FEATURED ARTICLE

Discussion of Animal Stem Cells in the Classroom: Engaging Students through the Lens of Veterinary Medicine

STEPHEN J. FARENGA, DANIEL NESS, MICHAEL HUTCHINSON

Abstract

Learning about stem cells within the context of treating pet illness or injury is an additional way for teachers to discuss the integration of science, technology, and veterinary medicine. We explain how practitioners in veterinary medicine harvest animal stem cells from adipose (fat) tissue in treating pet illness or injury. Further, we narrate how the veterinarian's approach to pet stem cell therapy demonstrates an important step in technological progress in science, one that may lead to medical advances for humans.

Key Words: *Stem cells; mesenchymal cells; cellular biology; molecular medicine; genetics; veterinarian medicine; technology in veterinarian medicine; autoimmune disease.*

Pets influence us on a daily basis by offering companionship, happiness, assistance, and even rehabilitation. Within the past decade, they have been members of more than 43 million households in the United States (Oyama & Serpell, 2013). In fact, the U.S. pet industry has grown to a nearly $60 billion business (Boler & Fahey, 2012). Nearly one-quarter of this amount is spent on pet health (Henderson, 2013). In response to the high premium placed on the well-being of pets, an increasing number of veterinary professionals have begun to offer advanced methods of treatment for debilitating illnesses that, in the recent past, left pet owners with fewer options for ailing or injured pets (Farenga et al., 2008). Alongside traditional treatments, veterinary medicine has advanced to the point where specialists are treating an increasing number of pets with stem cell therapy to improve numerous maladies in various animals, such as dogs, cats, horses, and birds (Black et al., 2007; Frisbie & Smith, 2010; Ganey et al., 2009; Marx et al., 2014).

Stem cells comprise a class of cells that divide for indefinite periods in culture and develop into specialized cells.

The purpose of this article is to explain the harvesting of adipose-derived mesenchymal stem cells (AD-MSCs) from the standpoint of veterinary medicine. In doing so, we provide science teachers with an alternative approach to teaching the subject of stem cells. Further, we demonstrate how the veterinarian's practice in stem cell therapy using adipose (fat) tissue has contributed to a large and important step in technological advancement for biology and for science in general. In examining this topic, however, it will be necessary to define several important terms. Doing so will help readers navigate through the discussion of stem cell therapy, while, at the same time, considering the importance of this topic from medical and technological perspectives.

What Are Stem Cells?

Stem cells comprise a class of cells that divide for indefinite periods in culture and develop into specialized cells. A stem cell can do two things that a non–stem cell cannot: first, it can renew itself by dividing; and second, it is capable of multilineage differentiation, which means that it can separate into different types of cells (Till & McCulloch, 1961). There are two types of stem cells that are familiar to most people: embryonic stem cells (ESCs) and adult stem cells (ASCs).

ESCs are undifferentiated cells that form after fertilization and are capable of dividing without differentiating for an extended period (Rippon & Bishop, 2004). It may help to visualize a timeline when thinking about ESCs and the terminology used to describe the biological processes associated with development (Figure 1). ESCs are sometimes categorized according to "potency," a term that describes their capacity to differentiate. The ability for ESCs to differentiate into different cell types as the organism develops decreases over time. As time progresses, these cells achieve a greater level of specialization that delimits their functions (Rippon & Bishop, 2004). This gives them the potential to transform from totipotency to

The American Biology Teacher, Vol. 77, No 6, pages. 405–412, ISSN 0002-7685, electronic ISSN 1938-4211. © 2015 by National Association of Biology Teachers. All rights reserved. Please direct all requests for permission to photocopy or reproduce article content through the University of California Press's Reprints and Permissions web page, www.ucpress.edu/journals.php?p=reprints. DOI: 10.1525/abt.2015.77.6.405.

