Cellular and Molecular Biology
E-ISSN : 1165-158X / P-ISSN : 0145-5680
www.cellmolbiol.org
Review

Application of stem cell for the regeneration of spiral ganglion neurons

F. Mohammadian[1*], A. Eatemadi[2,3], H. Daraee[2,3]

[1] Department of Medical Biotechnology, Faculty of Advance Medical Sciences, Tabriz University of Medical Sciences, Tabriz, Iran
[2] Department of Medical Biotechnology, School of advance Science in Medicine, Tehran University of Medical Sciences, Tehran, Iran
[3] Department of Medical Biotechnology, School of Medicine, Lorestan University of Medical Sciences, Lorestan, Iran

Correspondence to: mohamadianf@yahoo.com

Received August 26, 2016; **Accepted** January 25, 2017; **Published** January 30, 2017

Doi: http://dx.doi.org/10.14715/cmb/2017.63.1.2

Abstract: Over 278 million of people worldwide suffers from hearing loss, and this disease has significant detrimental effects emotionally and economically on individuals and the society in totality. Treatment using cochlear implant dramatically improve the perception, and production of speech, as well as the patient quality of life, with the different sensorineural hearing loss (SNHL). Yet, there are some challenges faced by a cochlear implant. In this review, we propose the regeneration of spiral ganglion neurons which is an interface neuron using human amniotic fluid mesenchymal stem cells (hAFMSCs), due to its high pluripotency potentials, this stem cell source can regenerate the spiral ganglion and this in-turn will bring back the inner ear hair-cells to functionality.

Key words: Mesenchymal; Stem cell; Inner ear; Spiral ganglion.

Introduction

Biological concepts for alleviating disease, including the restoring of damaged tissue with biological tools, holds great potential when applied as a means of hearing loss therapy (1–3). The most common world's disability is deafness, which increases with age to alter fully one-half of those over 65 years of age (4–7). Replacement of sensory cells occur spontaneously and following genetic manipulation in the vestibular (8), and auditory sensory epithelium (9). The discovery of an additional method to stimulate the growth of new auditory nerve fibers would create an opportunity for deafness treatment that is currently not possible.

Hearing impairment is a generally known disability. Apparently, there are different available models to replace or regenerate the cells within the mammalian cochlea. Generation of neurons or new sensory cells either by activating stem cells, cochlear progenitor cells or by the conversion of supporting cells would be an exciting approach (9–12). However, in a situation where spiral ganglion neurons and hair cells are severely absent or degenerated, a cell replacement therapy based on tissue implantation may offer an interesting and more immediate alternative. The transplanted cells would be anticipated to take the position of missing cochlear cells, and become fully incorporated with the auditory system both functionally and structurally(13–16).

Choice of transplantation of cells is a key issue, and there are different candidates for cell therapy. One of the options is using an embryonic neuronal tissue. Another better cell therapy alternative is to use stem cells(17–19). Embryonic stem (ES) cells are pluripotent and can differentiate into a variety of cell types (20,21). One of the challenges of stem cell transplantation into the human inner ear is to stimulate the implanted cells to a sensorineural or cochlear lineage, that is, cochlear sensory cells and spiral ganglion neurons.

In this review, we summarized the parts of human ear focusing on the defection of the spiral ganglion nerves as the cause of hearing impairment and the potentials for regenerating this nerves using hAFMSCs.

The human ear

External and middle ear

The external ear canal is connected to the tympanic membrane (eardrum). The middle ear possesses a chain of three bones that links the tympanic membrane to the cochlea. Tympanic membrane vibrations are relayed to the cochlea. Cochlea (Fig 1&4) (22) have three parallel fluid pockets. The vibration of the tympanic membrane brings about fluid waves in the cochlea. Organ of Corti (Fig 2) located within the cochlea, between the fluid chambers, they consist of the hair cells that include a hair-like projection from their climax (stereocilia). The physical movement of the stereocilia is converted into a nerve signal which is then transmitted via the spiral ganglion and the relay nuclei in the pons and midbrain to the auditory cortex in the temporal lobe (Fig 2) (16, 39). However, a defective SGN will terminate the signal from getting to the inner part of the ear and thus sensorineural hearing loss ensues.

