization of progenitor stem cells from canine allantois and amniotic fluids at the third trimester of gestation. Placenta **33**, 640-644.

Figueroa RJ, Koch TG, Betts DH, 2011: Osteogenic differentiation of equine cord blood multipotent mesenchymal stromal cells within coralline hydroxyapatite scaffolds in vitro. Vet Comp Orthop Traumatol **24**, 354-362.

Filioli Uranio M, Valentini L, Lange Consiglio A, Caira M, Guaricci AC, L'Abbate A, Catacchio CR, Ventura M, Cremonesi F, Dell'Aquila ME, 2011: Isolation, proliferation, cytogenetic, and molecular characterization and in vitro differentiation potency of canine stem cells from foetal adnexa: a comparative study of amniotic fluid, amnion, and umbilical cord matrix. Mol Reprod Dev **78**, 361-373.

Filioli Uranio M, Dell'Aquila ME, Caira M, Guaricci AC, Ventura M, Catacchio CR, Martino NA, Valentini L, 2014: Characterization and in vitro differentiation potency of early-passage canine amnion- and umbilical cord-derived mesenchymal stem cells as related to gestational age. Mol Reprod Dev **81**, 539-551.

Fuchs JR, Kaviani A, Oh JT, LaVan D, Udagawa T, Jennings RW, Wilson JM, Fauza DO, 2004: Diaphragmatic reconstruction with autologous tendon engineered from mesenchymal amniocytes. J Pediatr Surg **39**, 834-838.

Fuchs JR, Hannouche D, Terada S, Zand S, Vacanti JP, Fauza DO, 2005: Cartilage engineering from ovine umbilical cord blood mesenchymal progenitor cells. Stem Cells **23**, 958-964.

Gao Y, Zhu Z, Zhao Y, Hua J, Ma Y, Guan W, 2014: Multilineage potential research of bovine amniotic fluid mesenchymal stem cells. Int J Mol Sci **15**, 3698-3710.

Goldschlager T, Ghosh P, Zannettino A, Williamson M, Rosenfeld JV, Itescu S, Jenkin G, 2011: A comparison of mesenchymal precursor cells and amnion epithelial cells for enhancing cervical interbody fusion in an ovine model. Neurosurgery **68**, 1025-1034.

Gray FL, Turner CG, Ahmed A, Calvert CE, Zurakowski D, Fauza DO, 2012: Prenatal tracheal reconstruction with a hybrid amniotic mesenchymal stem cells-engineered construct derived from decellularized airway. J Pediatr Surg **47**, 1072-1079.

Guest DJ, Ousey JC, Smith MR, 2008: Defining the expression of marker genes in equine mesenchymal stromal cells. Stem Cells Cloning, **1**, 1-9.

Gulati BR, Kumar R, Mohanty N, Kumar P, Somasundaram RK, Yadav PS, 2013: Bone morphogenetic protein-12 induces tenogenic differentiation of mesenchymal stem cells derived from equine amniotic fluid. Cells Tissues Organs **198**, 377-389.

Hoynowski SM, Fry MM, Gardner BM, Leming MT, Tucker JR, Black L, Sand T, Mitchell KE, 2007: Characterization and differentiation of equine umbilical cord-derived matrix cells. Biochem Biophys Res Commun **362**, 347-353.

Iacono E, Brunori L, Pirrone A, Pagliaro PP, Ricci F, Tazzari PL, Merlo B, 2012a: Isolation, characterization and differentiation of mesenchymal stem cells from amniotic fluid, umbilical cord blood and Wharton's jelly in the horse. Reproduction **143**, 455-468.

Iacono E, Cunto M, Zambelli D, Ricci F, Tazzari PL, Merlo B, 2012b: Could fetal fluid and membranes be an alternative source for mesenchymal stem cells (MSCs) in the feline species? A preliminary study. Vet Res Commun **36**, 107-118.

Iacono E, Merlo B, Pirrone A, Antonelli C, Brunori L, Romagnoli N, Castagnetti C, 2012c: Effects of mesenchymal stem cells isolated from amniotic fluid and platelet-rich plasma gel on severe decubitus ulcers in a septic neonatal foal. Res Vet Sci **93**, 1439-1440.

Jager M, Bachmann R, Scharfstadt A, Krauspe R, 2006: Ovine cord blood accommodates multipotent mesenchymal progenitor cells. In Vivo **20**, 205-214.

Jang BJ, Byeon YE, Lim JH, Ryu HH, Kim WH, Koyama Y, Kikuchi M, Kang KS, Kweon OK, 2008: Implantation of canine umbilical cord blood-derived mesenchymal stem cells mixed with beta-tricalcium phosphate enhances osteogenesis in bone defect model dogs. J Vet Sci **9**, 387-393.

493 Jin GZ, Yin XJ, Yu XF, Cho SJ, Choi EG, Lee YS, Jeon JT, Yee ST, Kong IK, 2008: Generation of
494 neuronal-like cells from umbilical cord blood-derived mesenchymal stem cells of a RFP-transgenic
495 cloned cat. J Vet Med Sci **70**, 723-726.
496 Kang BJ, Ryu HH, Park SS, Koyama Y, Kikuchi M, Woo HM, Kim WH, Kweon OK, 2012:
497 Comparing the osteogenic potential of canine mesenchymal stem cells derived from adipose tissues,
498 bone marrow, umbilical cord blood, and Wharton's jelly for treating bone defects. J Vet Sci **13**, 299-
499 310.
500 Kang JG, Park SB, Seo MS, Kim HS, Chae JS, Kang KS, 2013: Characterization and clinical
501 application of mesenchymal stem cells from equine umbilical cord blood. J Vet Sci **14**, 367-371.
502 Kaviani A, Perry TE, Dzakovic A, Jennings RW, Ziegler MM, Fauza DO, 2001: The amniotic fluid
503 as a source of cells for fetal tissue engineering. J Pediatr Surg **36**, 1662-1665.
504 Kim EY, Lee KB, Yu J, Lee JH, Kim KJ, Han KW, Park KS, Lee DS, Kim MK, 2014: Neuronal
505 cell differentiation of mesenchymal stem cells originating from canine amniotic fluid. Hum Cell **27**,
506 51-58.
507 Kimura M, Toyoda M, Gojo S, Itakura Y, Kami D, Miyoshi S, Kyo S, Ono M, Umezawa A, 2012:
508 Allogeneic amniotic membrane-derived mesenchymal stromal cell transplantation in a porcine
509 model of chronic myocardial ischemia. J Stem Cells Regen Med **8**:171-180.
510 Klemmt PA, Vafaizadeh V, Groner B, 2011: The potential of amniotic fluid stem cells for cellular
511 therapy and tissue engineering. Expert Opin Biol Ther **11**, 1297-1314.
512 Koch TG, Heerkens T, Thomsen PD, Betts DH, 2007: Isolation of mesenchymal stem cells from
513 equine umbilical cord blood. BMC Biotechnol **30**, 7-26.
514 Kumar BM, Yoo JG, Ock SA, Kim JG, Song HJ, Kang EJ, Cho SK, Lee SL, Cho JH,
515 Balasubramanian S, Rho GJ, 2007: In vitro differentiation of mesenchymal progenitor cells derived
516 from porcine umbilical cord blood. Mol Cells **24**, 343-350.