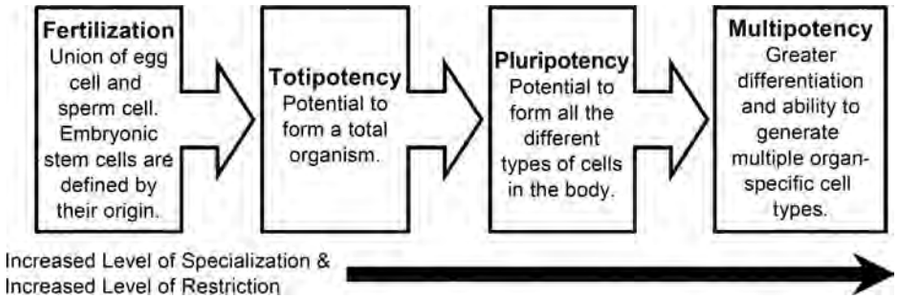


Figure 1. Embryonic stem cell development timeline.

pluripotency to multipotency (Figure 1). At present, we have not learned how to harvest embryonic stem cells without endangering the embryo. This endangerment is an important explanation for the controversy of harvesting embryonic stem cells and conducting embryonic stem cell research. That said, biologists and other research scientists have made advances in harvesting multipotent stem cells from adult tissue (Vieira et al., 2010).

ASCs differ from ESCs in their potential to differentiate. Given that ASCs are limited to the cell type for the specific tissue or organ where they are located, the field of regenerative medicine associated with these stem cells remains an actively researched area in biology (Cohen, 2013; Marx et al., 2014). At present, scientists are still conducting research to determine the origin of different types of ASCs (Ramdass & Koka, 2014). ASCs share some limited commonality with ESCs in that they have the capacity of both self-renewal and differentiation. ASCs may remain inactive until the appropriate conditions occur. For example, ASCs can be activated after an injury and migrate to the surrounding tissue. However, the role of regeneration varies with the type of tissue that has been injured, the severity of the injury, and the specific type of ASCs that are activated. In addition, research suggests that, in most instances, the quantity of ASCs is small, is difficult to harvest, and declines with age (Weissman, 2000).

○ What ASCs Can Do

Current scientific evidence suggests five broad areas in which ASC therapy can promote healing. First, ASCs provide an anti-inflammatory effect by modulating immune function. The literature is replete with positive results involving procedures that entail a variety of organ and tissue transplants as well as treatments that may involve the infusion of bodily fluids (Aggarwal & Pittenger, 2005; Meyerrose et al., 2007; Nasef et al., 2008; Wu et al., 2003). Second, ASCs migrate to damaged tissues through a biochemical signaling process and recruit other cells that secrete cytokines and other proteins. Third, ASCs support tissue remodeling over scar formation by the release of progenitor cells, which aid the regenerative process by producing a variety of growth factors that quicken recovery. However, unlike ASCs, they are incapable of renewal and are restricted in terms of the type of cells they can become. The role of progenitor cells is exemplified by endothelial progenitor cells that (1) are undifferentiated; (2) are located in the adult bone marrow or circulate in the blood; and (3) have the potential to promote revascularization, which is necessary for tissue growth (Hill et al., 2003; Barco et al., 2014). Fourth, ASCs inhibit apoptosis – namely, ASCs appear to turn off the process of cell death in which a programmed sequence of events leads to the elimination of cells around the injury site (Glennie et al., 2005; Plumas et al., 2005). And last, depending on their origin, ASCs can differentiate into a variety of tissue types. In canines and equines, clinical practice has demonstrated vast improvements in mobility due to the regeneration of bone, cartilage, tendon, and ligament tissue (Black et al., 2007; Ganey et al., 2009; Gingerich & Strobel, 2003; Marx et al., 2014; Nathan et al., 2003).

So, the challenge for researchers is fourfold: (1) to increase the number of ASCs grown in laboratories; (2) to manipulate the ASCs to produce specific cell types; (3) to determine the best use of ASCs in treating injury or disease; and (4) to address ethical concerns effectively. One possible solution to the challenge of stem cell research may come from what many would consider an unlikely source: adult mesenchymal stem cells (MSCs). MSCs can be isolated for therapeutic use by obtaining them from bone marrow, adipose tissue, lung tissue, the placenta, the umbilical cord, blood, and dental pulp. Specialists in veterinary medicine research have demonstrated that adipose tissue can provide the most promising results in obtaining the greatest concentration of MSCs (Frisbie & Smith, 2010; Vieira et al., 2010; Cohen, 2013; Barco et al., 2014; Chan et al., 2014; Marx et al., 2014). In fact, research suggests that adipose tissue, commonly known as "fat tissue," is one of the most productive sources of MSCs (Cohen, 2013; Marx et al., 2014; Ramdass & Koka, 2014) and has the potential to differentiate into a variety of tissue types (Figure 2). Further, a growing number of veterinarians are currently engaging in stem cell therapy in their clinical practice (Marx et al., 2014).