Auditory system development

The development of human auditory system starts from the fetus and it has its own sequential process. The structural parts of the auditory system develop early. Cochlea in the middle ear structural parts are well formed by 15 weeks' gestational age and are functional

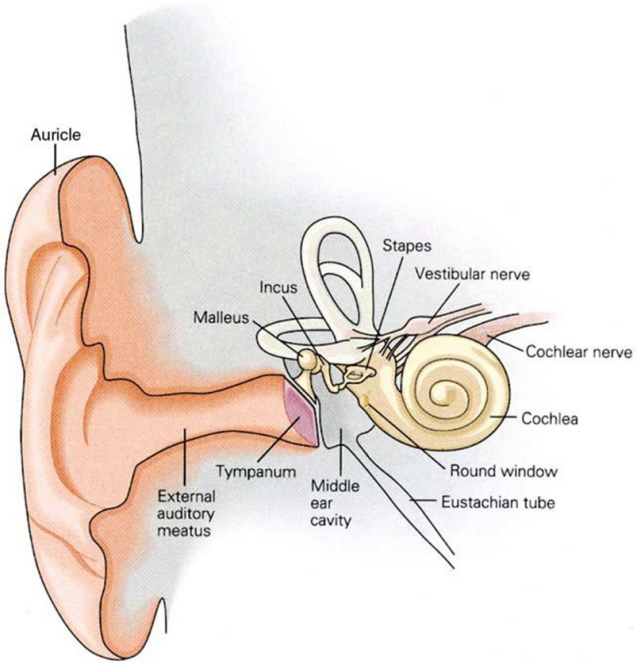

Figure 1. Showing the human ear structure. The external ear, particularly the prominent auricle, focuses sound into the external auditory meatus. Fluctuating increases and decreases in the pressure of air vibrate the tympanum. These vibrations are conveyed across the air-filled middle ear by three tiny, linked bones: the malleus, the incus, and the stapes. The vibration of the stapes stimulates the cochlea, the hearing organ of the inner ear (Reprinted from (24)).

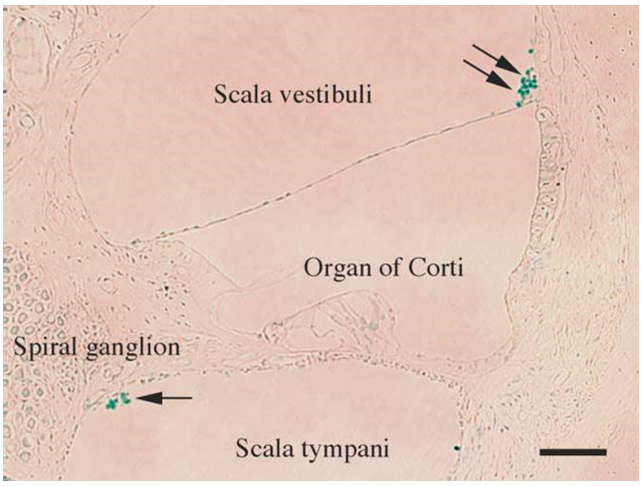

Figure 2. Image illustrating the survival of adult mouse NSCs in the normal guinea pig inner ear 2 weeks following transplantation. Blue-appearing LacZ-expressing implanted cells were found in the scala tympani and scala vestibuli of the inner ear. The implanted cells (arrow) were attached close to the spiral ganglion and the organ of Corti. Surviving transplanted cells (double arrows) were also observed in the scala vestibuli, attached to the lateral bony wall of the scala vestibuli, close to the Reissner membrane. Scale bar: 100 Am (Adapted from Hu et al. (24)).

by 20 weeks' gestation (25,26). The kinesthetic (movement), somaesthetic (touch), vestibular (motion-head), proprioceptive (position), and chemosensory (smell and touch) systems all are both anatomically and functionally operative before 20 weeks' gestation. The auditory system follows those systems in the chain of development.

At around 25 to 29 weeks' gestational age, the auditory system becomes functional, the ganglion cells of the spiral nucleus in the cochlea links the inner hair cells to the temporal lobe, and brain stem and of the cortex (26). The earliest evidence of an auditory evoked response is at 16 weeks' gestational age. During this stage, the ganglion cells in the cochlea are linked to nuclei in the brainstem that triggers a physiologic response. Loud noise in utero or in the NICU will produce changes in autonomic function at 25 to 26 weeks' gestation, blood pressure, heart rate, respiratory pattern, oxygenation, and gastrointestinal motility can all be affected (27). The neural links to the temporal lobe of the cortex become functional 28 to 30 weeks' gestational age.