517 Kunisaki SM, Jennings RW, Fauza DO, 2006a: Fetal cartilage engineering from amniotic
518 mesenchymal progenitor cells. Stem Cells Dev **15**, 245-253.
519 Kunisaki SM, Freedman DA, Fauza DO, 2006b: Fetal tracheal reconstruction with cartilaginous
520 grafts engineered from mesenchymal amniocytes. J Pediatr Surg **41**, 675-682.
521 Kunisaki SM, Fuchs JR, Kaviani A, Oh JT, LaVan DA, Vacanti JP, Wilson JM, Fauza DO, 2006c:
522 Diaphragmatic repair through fetal tissue engineering: a comparison between mesenchymal
523 amniocyte- and myoblast-based constructs. J Pediatr Surg **41**, 34-39.
524 Kunisaki SM, Fuchs JR, Steigman SA, Fauza DO, 2007: A comparative analysis of cartilage
525 engineered from different perinatal mesenchymal progenitor cells. Tissue Eng **13**, 2633-2644.
526 Lange-consiglio A, Corradetti B, Rutigliano L, Cremonesi F, Bizzaro D, 2011: *In vitro* studies of
527 horse umbilical cord matrix-derived cells: from characterization to labeling for magnetic resonance
528 imaging. Open Tissue Eng Regen Med J **4,** 120-133.
529 Lange-Consiglio A, Corradetti B, Bizzaro D, Magatti M, Ressel L, Tassan S, Parolini O, Cremonesi
530 F, 2012: Characterization and potential applications of progenitor-like cells isolated from horse
531 amniotic membrane. J Tissue Eng Regen Med **6**, 622-635.
532 Lange-Consiglio A, Corradetti B, Meucci A, Perego R, Bizzaro D, Cremonesi F, 2013a:
533 Characteristics of equine mesenchymal stem cells derived from amnion and bone marrow: in vitro
534 proliferative and multilineage potential assessment. Equine Vet J **45**, 737-744.
535 Lange-Consiglio A, Rossi D, Tassan S, Perego R, Cremonesi F, Parolini O, 2013b: Conditioned
536 medium from horse amniotic membrane-derived multipotent progenitor cells: immunomodulatory
537 activity in vitro and first clinical application in tendon and ligament injuries in vivo. Stem Cells Dev
538 **22**, 3015-3024.
539 Lange-Consiglio A, Tassan S, Corradetti B, Meucci A, Perego R, Bizzaro D, Cremonesi F, 2013c:
540 Investigating the efficacy of amnion-derived compared with bone marrow-derived mesenchymal
541 stromal cells in equine tendon and ligament injuries. Cytotherapy **15**, 1011-1020.

Lee KS, Cha SH, Kang HW, Song JY, Lee KW, Ko KB, Lee HT, 2013a: Effects of serial passage on the characteristics and chondrogenic differentiation of canine umbilical cord matrix derived mesenchymal stem cells. Asian-Australas J Anim Sci **26**, 588-595.

Lee KS, Nah JJ, Lee BC, Lee HT, Lee HS, So BJ, Cha SH, 2013b: Maintenance and characterization of multipotent mesenchymal stem cells isolated from canine umbilical cord matrix by collagenase digestion. Res Vet Sci **94**, 144-151.

Lim JH, Byeon YE, Ryu HH, Jeong YH, Lee YW, Kim WH, Kang KS, Kweon OK, 2007: Transplantation of canine umbilical cord blood-derived mesenchymal stem cells in experimentally induced spinal cord injured dogs. J Vet Sci **8**, 275-282.

Lin CS, Ning H, Lin G, Lue TF, 2012: Is CD34 truly a negative marker for mesenchymal stromal cells? Cytotherapy **14**, 1159-1163.

Lindenmair A, Hatlapatka T, Kollwig G, Hennerbichler S, Gabriel C, Wolbank S, Redl H, Kasper C, 2012: Mesenchymal stem or stromal cells from amnion and umbilical cord tissue and their potential for clinical applications. Cells **1**, 1061-1088.

Lovati AB, Corradetti B, Lange-Consiglio A, Recordati C, Bonacina E, Bizzaro D, Cremonesi F, 2011: Comparison of equine bone marrow-, umbilical cord matrix and amniotic fluid-derived progenitor cells. Vet Res Commun **35**, 103-121.

Luo H, Lu Y, Wu T, Zhang M, Zhang Y, Jin Y, 2013: Construction of tissue-engineered cornea composed of amniotic epithelial cells and acellular porcine cornea for treating corneal alkali burn. Biomaterials **34**, 6748-6759.

Mann A, Yadav RP, Singh J, Kumar D, Singh B, Yadav PS, 2013: Culture, characterization and differentiation of cells from buffalo (Bubalus bubalis) amnion. Cytotechnology **65**, 23-30.