○ Adipose Tissue: A Successful Source of Adult Stem Cells

AD-MSCs can be produced in great quantity in clinical situations, have the ability to become different cell types, and are used to treat a variety of conditions (Marx et al., 2014). AD-MSCs are part of the tissue stroma, are self-renewable, and can differentiate into cells with the specific function required to alleviate certain debilitating conditions (Le Blanc, 2006).

Research in veterinary medicine has shown that stem cells can be harvested more easily from adipose tissue than from other stem cell sources in the body (di Summa et al., 2010; Marx et al., 2014). Adipose tissue contains a plentiful supply of MSCs. In addition, veterinarians have reported in clinical trials that obtaining AD-MSCs from subcutaneous or abdominal adipose tissue tends to be far less painful for an animal receiving medical treatment, compared with a number of standard procedures (see the case study below). Initial studies suggest that AD-MSCs have an impressive track record for promoting healing and repair (Cohen, 2013; Bashir et al., 2014; Chan et al., 2014; Marx et al., 2014; Ramdass & Koka, 2014). Even though AD-MSCs have a mesodermal origin, a number of studies demonstrate beneficial outcomes on body structures

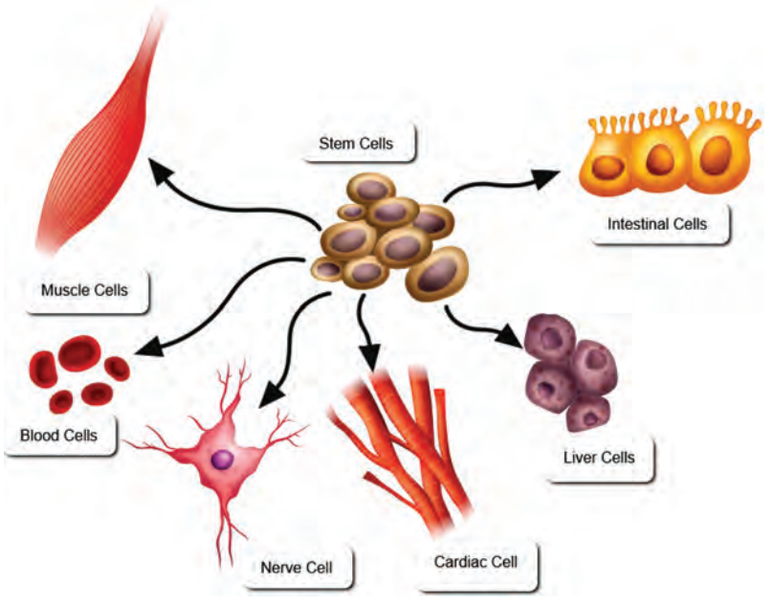


Figure 2. Capabilities of adipose-derived mesenchymal stem cells. Diagram used with permission of Animal General Inc., Cranberry, PA: 2014.

Table 1. Three methods for mesenchymal stem cell treatment.

Procedure	Donor		Recipient	Description
Autograft	Dog A Rex	to to	Dog A Rex	Organism donates required cells or tissue to itself for treatment
Allograft	Dog A Rex	to to	Dog B Fido	Donor of the same species, but different tissue taken from Dog A and used to treat Dog B
Xenograft	Sheep Dolly	to to	Dog A Rex	Donor is from a different species

that emanate from ectodermal and endodermal lineages (di Summa et al., 2010; Marsella et al., 2011; Cohen, 2013; Barco et al., 2014).

○ Therapeutic Use of AD-MSCs in Veterinary Medicine

Veterinarians have treated animals with stem cell therapy mainly in preclinical trials. Some trials have suggested benefits that are dependent on four conditions: (1) the animal being treated; (2) diagnosed illness; (3) method of treatment; and (4) appropriate follow-up and rehabilitation. In fact, a number of studies have reported positive effects of AD-MSC therapy in the treatment of debilitating conditions. These conditions include, but are not limited to, the following: the regeneration and repair of tissues and cells associated with cardiac-related illness such as postischemia (Wu et al., 2003; Perin et al., 2014), diabetes, osteoarthritis, tendon injury, and sepsis (Aggarwal & Pittenger, 2005; Bashir et al., 2014; Ramdass & Koka, 2014). An important factor in treatment and recovery is that MSCs in general, and AD-MSCs in particular, have the potential for an immunomodulatory effect, thereby lessening the chances of host graft rejection (Aggarwal & Pittenger, 2005; Le Blanc, 2006). In other words, the AD-MSCs used in treatment will not trigger a response from the organism's immune system and, therefore, will likely have both innate and adaptive immunity. Remarkably, as shown in Table 1, this has been demonstrated in procedures that involve autografts, allografts, and xenografts (Wu et al., 2003; Aggarwal & Pittenger, 2005; Le Blanc, 2006; Meyerrose et al., 2007). Further, the mesenchymal stem cell suspension appears to bring about ameliorative actions that may reduce nonspecific immune responses, such as acute and chronic inflammation (Le Blanc, 2006; Nasef et al., 2008; Marsella et al., 2011).