The cochlea (the receptor organ) and the auditory cortex are the two parts of the auditory system that are most crucial in the developmental processes (23). However, they all relate to the signals received from the neurons of the spiral ganglion and cochlear nuclei of the cochlea. It is the cochlea and auditory cortex in the temporal lobe that is most affected by the environment and the care practices of the NICU.

The cochlea

The mammalian cochlea is a fully-developed and well-structured organ comprising of a large variety of cell types. Although hearing loss is related to the loss of hair cells, the cochlea sensory transducers, hearing impairment also arises from dysfunction of several cochlear cell kinds. For instance, in human, the primarily inherited form of deafness is associated with connexin 26 gene mutation; a cytoplasmic gap junction protein found in several cochlear supporting cells (28). Auditory nerve disease like auditory neuropathy (29) and acoustic Schwannoma (30), involving the auditory neurons and glia degeneration, respectively, also leads to hearing loss (Fig 4).

A biological approach to disease amelioration, involving damaged tissue replacement using biological tools, is promising when applied as an approach to treating hearing loss (2,31). Hair cell regeneration has been

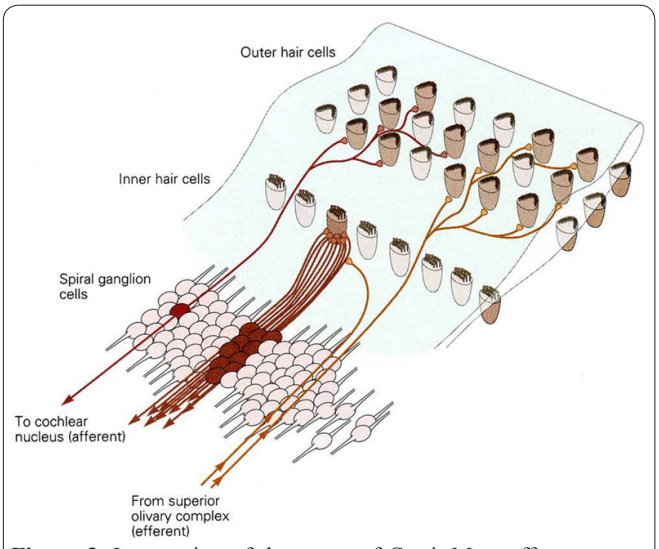

Figure 3. Innervation of the organ of Corti. Most afferent axons end on inner hair cells, each of which constitutes the sole terminus for an average of 10 axons. A few afferent axons of small caliber provide diffuse innervations to the outer hair cells. Efferent axons largely innervate outer hair cells and do so directly. In contrast, efferent innervation of inner hair cells is sparse and is predominantly axoaxonic, at the endings of afferent nerve fibers (Reprinted from (74)).

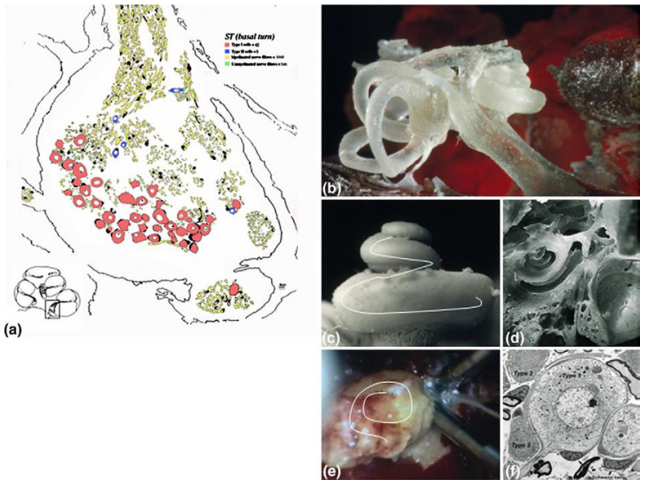

Figure 4. Showing the anatomy of the human inner ear. (a) Computer-based simulation of the human spiral ganglion showing type I (brown) and type II (blue) cells in combination with myelinated (yellow) and un-myelinated (green) nerve fibers. (b) Plastic corrosion cast of a human inner ear showing the vestibular apparatus, the cochlea, and the internal acoustic meatus. (c) Corrosion cast of the human cochlea with spiral ganglion outlined. (d,e) Microdissection of a human cochlea (cadaver) and (e) at surgery (spiral ganglion outlined). (f) Transmission electron microscopy (TEM) showing Type I and Type II ganglion cells (Adapted from (75)).