Martino NA, Lange-Consiglio A, Cremonesi F, Valentini L, Caira M, Guaricci AC, Ambruosi B, Sciorsci RL, Lacalandra GM, Reshkin SJ, Dell'Aquila ME, 2012: Functional expression of the

566 extracellular calcium sensing receptor (CaSR) in equine umbilical cord matrix size-sieved stem
567 cells. PLoS One **6**, e17714.
568 Mattioli M, Gloria A, Turriani M, Mauro A, Curini V, Russo V, Tetè S, Marchisio M,
569 Pierdomenico L, Berardinelli P, Colosimo A, Muttini A, Valbonetti L, Barboni B, 2012: Stemness
570 characteristics and osteogenic potential of sheep amniotic epithelial cells. Cell Biol Int **36**, 7-19.
571 Mauro A, Turriani M, Ioannoni A, Russo V, Martelli A, Di Giacinto O, Nardinocchi D, Berardinelli
572 P, 2010: Isolation, characterization, and in vitro differentiation of ovine amniotic stem cells. Vet
573 Res Commun **34**, S25-28.
574 McGeady TA, Quinn PJ, FitzPatrick ES, Ryan MT, 2006: Foetal Membranes. In: Veterinary
575 Embryology. Blackwell Publishing, pp. 66-75.
576 Miller D, Packthongsuk K, Rathbun T, Boyle D, Troyer D, Davis DL, 2012: Confocal imaging of
577 trans-epithelial trafficking by immune and umbilical cord stem cells in the neonatal porcine
578 intestine. Anat Histol Embryol **41**, 461-468.
579 Mitchell KE, Weiss ML, Mitchell BM, Martin, P, Davis D, Morales L, Helwig B, Beerenstrauch M,
580 Abou-Easa K, Hildreth T, Troyer D, Medicetty S, 2003: Matrix cells from Wharton's jelly form
581 neurons and glia. Stem Cells **21**, 50-60.
582 Mohanty N, Gulati BR, Kumar R, Gera S, Kumar P, Somasundaram RK, Kumar S, 2014:
583 Immunophenotypic characterization and tenogenic differentiation of mesenchymal stromal cells
584 isolated from equine umbilical cord blood. In Vitro Cell Dev Biol Anim **50**, 538-548.
585 Nanaev AK, Kohnen G, Milovanov AP, Domogatsky SP, Kaufmann P, 1997: Stromal
586 differentiation and architecture of the human umbilical cord. Placenta **18**, 53-64.
587 Paebst F, Piehler D, Brehm W, Heller S, Schroeck C, Tarnok A, Burk J, 2014: Comparative
588 immunophenotyping of equine multipotent mesenchymal stromal cells: an approach toward a
589 standardized definition. Cytometry A **85**, 678-687.

590 Park SS, Byeon YE, Ryu HH, Kang BJ, Kim Y, Kim WH, Kang KS, Han HJ, Kweon OK, 2011:
591 Comparison of canine umbilical cord blood-derived mesenchymal stem cell transplantation times:
592 involvement of astrogliosis, inflammation, intracellular actin cytoskeleton pathways, and
593 neurotrophin-3. Cell Transplant **20**, 1867-1880.
594 Park SB, Seo MS, Kim HS, Kang KS, 2012: Isolation and characterization of canine amniotic
595 membrane-derived multipotent stem cells. PLoS One **7**, e44693.
596 Passeri S, Nocchi F, Lamanna R, Lapi S, Miragliotta V, Giannessi E, Abramo F, Stornelli MR,
597 Matarazzo M, Plenteda D, Urciuoli P, Scatena F, Coli A, 2009: Isolation and expansion of equine
598 umbilical cord-derived matrix cells (EUCMCs). Cell Biol Int **33**, 100-105.
599 Peng SY, Chou CJ, Cheng PJ, Tseng TY, Cheng WT, Shaw SW, Wu SC, 2014:
600 Intramuscular transplantation of pig amniotic fluid derived progenitor cells has therapeutic potential
601 in a mouse model of myocardial infarction. Cell Transplant, doi: 10.3727/096368914X680109.
602 Pratheesh MD, Gade NE, Katiyar AN, Dubey PK, Sharma B, Saikumar G, Amarpal, Sharma GT,
603 2013: Isolation, culture and characterization of caprine mesenchymal stem cells derived from
604 amniotic fluid. Res Vet Sci **94**, 313-319.
605 Pratheesh MD, Gade NE, Dubey PK, Nath A, Sivanarayanan TB, Madhu DN, Sharma B, Amarpal,
606 Saikumar G, Sharma GT, 2014: Molecular characterization and xenogenic application of Wharton's
607 jelly derived caprine mesenchymal stem cells. Vet Res Commun **38**, 139-148.
608 Raoufi MF, Tajik P, Dehghan MM, Eini F, Barin A, 2011: Isolation and differentiation of
609 mesenchymal stem cells from bovine umbilical cord blood. Reprod Domest Anim **46**, 95-99.
610 Reed SA, Johnson SE, 2008: Equine umbilical cord blood contains a population of stem cells that
611 express Oct4 and differentiate into mesodermal and endodermal cell types. J Cell Physiol **215**, 329-
612 336.
613 Rossdale PD. Parturition in the thoroughbred mare with particular reference to blood deprivation in
614 the new-born. Vet Record 1958: **70**, 142–151.