How Veterinarians Conduct AD-MSC Therapy

The following is a generalizable method by which veterinarians obtain AD-MSCs in clinical practice. For our purposes, this procedure outlines the use of AD-MSCs for improving conditions of mobility affected by chronic inflammation, osteoarthritis, tendon injury, or lameness in dogs, horses, and cats.

To begin with, surgeons must use aseptic techniques at all times. The first task of the veterinarian surgeon is to obtain adipose tissue from the animal requiring treatment (Figure 3). The surgeon then places the adipose tissue in a sterile container and cuts the tissue into smaller pieces to increase the tissue's surface area. The cut-up tissue is then placed in a test tube along with antibiotic wash (Figure 4). Next, digestive serums are injected into the adipose tissue. The technician then mixes the tissue and places the tissue-filled test tube in a water bath for the purpose of incubation. It is then placed in a centrifuge to remove supernatant fluid. The cell pellet, which contains potential mesenchymal stem cells, can be found at the bottom of the test tube container. The solution containing the cell pellet must be filtered and placed back in the test tube container (Figure 5). The process of centrifuging and filtering is repeated twice. Finally, the cell pellet is now ready to be used for treatment.

Getting AD-MSCs to Work

There are multiple methods of administering AD-MSCs to the animal in treatment. The two most common are the following: The first is to inject the stem cells and solution directly into the affected tissue or joint. The second is to deliver the stem cells in solution through an intravenous drip. Research has also demonstrated benefits by injecting stem cells directly into muscles or infected organs (Jung et al., 2009; di Summa et al., 2010; Bahrani, 2012; Marx et al., 2014; Ramdass & Koka, 2014). In some cases, direct injection and the intravenous drip are both used in treatment. Stem cells are

Figure 3. Adipose tissue obtained from a dog requiring treatment. Photograph used with permission of Animal General Inc., Cranberry, PA: 2014.

Figure 4. Adipose tissue placed in container with antibiotic wash and digestive fluids. Photograph used with permission of Animal General Inc., Cranberry, PA: 2014.

homeostatically regulated (Domen & Weissman, 1999; Ramdass & Koka, 2014), which means that, through physiological processes, they can maintain internal equilibrium on their own. Complex feedback loops exist, which control how stem cells self-renew, differentiate, commit cell death, and emigrate. The function of the stem cell is controlled by a combination of external and internal mechanisms. Internal cellular regulation is directed by signaler proteins produced by their own cellular machinery. In general, three external mechanisms regulate a cell's developmental choices: chemicals secreted by other cells, physical contact among surrounding cells, and molecules found in the microenvironment. Regenerative medicine models show that mesenchymal stem cells respond to chemicals secreted by other cells at the site of the damaged tissue (Cohen, 2013; Marx et al., 2014). The injury site attracts the mesenchymal stem cells and other proteins, which, in turn, establish chemical pathways to the injury as a means of starting the process of repair. When the mesenchymal stem cells get to the injury site, they have the potential to repair damaged tissue by proliferation and matrix production. Receptor proteins on cell membranes, called "integrins," help regulate a variety of cellular processes (Clark & Brugge, 1995). Integrins are critical to adhesive interactions, which control the migration, proliferation, differentiation, and communication of cells and are associated with a vital communication process known as "signaling" that transmits environmental information from the outside in (to the cell) and from the inside out (from the cell to the surrounding tissue).

Cellular proliferation and adhesion are regulated by elaborate interactions among cell biochemistry, structural mechanisms, and surface bonding (Evans & Calderwood, 2007). At least one of the factors in cell growth and adhesion appears to be regulated by the pressure found around the surrounding cells. An on-and-off mechanism that controls the molecular interactions seems to be regulated by the pressure applied at the intercellular sites. This helps explain the formation of the extracellular matrix, where the cells adhere to the repair site, produce new cells, and form and connect to new tissue. The extracellular matrix supplies the architecture and strength to the surrounding cells. At the time of tissue repair, combinations of protein fibers and nonfibrous substances serve a connective function between groups of cells in the stroma. At the repair site, chemokines allow for cellular communication between and among groups of cells. However, the signaling processes of intercellular and intracellular communication are not fully understood. Research scientists are still looking for the exact mechanisms to control the regenerative process in order to develop more effective treatments.