shown to repair the damaged cochlea of birds and a low level of hair cell genesis continues into adulthood in mammalian vestibular systems, the mature mammalian cochlea has not exhibited any inherent ability to regenerate after trauma. However, injection of viral particles engineered to deliver the transcription factor (Math1) into the cochlea has given rise to new cochlear hair cells production (9,32). An alternative method of replacement of cells is the therapeutic application of exogenous cells capable of differentiating into cochlear cell types. Multipotent cell lines development sourced from stem cells is a practical source of exogenous cells ablated or damaged cochlear cells replacement (33).

Several human and murine stem cell lines have been confirmed to be a good tool for cellular replacement. , the v-myc immortalized murine clonal 17.2 neural stem cell (cNSC) line (34), derived from the cerebellum of the fetus, has been successfully used in different cellular replacement studies consisting of the brain and spinal cord. Transplanted cNSCs have been noted to move to the site of a brain lesion and differentiate into native cell types, such as oligodendrocytes (35), astrocytes, microglia, cortical neurons (34), neurons and spinal cord glia (36). cNSCs are thus capable of both functional recoveries following injury and replacement of damaged (37). In addition, these cNSCs express different markers that are expressed in cochlear tissues, like connexin 26 and the hair cell marker myosin 7a (38,39). Thus, they may also be a useful tool for cellular replacement within the cochlea.

Differentiation of the hair cells (Fig 3) in the cochlea starts early in gestation (10–12 weeks). Stereocilia development on the apex of the hair cells. It starts from the inner hair cells, and later on the outer hair cells. Hair cells development starts from the cochlea base to the apical regions. This is true for both outer and inner hair cells. There are many numbers of hair cells produce early in development, if not used or connected, some disappear. It is a process similitude to the excess ganglion cells of the retina.

More than 90% of the cochlear ganglion cells stimulates inner hair cells. Each axon triggers a single hair cell, but each inner hair cell targets its output to up to 10 nerve fibers. Neural information for hearing sterns almost solely from inner hair cells. At any point along the course of the spiral ganglion in the cochlea, the neurons respond best to the optimal or prime frequency of the inner hair cell. Thus, the tonotopic organization of the auditory cortex, as well as relay nuclei, begins with the postsynaptic site on the inner hair cells. The acoustic sensitivity of axons in the cochlear nerve mirrors the innervations pattern of the spiral ganglion cell. Like the hair cells, each axon has a characteristic frequency of sound for maximal response. There is a tuning curve for the ganglion cell nerve fibers, just as there is for hair cells (23,40).

Inner ear

More than 10% of the world population are challenged with hearing impairment and a resulting deterioration of their communication performance. Hearing impairment is often as a result of injuries affecting inner ear mechanosensory hair cells. Yet, clinical treatment alternatives are limited but the cochlear implant (cochlear prosthesis) has led to a breakthrough in the rehabilitation of the auditory. The cochlear implant bypasses the sensory hair cells and directly stimulates the remaining spiral ganglion neurons (SGNs), partly restoring part of the hearing function even in profoundly deaf patients. Patients benefit from a cochlear implant depends on the integrity of SGNs and functional excitable neurites available for electrical stimulation. The survival and function of SGNs, in turn, depend on trophic inputs provided by their presynaptic and postsynaptic target cells as well as neighboring tissues. Loss of the sensory cells will thus deprive adult SGNs of trophic factors and cause their subsequent degeneration. Identifying possible preventive factors that could arrest this progressive degeneration is of clinical value as it could further enhance the benefits that severely hearing impaired patients get from a cochlear implant.

In a situation where the sensory cells or SGNs are damaged permanently, a simple preventive strategy might not be effective. New hair cells have been suggested to be formed by regeneration (41) or phenotypic trans-differentiation (8),(32) within the adult mammalian inner ear. The regenerative potential of adult SGNs, however, remains to be tested.