615 Rossi B, Merlo B, Colleoni S, Iacono E, Tazzari PL, Ricci F, Lazzari G, Galli C, 2014: Isolation
616 and in vitro characterization of bovine amniotic fluid derived stem cells at different trimesters of
617 pregnancy. Stem Cell Rev **10**, 712-724.
618 Rutigliano L, Corradetti B, Valentini L, Bizzaro D, Meucci A, Cremonesi F, Lange-Consiglio A,
619 2013: Molecular characterization and in vitro differentiation of feline progenitor-like amniotic
620 epithelial cells. Stem Cell Res Ther **4**, 133.
621 Ryu HH, Kang BJ, Park SS, Kim Y, Sung GJ, Woo HM, Kim WH, Kweon OK, 2012: Comparison
622 of mesenchymal stem cells derived from fat, bone marrow, Wharton's jelly, and umbilical cord
623 blood for treating spinal cord injuries in dogs. J Vet Med Sci **74**, 1617-1630.
624 Sartore S, Lenzi M, Angelini A, Chiavegato A, Gasparotto L, De Coppi P, Bianco R, Gerosa G,
625 2005: Amniotic mesenchymal cells autotransplanted in a porcine model of cardiac ischemia do not
626 differentiate to cardiogenic phenotypes. Eur J Cardiothorac Surg **28**, 677-684.
627 Schuh EM, Friedman MS, Carrade DD, Li J, Heeke D, Oyserman SM, Galuppo LD, Lara DJ,
628 Walker NJ, Ferraro GL, Owens SD, Borjesson DL, 2009: Identification of variables that optimize
629 isolation and culture of multipotent mesenchymal stem cells from equine umbilical-cord blood. Am
630 J Vet Res **70**, 1526-1535.
631 Seo MS, Jeong YH, Park JR, Park SB, Rho KH, Kim HS, Yu KR, Lee SH, Jung JW, Lee YS, Kang
632 KS, 2009: Isolation and characterization of canine umbilical cord blood-derived mesenchymal stem
633 cells. J Vet Sci **10**, 181-187.
634 Seo MS, Park SB, Kang KS, 2012: Isolation and characterization of canine Wharton's jelly-derived
635 mesenchymal stem cells. Cell Transplant **21**, 1493-1502.
636 Seo MS, Park SB, Kim HS, Kang JG, Chae JS, Kang KS, 2013: Isolation and characterization of
637 equine amniotic membrane-derived mesenchymal stem cells. J Vet Sci **14**, 151-159.

638 Shaw SW, Bollini S, Nader KA, Gastadello A, Mehta V, Filppi E, Cananzi M, Gaspar HB, Qasim
639 W, De Coppi P, David AL, 2011: Autologous transplantation of amniotic fluid-derived
640 mesenchymal stem cells into sheep fetuses. Cell Transplant **20**, 1015-1031.
641 Singh J, Mann A, Kumar D, Duhan JS, Yadav PS, 2013: Cultured buffalo umbilical cord matrix
642 cells exhibit characteristics of multipotent mesenchymal stem cells. In Vitro Cell Dev Biol Anim
643 **49**, 408-416.
644 Sobolewski K, Bańkowski E, Chyczewski L, Jaworski S, 1997: Collagen and glycosaminoglycans
645 of Wharton's jelly. Biol Neonate **71**, 11-21.
646 Sreekumar TR, Ansari MM, Chandra V, Sharma GT, 2014: Isolation and characterization of buffalo
647 Wharton's Jelly derived mesenchymal stem cells. J Stem Cell Res Ther **4**, doi:10.4172/2157-
648 7633.1000207.
649 Takechi K1, Kuwabara Y, Mizuno M, 1993: Ultrastructural and immunohistochemical studies of
650 Wharton's jelly umbilical cord cells. Placenta **14**, 235-245.
651 Toupadakis CA, Wong A, Genetos DC, Cheung WK, Borjesson DL, Ferraro GL, Galuppo LD,
652 Leach JK, Owens SD, Yellowley CE, 2010: Comparison of the osteogenic potential of equine
653 mesenchymal stem cells from bone marrow, adipose tissue, umbilical cord blood, and umbilical
654 cord tissue. Am J Vet Res **71**, 1237-1245.
655 Turner CG, Klein JD, Steigman SA, Armant M, Nicksa GA, Zurakowski D, Ritz J, Fauza, DO,
656 2011: Preclinical regulatory validation of an engineered diaphragmatic tendon made with amniotic
657 mesenchymal stem cells. J Pediatr Surg **46**, 57-61.
658 Van Loon VJ, Scheffer CJ, Genn HJ, Hoogendoorn AC, Greve JW, 2014: Clinical follow-up of
659 horses treated with allogeneic equine mesenchymal stem cells derived from umbilical cord blood
660 for different tendon and ligament disorders. Vet Q, **34**, 92-97.

Vidane AS, Souza AF, Sampaio RV, Bressan FF, Pieri NC, Martins DS, Meirelles FV, Miglino MA, Ambrósio CE, 2014: Cat amniotic membrane multipotent cells are nontumorigenic and are safe for use in cell transplantation. Stem Cells Cloning **7**, 71-78.

Violini S, Gorni C, Pisani LF, Ramelli P, Caniatti M, Mariani P, 2012: Isolation and differentiation potential of an equine amnion-derived stromal cell line. Cytotechnology **64**, 1-7.

Weber B, Emmert MY, Behr L, Schoenauer R, Brokopp C, Drogemuller C, Modregger P, Stampanoni M, Vats D, Rudin M, Bürzle W, Farine M, Mazza E, Frauenfelder T, Zannettino AC, Zünd G, Kretschmar O, Falk V, Hoerstrup SP, 2012: Prenatally engineered autologous amniotic fluid stem cell-based heart valves in the fetal circulation. Biomaterials **33**, 4031-4043.

Weber B, Kehl D, Bleul U, Behr L, Sammut S, Frese L, Ksiazek A, Achermann J, Stranzinger G, Robert J, Sanders B, Sidler M, Brokopp CE, Proulx ST, Frauenfelder T, Schoenauer R, Emmert MY, Falk V, Hoerstrup SP, 2013: In vitro fabrication of autologous living tissue-engineered vascular grafts based on prenatally harvested ovine amniotic fluid-derived stem cells. J Tissue Eng Regen Med, doi: 10.1002/term.1781.

Weiss ML, Mitchell KE, Hix JE, Medicetty S, El-Zarkouny SZ, Grieger D, Troyer DL, 2003: Transplantation of porcine umbilical cord matrix cells into the rat brain. Exp Neurol **182**, 288-299.

Yadav PS, Mann A, Singh V, Yashveer S, Sharma RK, Singh I, 2011: Expression of pluripotency genes in buffalo (Bubalus bubalis) amniotic fluid cells. Reprod Domest Anim **46**, 705-711.