The therapeutic use of AD-MSCs in veterinary medicine has the potential not only to improve the lives of our pets, but also to supply clinical trials as evidence and as a basis for beginning human trials and treating human illness. Although the reports from some studies suggest positive results, more data are needed to answer specific

Figure 5. (Top layer) Yellow unsaturated fatty layer. (Middle layer) White fibrin and undigested fatty layer. (Bottom layer) Red layer + pellet (stromal vascular fraction [or SVF] with stem cells). Photograph used with permission of Animal General Inc., Cranberry, PA: 2014.

questions regarding AD-MSC treatment. The field of regenerative medicine is probably one of the most pioneering areas that demonstrate the need for teaching and learning of scientific and mathematical inquiry skills. Prior to discussion, students will need a foundational understanding of the differences between the use of adult stem cells and of embryonic stem cells. Further, students will be faced with making decisions that are generated by the impact from the advances of science and technology on the field of medicine. The following case study involving Sadie exemplifies the cutting-edge research in stem cell therapies as it applies to veterinary medicine. This successful and encouraging case provides evidence that AD-MSC therapy has the potential to transcend current frontiers in regenerative medicine for both animals and humans. Upon examining the case of Sadie, have students engage in activities that are based on the questions that follow.

○ Extension Activity Involving a Canine Case Study: Case Notes & Comments

The patient, Sadie, is a 4½-year-old female cockapoo – a dog breed that is half cocker spaniel and half poodle. Prior to treatment on November 20, 2012, she was suffering from atopic dermatitis ("atopy" for short), which is an autoimmune disease caused by complex genetic and environmental factors that adversely affects skin (Marsella et al., 2011). Atopy is characterized by incessant scratching that often leads to severe sores, lesions, and ulcers (Figure 6). In many instances, secondary infections are common, many of which lead to additional complications. Given her condition, Sadie was allergic to almost everything within her environment.

The traditional protocol for standard treatment of this condition includes omega-3 fatty acids, steroid-based medications that may be given by pill or by long-acting injections, medicated baths, and antibiotics to fight secondary infections. Unfortunately, none of these treatment methods proved effective in Sadie's case, and it seemed that Sadie might have to be euthanized. But a final option was suggested: that Sadie be treated with stem cell

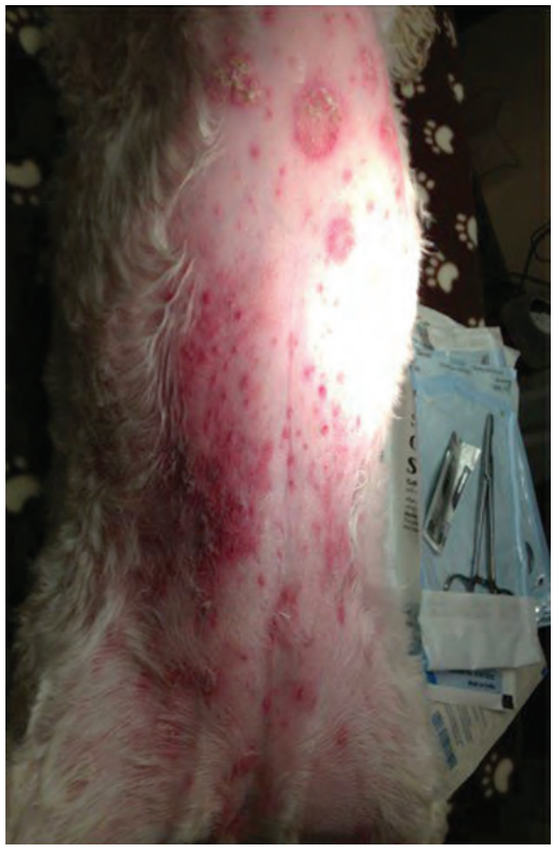


Figure 6. Sadie on day 0 of treatment. Photograph used with permission of Animal General Inc., Cranberry, PA: 2014.

therapy based on AD-MSCs. The use of AD-MSC to treat atopy was considered experimental. Prior to this point, AD-MSCs were primarily used to treat arthritis, ligament issues, and bone fractures. It should be noted that the use of AD-MSCs is not necessarily a cure for atopy. However, Sadie has remained symptom free since the final AD-MSC treatment. Practitioner-based evidence suggests that the AD-MSC treatment lasts for ~18 months prior to a second administration of the treatment (M. Hutchinson, personal observation).