Spiral ganglion

In animal models, the loss of SGNs is fast and extensive with up to 60% of neurons lost six weeks after deafness in the guinea pig (42) or after 10 weeks in the rat (43). In a quest to stop spiral ganglion degeneration, and manage the SGNs population available for stimulation through a cochlear implant, the application of exogenous neurotrophins is being investigated as a possible adjunct therapy to the cochlear implant. Delivery of exogenous neurotrophins to the cochlea (BDNF and NT3 precisely) has contributed the most potential intervention (44,45). This method was successful in

the deafness of animal models, in which delivery of exogenous NT3 and/or BDNF was shown efficient in promoting SGN survival and re-growing the peripheral processes in vivo (42,46–48), even when started sometime after the deafness begins (49). Although we now know the ability of neurotrophins in protecting the SGN population, yet, understanding of their impact on SGN function is little. To check the benefit of exogenous neurotrophin delivery in providing an efficient therapeutic adjunct to a cochlear implant, it is crucial that we find out how the ion channels that regulate neuronal activity are controlled. This is important given that clinical outcome still show SGN survival alone is inadequate to secure favorable sound perception by implant recipients (50).

Auditory development processes

Development of the human auditory system comprises of four basic factors that are crucial to the process.

Genetic endowment, activity independent

The basic structures of the auditory system are the result of cell differentiation, multiplication, migration, and basic cell position. These are controlled by genetic code or genetic endowment. These process will be initiated without stimulation or facilitation from outside. Some gene expression is altered by outside stimulation and environment; but the main structure, cell locations, and other parameters are the result of the genetic code. It is possible to alter genetic processes but not to improve them. In the case of the auditory system, the shape and structure of the ears, the middle ear, the nerve tracks, the main structure of the cochlea, and the nuclei are likewise genetically coded (26).

Individual genes expression that control the development of the auditory system may be altered by exposure to conditions arising from the environment. Gene expression of any single gene can be changed without altering the DNA structure in a process termed epigenetics, and in the past few years, it is the basis for major genetic research. Gene expression alteration arises as a result of exposure to three types of environmental factors. Alteration of gene expression can either be through toxic or chemical exposure, nutritional deficiencies or excesses, and intense or constant abnormal sensory stimulation.

Endogenous stimulation-dependent

Endogenous stimulation is nerve cell activity emanating from the brain, peripheral nerves or sensory organs, without stimulation from outside. The spontaneous irregular firing of ganglion cells of the spiral and cochlear nucleus is the first stage of this endogenous activity. This is necessary to promote the growth of axons for cell-to-cell interactions. In human, this starts before 20th week gestation. The irregular firing becomes regular; and with further development at 22 weeks, they become synchronous waves of ganglion cell firing. This is crucial for axons and midbrain nuclei targeting. They continue to the cerebral cortex temporal lobe by 28 to 29 weeks' gestation. These endogenous stimuli can be easily blocked by alcohol, drugs, and toxic chemicals in the environment. The effect of loud sounds or intense noise on the endogenous ganglion cell activity is not understood (26).

Exogenous or activity-dependent processes

The auditory system needs auditory stimulation as a part of development during the last 10 to 12 weeks of fetal life (28-40 weeks' gestational age) and continuing for several years after birth, unlike vision where visual experiences and stimulation are not needed until after birth at term. Starting at 28 to 29 weeks, the hair cells and their cochlea connections sufficiently mature to start tuning for specific sound frequencies. The hair cells for the lower-frequency sounds are tuned first. The fetus is protected from most high-frequency sounds in utero. The internal in utero environment is sufficiently quiet to permit the recognition and response to sounds, internal and external. Exposure to outside intense low-frequency noise (70–80 dB) will block the ability to tune the hair cells to the very specific prime frequency in utero or in the NICU.

Effects of environment and sensory interference

Environmental factors have a clear effect on the auditory development of the fetus in utero and the infant in a NICU, at home, as well day care. In Utero, all intense (N60 dB) low-frequency noise should be avoided and particularly after 20 or 22 weeks' of gestation. Fetus in utero, after 28 to 29 weeks, needs exposure to family voices, mother's voice, music (simple melodies), and family and environment meaningful sounds. The background noise level needs to be kept to less than 50 dB, particularly in the lower frequencies, for the infant to separate the music or speech.