Zheng YM, Zhao HY, Zhao XE, Quan FS, Hua S, He XY, Liu J, He XN, Lin H, 2009: Development of cloned embryos from porcine neural stem cells and amniotic fluid-derived stem cells transfected with enhanced green fluorescence protein gene. Reproduction **137**, 793-801.

Zheng YM, Zhao XE, An ZX, 2010: Neurogenic differentiation of EGFP gene transfected amniotic fluid-derived stem cells from pigs at intermediate and late gestational ages. Reprod Domest Anim **45**, e78-82.

689 **Table 1**. AMSCs characterization.

Species	Phenotype	*In vitro* differentiation	Ref.
Sheep	**ICC**: Oct4+,TERT+	Osteogenic, myogenic	(Mauro et al. 2010)
Horse	**FCA**: CD44+,CD90+,CD14+,CD45+,CD29-,MHCI-,MHCII- **ICC**: cKit+,Oct4+,CD105+	Adipogenic, chondrogenic, osteogenic	(Coli et al. 2011)
	RT-PCR: OCT4+,SOX2+,NANOG+,CD105+,CD34-	Adipogenic, chondrogenic, osteogenic	(Violini et al. 2012)
	ICC: Tra1-60+,SSEA3+,SSEA4+,Oct4+ **RT-PCR**: CD29+,CD105+,CD44+,CD166+,CD34-, MHCI+,MHCII-	Adipogenic, chondrogenic, osteogenic, neurogenic	(Lange-Consiglio et al. 2012)
	FCA: CD44+,CD90+,CD105+,CD19-,CD20-,CD28-,CD31-, CD34-,CD38-,CD41a-,CD62L-,CD62P-,CD200-	Adipogenic, chondrogenic, osteogenic	(Seo et al. 2013)
	AP+ **ICC**: Tra1-60+, SSEA4+,Oct4+ **RT-PCR**: CD29+,CD105+,CD44+,CD166+,CD34-, MHCI+,MHCII-	Adipogenic, chondrogenic, osteogenic, neurogenic	(Lange-Consiglio et al. 2013a)
	FCA: Oct4+,SSEA4+,cMyc+,CD34- **RT-PCR:** CD29+,CD44+, CD166+, CD105+, CD34- MHCI+,MHCII-		(Corradetti et al. 2014)
Dog	**RT-PCR**: OCT4+,NANOG-,CD44+,CD184+,CD29+,CD34-,CD45-, DLA79-,DLA88-,DLA-DRA1-,DLA-DQA1-	Adipogenic, chondrogenic, osteogenic, neurogenic	(Filioli Uranio et al. 2011)
	FCA: CD90+,CD105+,CD3-,CD11c-,CD28-,CD34-,CD38-, CD41a-,CD45-,CD62L- **qPCR**: OCT4+,SOX2+,NANOG+,KLF4+	Adipogenic, chondrogenic, osteogenic, neurogenic	(Park et al. 2012)

	RT-PCR: OCT4+,NANOG-,CD29+,CD44+,CD184+,CD34-, CD45-,DLA79-,DLA88-,DLA-DRA1-,DLA-DQA1-	Osteogenic, neurogenic	(Filioli Uranio et al. 2014)
Cat	**FCA**: CD44+,CD90+,CD105+,CD14-,CD34-,CD45-,CD73-	Adipogenic, chondrogenic, osteogenic	(Iacono et al. 2012b)
	FCA: CD73+,CD90+,CD34±,CD45-,CD79-	Adipogenic, chondrogenic, osteogenic	(Vidane et al. 2014)
Buffalo	**AP**+ **RT-PCR**: OCT4+,SOX2+,NANOG+,AP+,SCF+,NESTIN+		(Dev et al. 2012c)
	AP+ **RT-PCR**: OCT4+,SOX2+,NANOG+	Osteogenic	(Mann et al. 2013)
Bovine	**RT-PCR**: OCT4+,Myc+,CD44+,CD166+,CD73+,CD29+, CD105-,CD45-,CD14-,CD34-,MHCI+,MHCII-	Adipogenic, chondrogenic, neurogenic	(Corradetti et al. 2013)

690 +: positive, -: negative; ±: weakly positive; ICC: immunocytochemistry.

691 **Table 2**. AFMSCs characterization.

Species	Phenotype	*In vitro* differentiation	Ref.
Sheep	**ICC:** FSP+,CD31-,Vimentin+,αSMA+,Desmin-,CK8+,CK18+		(Kaviani et al. 2001)
	ICC: Vimentin+,CK8+,CK18+,CD31-	Chondrogenic	(Kunisaki et al. 2006a)
		Chondrogenic	(Kunisaki et al. 2006b)
	FCA: CD29+,CD44+,CD90±,CD105±,CD31-	Adipogenic, osteogenic	(Kunisaki et al. 2007)
	ICC: Oct4+,TERT+	Osteogenic, myogenic	(Mauro et al. 2010)
	FCA: CD44+,CD58+,CD166+,CD14-,CD31-,CD45-	Adipogenic, osteogenic	(Shaw et al. 2011)
	FCA: CD29+,CD44+,CD105+,CD166+,CD31-		(Gray et al. 2012)
	ICC: SSEA4+,STRO4+,CD29+,CD44+,CD166+ **RT-PCR**: OCT4+,NANOG+,SOX2+		(Weber et al. 2012)
	ICC: TERT+,Sox2+,Nanog+ **semiqPCR**: TERT+,NANOG+,SOX2±,OCT4±	Osteogenic	(Colosimo et al. 2013a)
	FCA: Sox2+,Nanog+,TERT+,CD166+,CD29±,CD58±,CD14-,CD31-,CD45-,c-Kit- **ICC**: Sox2+,Nanog+,TERT+,c-Kit-	Adipogenic, osteogenic, tenogenic	(Colosimo et al. 2013b)
	FCA: TERT+,Oct3/4+,Nanog+,Sox2+,CD29+,CD58+,CD90+,CD166±,CD14-,CD31-,CD45-,CD49f-,CD117-,HLA-ABC+, HLA-DR-	Osteogenic	(Di Tomo et al. 2013)
	FCA: CD44+,CD166+,CD29+,STRO4+ **ICC**: CD29+,CD44+,STRO4+,CD166±,CD11b-	Chondrogenic, osteogenic, endothelial, myogenic/myofibroblasts	(Weber et al. 2013)