○ Student Inquiry

Many students may be surprised to learn that there are many similarities with regard to atopy in canines and humans (Marsella et al., 2011). Students can investigate the standard of care for treating atopy. Afterward, have students examine the photographs of Sadie from Day 0, Day 30, and Day 60 (Figure 7).

After reading the case notes and making careful observations of clinical data provided in Figure 7, have students answer the following questions:

1. What scientific, technological, and humanitarian questions do the photographs raise?
2. What was the importance of using the AD-MSCs in Sadie's case?

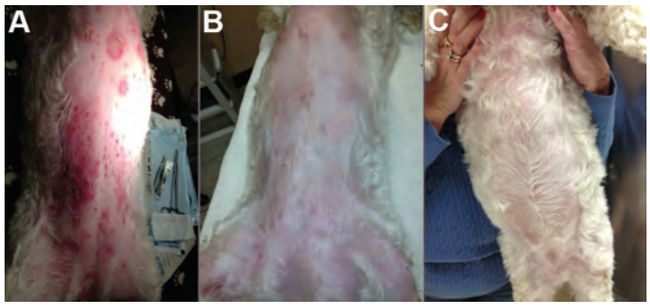


Figure 7. Progression of AD-MSC Treatment from day 0 to day 60. (**A**) Day 0; (**B**) Day 30; (**C**) Day 60. Photographs used with permission of Animal General Inc., Cranberry, PA: 2014. As of September 15, 2013, Sadie is still 100% clear. Based on three dogs with similar atopic conditions, we are expecting 17 to 18 months before retreatment is necessary.

3. What are your thoughts on the use of AD-MSCs and stem cells in general in treating disease, even if the treatment may not be a cure?
4. Explain your position to the ethical implications in using experimental treatments prior to demonstrating their clinical effectiveness.
5. What challenges are presented in the use of AD-MSCs? Discuss these challenges.
6. How has the case study of Sadie changed your understanding of science and technology and their effects on applied medicine?
7. What treatment measures would you have considered if Sadie were your companion?

References

Aggarwal, S. & Pittenger, M.F. (2005). Human mesenchymal stem cells modulate allogeneic immune cell responses. *Blood, 105,* 1815–1822.

Bahrani, H., Razmkhah, M., Ashraf, M.J., Tanideh, N., Chenari, N., Khademi, B. & Ghaderi, A. (2012). Differentiation of adipose-derived stem cells into ear auricle cartilage in rabbits. *Journal of Laryngology and Otology, 126,* 770–774.

Barco, R., Encinas, C., Valencia, M., Carrascal, M.T., García-Arranz, M. & Antuña, S. (2014). Use of adipose-derived stem cells in an experimental rotator cuff fracture animal model. *Revista Española de Cirugía Ortopédica y Traumatología, 59,* 3–8.

Bashir, J., Sherman, A., Lee, H., Kaplan, L. & Hare, J.M. (2014). Mesenchymal stem cell therapies in the treatment of musculoskeletal diseases. *PM&R, 6,* 61–69.

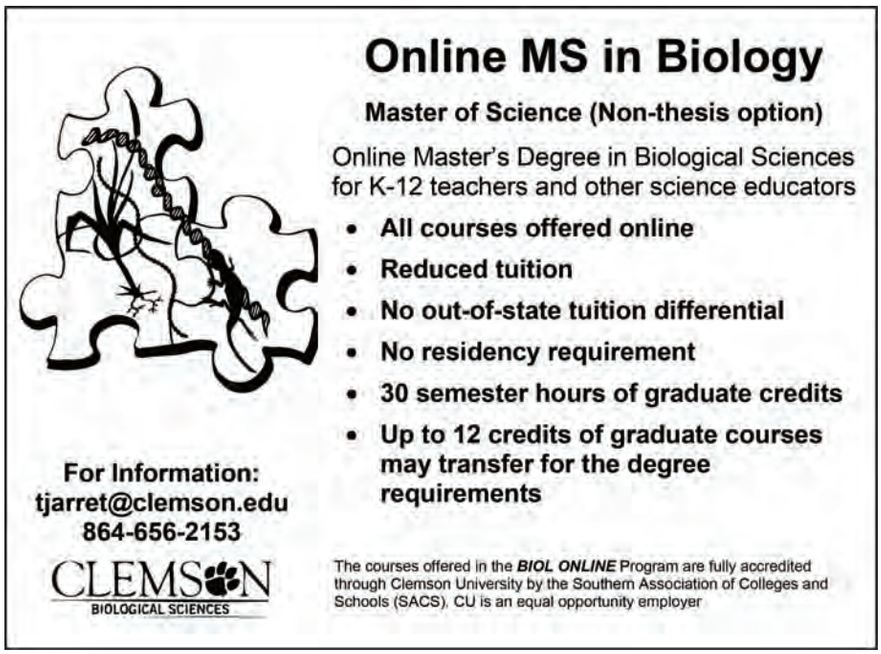
Black, L.L., Gaynor, J., Gahring, D., Adams, C., Aron, D., Harman, R. & others (2007). Effect of adipose-derived mesenchymal stem and regenerative cells on lameness in dogs with chronic osteoarthritis of the coxofemoral joints: a randomized, double-blinded, multicenter controlled trial. *Veterinary Therapeutics, 8,* 272–284.

Boler, B.M.V. & Fahey, G.C., Jr. (2012). Current status of practical applications: pets. In T.R. Callaway & S.C. Ricke (Eds.), *Direct-Fed Microbials and Prebiotics for Animals: Science and Mechanisms of Action* (pp. 75–78). New York, NY: Springer.

Chan, L.L.Y., Cohen, D.A., Kuksin, D., Paradis, B.D. & Qiu, J. (2014). Automated enumeration and viability measurement of canine stromal vascular fraction cells using fluorescence-based image cytometry method. *Journal of Fluorescence, 24,* 983–989.

Clark, E.A. & Brugge, J.S. (1995). Integrins and signal transduction pathways: the road taken. *Science, 268,* 233–239.

Cohen, D.A. (2013, November). Gene expression profiling of canine adipose derived stem cell preparations. Paper presented at the Third



Annual Conference of the North American Veterinary Regenerative Medicine Association, Atlanta, GA.

di Summa, P.G., Kingham, P.J., Raffoul, W., Wiberg, M., Terenghi, G. & Kalbermatten, D.F. (2010). Adipose-derived stem cells enhance peripheral nerve regeneration. *Journal of Plastic, Reconstructive & Aesthetic Surgery, 63,* 1544–1552.

Domen, J. & Weissman, I.L. (1999). Self-renewal, differentiation or death: regulation and manipulation of hematopoietic stem cell fate. *Molecular Medicine Today, 5,* 201–208.

Evans, E.A. & Calderwood, D.A. (2007). Forces and bond dynamics in cell adhesion. *Science, 316,* 1148–1153.

Farenga, S.J., Ness, D. & Hutchinson, M. (2008). Developing an awareness of pet stewardship. *Science Scope, 32,* 58–63.

Frisbie, D.D. & Smith, R.K.W. (2010). Clinical update on the use of mesenchymal stem cells in equine orthopaedics. *Equine Veterinary Journal, 42,* 86–89.

Ganey, T., Hutton, W.C., Moseley, T., Hedrick, M. & Meisel, H.J. (2009). Intervertebral disc repair using adipose tissue-derived stem and regenerative cells: experiments in a canine model. *Spine, 34,* 2297–2304.

Gingerich, D. & Strobel, J. (2003). Use of client-specific outcome measures to assess treatment effects in geriatric, arthritic dogs: controlled clinical evaluation of a nutraceutical. *Veterinary Therapeutics, 4,* 56–66.

Glennie, S., Soeiro, I., Dyson, P.J., Lam, E.W. & Dazzi, F. (2005). Bone marrow mesenchymal stem cells induce division arrest anergy of activated T cells. *Blood, 105,* 2821–2827.

Henderson, S. (2013). Spending on pets: "Tails" from the Consumer Expenditure Survey. Beyond the Numbers, 2(16). [Online]. Washington, DC: U.S. Bureau of Labor Statistics. Available at http://www.bls.gov/opub/btn/volume-2/spending-on-pets.htm

Hill, J.M., Zalos, G., Halcox, J.P.J., Schenke, W.H., Waclawiw, M.A., Quyyumi, A.A. & Finkel, T. (2003). Circulating endothelial progenitor cells, vascular function, and cardiovascular risk. *New England Journal of Medicine, 348,* 593–600.