	,Vimentin+,αSMA±,Desmin± **qPCR**: NANOG+,STAT3+		
Swine	**ICC**: SSEA4±,Oct4±,CD90+,Vimentin+,vWf+,SM22+,VE-Cadherin±,αSMA±,CD31±,CD105-,CD117-,CD34-,CD45-,Sca-1-,CardiacTnI-		(Sartore et al. 2005)
	RT-PCR: OCT4+,SOX2+,THY+	Adipogenic, osteogenic, neurogenic, myogenic, endothelial	(Zheng et al. 2009)
	RT-PCR: OCT4+,SOX2+,THY+	Neurogenic	(Zheng et al. 2010)
	FCA: CD44+,CD166+,CD34-,CD45-,CD54- **ICC**: Oct4+,Nanog+,Tra1-60+,Tra1-81+,SSEA4+,SSEA1- **RT-PCR**: CD90+,CD117+,HLA-ABC+,CD45-	Adipogenic, neurogenic, cardiomyogenic; Multilineage-differentiation through EBs	(Chen et al. 2011)
	FCA: CD44+,CD90+,CD31-,CD4a-		(Peng et al. 2014)
Horse	**ICC**: Oct4+,SSEA4+,Tra1-60± **RT-PCR**: CD29+,CD44+,CD105+,CD34-,ELA-ABC+,ELA-DR-	Adipogenic, chondrogenic, osteogenic	(Lovati et al. 2011)
	FCA: CD44+,CD90+,CD105+,CD14-,CD34-,CD45-,CD73-	Adipogenic, chondrogenic, osteogenic	(Iacono et al. 2012a)
		Adipogenic, chondrogenic, osteogenic	(Iacono et al. 2012c)
	AP+ **FCA**: CD29+,CD44+,CD90+,CD73(clone 5F/B9)-,CD73(clone AD2)+,CD34-,CD45- **ICC**: CD73+,CD90+,CD34-,CD45- **RT-PCR**: CD73+,CD90+,CD105+,CD14-,CD34-,CD45-	Adipogenic, chondrogenic, osteogenic, tenogenic	(Gulati et al. 2013)

Dog	**RT-PCR**: OCT4+,NANOG-,CD44+,CD184-,CD29-,CD34-,CD45-,DLA79+,DLA-DRA1+,DLA-DQA1-,DLA 88-	Adipogenic, chondrogenic, osteogenic, neurogenic	(Filioli Uranio et al. 2011)
	ICC: Vimentin+, Nestin+, CK18+	Adipogenic, chondrogenic, osteogenic, neurogenic	(Fernandes et al. 2012)
	FCA: CD29+,CD44+,CD90+,CD34- **ICC**: Oct4+,Nanog+,Sox2+,CD29+,CD44+,CD90+,CD34- **RT-PCR**: OCT4+,NANOG+,SOX2+	Adipogenic, chondrogenic, osteogenic, hepatogenic	(Choi et al. 2013)
		Neurogenic	(Kim et al. 2014)
Cat	**FCA**: CD44+,CD90+,CD105+,CD34±,CD14-,CD45-,CD73-	Adipogenic, chondrogenic, osteogenic	(Iacono et al. 2012b)
Buffalo	**AP**+ **RT-PCR**: OCT4+,NESTIN+,FGF5+		(Dev et al. 2012a)
	AP+ **RT-PCR**:OCT4+,NANOG+,SOX2+,AP+,SCF+,CyclinA+,NESTIN+,FGF5+	Neurogenic	(Dev et al. 2012b)
	AP+ **qPCR**: OCT4+,NANOG+,SOX2+		(Yadav et al. 2012)
Caprine	**FCA**: Nanog+,Sox2+ **ICC**: Oct4+,Nanog+,Sox2+,SSEA4+,SSEA1+ **RT-PCR**: CD73+,CD90+,CD105+,CD34-	Adipogenic, chondrogenic, osteogenic	(Pratheesh et al. 2013)

Bovine	**RT-PCR**: OCT4+,MYC+,CD29+,CD44+,CD73+,CD166+,CD14-,CD34-,CD45-,CD105-,CD117+,MHCI+,MHCII-	Adipogenic, chondrogenic, neurogenic	(Corradetti et al. 2013)
	ICC: Oct4+,CD29+,CD44+,CD71+,CD34- **RT-PCR**: OCT4+,CD29+,CD44+,CD73+,CD166+,CD34-,CD45-	Adipogenic, chondrogenic, neurogenic	(Gao et al. 2014)
	FCA: CD34+,CD44+,CD90+,CD105+,CD14- **ICC**: Oct4-,SSEA4±,αSMA+,Vimentin+,N-Cadherin+,E-Cadherin-,CK- **qPCR:** OCT4-,NANOG-,SOX2-	Adipogenic, chondrogenic, osteogenic	(Rossi et al. 2014)

692 +: positive, -: negative; ±: weakly positive; ICC: immunocytochemistry.

693

694 **Table 3**. UCBMSCs characterization.

Species	Phenotype	*In vitro* differentiation	Ref.
Sheep		Chondrogenic	(Fuchs et al. 2005)
		Adipogenic, chondrogenic, osteogenic	(Jager et al. 2006)
	FCA: CD44+,CD38-,CD45-,CD41/61-		(Fadel et al. 2011)
Horse		Adipogenic, chondrogenic, osteogenic	(Koch et al. 2007)
	ICC: Oct4-,SSEA1-,SSEA3-,SSEA4-,Tra1-60-,Tra1-81-,AP+,CD29+,CD44+,CD90+,CD14-,CD79α-,Coll I+,Coll II+,Coll III+,Coll IX/XI+,MHCI±,MHCII-	Adipogenic, chondrogenic, Osteogenic	(Guest et al. 2008)
	AP + **ICC**: Oct4+,Tra1-60+,Tra1-81+,SSEA1+,SSEA4±	Adipogenic, chondrogenic, hepatogenic, myogenic	(Reed and Johnson, 2008)
		Chondrogenic	(Berg et al. 2009)
	ICC: Vimentin±,αSMA±,CD18-,PanCK-,Factor VIII-,Osteonectin-,Osteocalcin-	Adipogenic, chondrogenic, osteogenic	(Schuh et al. 2009)
		Osteogenic	(Toupadakis et al. 2010)
		Osteogenic	(Figueroa et al. 2011)
	FCA: CD86-,MHCI-,MHCII-		(Carrade et al. 2011)
	FCA: CD29+,CD44+,CD90+,CD86-,MHCI+,MHCII-,F6B-		(Carrade et al. 2012)
	FCA: CD29+,CD44+,CD90+,CD79α-,MHCII-	Adipogenic, chondrogenic,	(De Shauwer et al. 2011)