Jung, M., Kaszap, B., Redöhl, Steck, E., Breusch, S., Richter, W. & Gotterbarm, T. (2009). Enhanced early tissue regeneration after matrix-assisted autologous mesenchymal stem cell transplantation in full thickness chondral defects in a minipig model. *Cell Transplantation, 18,* 923–932.

Le Blanc, K. (2006). Mesenchymal stromal cells: tissue repair and immune modulation. *Cytotherapy, 8,* 559–561.

Marsella, R., Thierry, O. & Carlotti, D.-N. (2011). Current evidence of skin barrier dysfunction in human and canine atopic dermatitis. *Veterinary Dermatology, 22,* 239–248.

Marx, C., Silveira, M.D., Selbach, I., da Silva, A.S., Braga, L.M., Camassola, M. & Nardi, N.B. (2014). Acupoint injection of autologous stromal vascular fraction and allogeneic adipose-derived stem cells to treat hip dysplasia in dogs. *Stem Cells International 2014,* 391274.

Meyerrose, T.E., De Ugarte, D.A., Hofling, A.A., Herrbrich, P.E., Cordonnier, T.D., Shultz, L.D. & others (2007). In vivo distribution of human adipose-derived mesenchymal stem cells in novel xenotransplantation models. *Stem Cells, 25,* 220–227.

Nasef, A., Ashammakhi, N. & Fouillard, L. (2008). Immunomodulatory effect of mesenchymal stromal cells: possible mechanisms. *Regenerative Medicine, 3,* 531–546.

Nathan, S.T., Das De, S., Thambyah, A., Fen, C., Goh, J. & Hin Lee, E. (2003). Cell-based therapy in the repair of osteochondral defects: a novel use for adipose tissue. *Tissue Engineering, 9,* 733–744.

Oyama, M.A. & Serpell, J.A. (2013). General Commentary: rethinking the role of animals in human well-being. *Frontiers in Psychology, 4,* 374.

Perin, E.C., Sanz-Ruiz, R., Sánchez, P.L., Lasso, J., Pérez-Cano, R., Alonso-Farto, J.C. & others (2014). Adipose-derived regenerative cells in patients with ischemic cardiomyopathy: the PRECISE Trial. *American Heart Journal, 168,* 88–95.

Plumas, J., Chaperot, L., Richard, M.-J., Molens, J.-P., Bensa, J.-C. & Favrot, M.-C. (2005). Mesenchymal stem cells induce apoptosis of activated T cells. *Leukemia, 19,* 1597–1604.

Ramdass, B. & Koka, P.S. (2014). Ligament and tendon repair through regeneration using mesenchymal stem cells. *Current Stem Cell Research & Therapy, 10,* 84–88.

Rippon, H.J. & Bishop, A.E. (2004). Embryonic stem cells. *Cell Proliferation, 37,* 23–34.

Till, J.E. & McCulloch, E.A. (1961). A direct measurement of the radiation sensitivity of normal mouse bone marrow cells. *Radiation Research, 14,* 213–222.

Vieira, N.M., Brandalise, V., Zucconi, E., Secco, M., Strauss, B.E. & Zatz, M. (2010). Isolation, characterization, and differentiation potential of canine adipose-derived stem cells. *Cell Transplantation, 19,* 279–289.

Weissman, I.L. (2000). Translating stem and progenitor cell biology to the clinic: barriers and opportunities. *Science, 287,* 1442–1446.

Wu, G.D., Nolta, J.A., Jin, Y.-S., Barr, M.L., Hong, Y., Starnes, V.A. & Cramer, D.V. (2003). Migration of mesenchymal stem cells to heart allografts during chronic rejection. *Transplantation, 75,* 679–685.

STEPHEN J. FARENGA is a Professor of Science Education at Queens College, City University of New York, 65-30 Kissena Blvd, Flushing, NY 11367; e-mail: sfarenga@qc.cuny.edu. DANIEL NESS is an Associate Professor at St. John's University, 8000 Utopia Pkwy, Jamaica, NY 11439; e-mail: nessd@stjohns.edu. MICHAEL HUTCHINSON, DVM, is a Clinical Director of Animal General, Cranberry Township, PA 16066; e-mail: info@animalgeneral.net.