		osteogenic	
	FCA and Confocal Microscopy: CD29+,CD44+,CD90+,MHCII-,CD105±,CD45-,CD73±,CD79α-,monocyte marker-	Adipogenic, chondrogenic, osteogenic	(De Shauwer et al. 2012)
		Adipogenic, chondrogenic, osteogenic	(Burk et al. 2013)
	FCA: CD29+,CD44+,CD73+,CD90+,CD105+,CD45-CD79α-,MHCI+,MHCII-,monocyte marker- **RT-qPCR**: CD40+,CD80±,CD86-,TGFβ+,HGF±,IDO-,TNFα-	Adipogenic, chondrogenic, osteogenic,	(De Shauwer et al. 2013)
	FCA: CD90+,CD105+,CD20-,CD28-,CD38-,CD62L-CD200-,CD31-,CD62P-,CD34-,CD41a-	Adipogenic, chondrogenic, osteogenic	(Kang et al. 2013)
	FCA: CD29+,CD44+,CD73+,CD90+,CD105+,CD45-CD79α-,MHCI+,MHCII-,monocyte marker- **qPCR**: CD40+,CD80+,CD86-,TGFβ+,HGF±,IDO-,TNFα-	Adipogenic, chondrogenic, osteogenic,	(De Schauwer et al. 2014)
	FCA: CD29+,CD44+,CD73+,CD90+,CD34-,CD45- **ICC**: CD73+,CD90+,CD34-,CD45- **RT-PCR**:OCT4+,NANOG+,SOX2+,CD73+,CD90+,CD105+,CD14-,CD34-,CD45-	Tenogenic	(Mohanty et al. 2014)
	FCA: CD29±,CD44+,CD73-,CD90-,CD105-,CD14-CD34-,CD45-,CD79α-,MHCII-		(Paebst et al. 2014)
Dog		Osteogenic	(Jang et al. 2008)
	FCA: Oct4+,CD29+,CD33+,CD44+,CD105+,CD184+,CD4-,CD8a-,CD10-,CD14-,CD24-,CD31-,CD34-,CD38-,CD41a-,CD45-,CD49b-,CD41/61-,CD62p,CD73-,CD90-,CD133-,HLA-	Chondrogenic, osteogenic, neurogenic	(Seo et al. 2009)

		DR-		
		FCA: CD44+,CD73+,CD90+,CD105+,CD14-,CD34-,CD45-	Osteogenic	(Kang et al. 2012)
	Swine	**ICC**: CD29+,CD49b+,CD105+,CD45-,CD133-,SCF+,LIF+,G-CSF+	Adipogenic, chondrogenic, osteogenic	(Kumar et al. 2007)
	Cat	**FCA**: CD9+,CD44+,CD18-,CD45- **ICC**: Vimentin+	Neurogenic	(Jin et al. 2008)
	Bovine	**RT-PCR**: OCT4+,CD73+	Adipogenic, chondrogenic, osteogenic	(Raoufi et al. 2011)

695 + :positive; - : negative; ± : weakly positive; ICC: immunocytochemistry

696 **Table 4**. WJMSCs and UCMMSCs characterization.

Species	Phenotype	*In vitro* differentiation	Ref.
Swine	**Immunoblotting**: c-kit+,αSMA+	Neurogenic	(Mitchell et al. 2003)
	AP+ **ICC:** Oct4+,Nanog+ **RT-PCR**: OCT4+,NANOG+,SOX2+ **qPCR**: OCT4+,NANOG+,SOX2+		(Carlin et al. 2006)
Horse	**FCA**: CD54+,CD90+,CD105+,CD146+,Oct4+,SSEA4+,c-kit+, SSEA3±,Tra1-60±,HLA-ABC-,HLA-1AG-,MHCII-	Adipogenic, chondrogenic, osteogenic	(Hoynowski et al. 2007)
	RT-PCR: Oct4+, Sox2+		(Cremonesi et al. 2008)
		Adipogenic, chondrogenic, osteogenic	(Passeri et al. 2009)
		Osteogenic	(Toupadakis et al. 2010)
	FCA: Oct4+,c-myc+,CD34-,CD105-,SSEA4-,SSEA3-,Tra1-60- **RT-PCR**: CD29+,CD44+,CD105+,CD166+,CD14-CD34-, MHCI+,MHCII-	Adipogenic, chondrogenic, osteogenic, neurogenic	(Corradetti et al. 2011)
	RT-PCR: CD29+,CD44+,CD105+,CD166+,CD14-CD34-, MHCI+,MHCII-	Adipogenic, chondrogenic, osteogenic, neurogenic	(Lange-Consiglio et al. 2011)
	RT-PCR: CD29+,CD44+,CD105+,CD34-,MHCI+,MHCII- **ICC**: Oct4+,SSEA4+,Tra1-60±	Adipogenic, chondrogenic, osteogenic	(Lovati et al. 2011)
	RT-PCR: CaSR+		(Martino et al. 2011)
	FCA: CD86-,MHCI-,MHCII-		(Carrade et al. 2011)

	FCA: CD29+,CD44+,CD90+,CD86-,MHCI+,MHCII-,F6B-		(Carrade et al. 2012)
		Adipogenic, chondrogenic, osteogenic	(Burk et al. 2013)
	FCA: CD29+,CD44+,CD90+,CD45-,CD73-,CD79α-,CD105-, MCHII-,Monocyte marker-		(De Schauwer et al. 2013)
	FCA: CD44+,CD90+,CD105+,CD34-,MHCII- **ICC:** MHCII-	Adipogenic, chondrogenic, osteogenic	(Barberini et al. 2014)
	FCA: CD29+,CD44+,CD73+,CD90+,CD105+,CD45-,CD79α- MHCI+,MHCII-,monocyte marker- **qPCR**: CD40+,CD80+,CD86-,TGFβ+,HGF±,IDO-TNFα-	Adipogenic, chondrogenic, osteogenic,	(De Schauwer et al. 2014)
	FCA: CD29+,CD44±,CD73-,CD90-,CD105-,CD14-,CD34-, CD45-,CD79α-,MHCII-		(Paebst et al. 2014)
		Chondrogenic	(Co et al. 2014)
Caprine	**AP**+ **RT-PCR:** NANOG-		(Babaei et al. 2008)
	AP+ **FCA:** CD44+,CD34- **ICC:** αSMA+		(Azari et al. 2011)
	FCA: STRO1+,CD73+,CD105+,CD34- **ICC:** STRO1+,CD73+,CD105+,CD34-	Adipogenic, chondrogenic, osteogenic	(Pratheesh et al. 2014)
Dog	**RT-PCR**: CD29+,CD44+,CD184+,OCT4+,CD34-,CD45-, DLA-DQA1-,DLA79-,DLA88-	Adipogenic, chondrogenic, osteogenic, neurogenic	(Filioli Uranio et al. 2011)

	FCA: CD44+,CD73+,CD90+,CD105+,CD14-,CD34-,CD45-	Osteogenic	(Kang et al. 2012)
	FCA: CD44+,CD73+,CD90+,CD105+,CD14-,CD34-,CD45-	Adipogenic, osteogenic, neurogenic	(Ryu et al. 2012)
	FCA: CD90+,CD105+,CD11c-,CD34-,CD3-,CD45-,CD28-, CD38-,CD62L,CD41a-	Adipogenic, chondrogenic, osteogenic, neurogenic	(Seo et al. 2012)
	RT-PCR: CD44+,CD54+,CD61+,CD80+,CD90+,CD105+	Chondrogenic	(Lee et al. 2013a)
	FCA: CD44+,CD90+,CD105+,CD184+,CD29-,CD33-,CD34-,CD45- **ICC**:Oct3/4+,Nanog+,Sox2+,SSEA4+,CD44+,CD90+,CD105+,CD184+ **RT-PCR**: OCT3/4+,NANOG+,SOX2+,CD44+,CD90+,CD105+, CD184+,CD29-,CD34-,CD45-	Adipogenic, chondrogenic, osteogenic, neurogenic	(Lee et al. 2013b)
	FCA: CD29+,CD44+,CD184+,OCT4+,CD34-,CD45-, DLA-DRA-,DLA-DQA1-,DLA79-,DLA88-	Osteogenic, neurogenic	(Filioli Uranio et al. 2014)
Bovine	**RT-PCR**: OCT4+,ITSN1+	Adipogenic, chondrogenic, osteogenic, neurogenic	(Cardoso et al. 2012a)
		Neurogenic	(Cardoso et al. 2012b)
Buffalo	**FCA:** CD29+,CD73+,CD90+,CD105+,CD34-,CD36-,CD45- **RT-PCR**: OCT4+,NANOG+,SOX2+	Adipogenic, osteogenic	(Singh et al. 2013)
	AP+ **ICC**: Oct4+,Nanog+,Sox2+,CD73+,CD90+,CD105+,CD34- **RT-PCR**: CD73+,CD90+,CD105+ **qPCR**: OCT4+,NANOG+,SOX2+	Adipogenic, chondrogenic, osteogenic	(Sreekumar et al. 2014)

697 + : positive; - : negative; ± : weakly positive; ICC: immunocytochemistry

698 **Table 5**. Clinical applications of foetal MSCs in domestic animals.

MSCs source	Species	Clinical application	Ref.
AM	Sheep	Diskectomy with bone graft	(Goldschlaget et al. 2011)
	Horse	**Spontaneous** tendon and ligament injuries	(Lange-Consiglio et al. 2012; 2013b; 2013c)
	Pig	Myocardial ischemia	(Kimura et al. 2012)
AF	Sheep	Diaphragmatic repair with graft	(Fuchs et al. 2004; Turner et al. 2011)
		Prenatal tracheal reconstruction with scaffold	(Kunisaki et al. 2006b; Gray et al. 2012)
		Diaphragmatic repair	(Kunisaki et al. 2006c)
		Prenatal treatment with lentiviral transducted cells	(Shaw et al. 2011)
		Prenatal cell-based heart valves implantation	(Weber et al. 2012)
		Tendon injuries with nucleofected cells	(Colosimo et al. 2013a)
	Swine	Cardiac ischemia	(Sartore et al. 2005)
		Myocardial infarction in mouse	(Peng et al. 2014)
	Horse	**Spontaneous** decubitus ulcers with PRP	(Iacono et al. 2012c)
UCB	Horse	Intra-articular injection	(Carrade et al. 2011)
		Spontaneous tendinitis of superficial digital flexor tendon	(Kang et al. 2013)
		Spontaneous tendon and ligament disorders	(Van Loon et al. 2014)

	Dog	Spinal cord injury	(Lim et al. 2007; Park et al. 2011)
		Diaphyseal defect in the radius	(Jang et al. 2008)
		Bone regeneration in ectopic implantations	(Byeon et al. 2010)
		Segmental bone defects	(Kang et al. 2012)
WJ/UCM	Swine	Transplantation into rat brains	(Weiss et al. 2003)
		Allogeneic engraftment in the neonatal intestine	(Miller et al. 2012)
	Caprine	Skin wounds	(Azari et al. 2011)
		Skin wounds in rat	(Pratheesh et al. 2014)
	Horse	Intra-articular injection	(Carrade et al. 2011)
	Dog	Segmental bone defects	(Kang et al. 2012)
		Spinal cord injuries	(Ryu et al. 2012)

699