Human amniotic fluid mesenchymal stem cells

Human amniotic fluid mesenchymal stem cells (hAFMSCs) have drawn an increasing attention recently as a potential reserve of stem cells, which can be useful for regenerative medicine clinical application. Several types of research have been carried out to date in terms of possibility for the differentiation of these cells, with several reports showing that, cells from the amniotic fluid high plasticity (51). Cells from the amniotic fluid possess immunomodulatory property both *in vivo* and *in vitro*, which could make them beneficial in an allotransplantation setting. In regenerative medicine, stem cells depict a useful tool for maintaining or regenerating the functions of defective and damaged organs and tissues (51). Stem cells are typically classified according to their ability to differentiate toward different cell types, these cells are proposed to be an important source for spiral ganglion nerves cells regeneration following hair cell loss.

Can hAFMSCS be used to the regeneration of spiral ganglion?

A substitution approach using cell therapy has been used as a treatment for severe neurological disorders such as Parkinson disease (52). Applying a similar strategy to the impaired inner ear raises many practical questions. Hearing impairment in most cases are as a

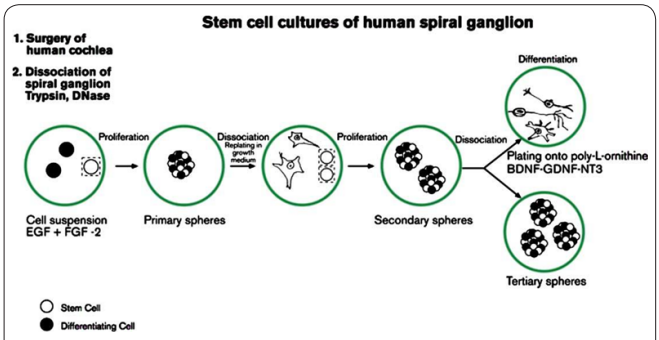

Figure 5. Human spiral ganglion was dissected during surgery, cells were isolated and then cultured. In other to induce differentiation, cells were plated on pre-coated dishes and cultured in the presence of neurotrophic factors (Adapted from (75)).

result of death, or dysfunction of spiral ganglion neuron. We propose the replacement of cells from human amniotic fluid mesenchymal stem cells replacing spiral ganglion neurons after impairment (Fig 5).

If a transplantation approach is to be successful in treating inner ear injuries, it is, of course, essential that the cells not only survive, but also migrate to functionally relevant regions and differentiate into an appropriate cell fate, that is, a neuronal fate when attempting to replace auditory neurons.

We hypothesize that once the cells of this hAFMSCs line migrate within the cochlea, they receive signals from the microenvironment and will upregulate genetic cell fate programs expressed by local endogenous cells.

SGNs degenerate after hair cell loss in both humans (53–55), and animal models of sensory neural hearing loss (42,48,56,57). This is caused by the loss of the endogenous supply of the pro-survival neurotrophin (NT) peptides brain-derived neurotrophic factor (BDNF) and neurotrophin-3 (NT- 3), produced by the inner hair cells and support cells of the organ of Corti (58–61). Acute SGN degeneration may limit the efficacy of hearing rehabilitation by a cochlear implant. The prevention of SGN degeneration following an SNHL may, therefore, promote the clinical consequence for implant patients.

Conclusion

Our ear is one of the vital organ, in which a loss of its function may lead to a severe consequence, medically, economically and socially(62–66). hAFMSCs having great pluripotent potentials can be taken advantage of by using it for the regeneration of spiral ganglion which is one of the most vital part of the inner ear(67–71). We propose the regeneration potentials of spiral ganglion can meet up with this demand. The full mechanism by which this process occur should be studied in the future as it may be the best and last resort for patient with SNHL(72,73).

Acknowledgments
The authors thank Department of Medical Biotechnology, School of advance Science in Medicine, Tehran University of Medical Sciences and Department of Medical Biotechnology, Faculty of Advance Medical Sciences, Tabriz University of Medical Sciences.

References

1. Holley MC. Application of new biological approaches to stimulate sensory repair and protection. Br Med Bull. 2002;63:157–69.
2. Kanzaki S, Kawamoto K, Oh SH, Stover T, Suzuki M, Ishimoto S, et al. From gene identification to gene therapy. Audiol Neurootol. 2002;7(3):161–4.
3. Coskunpinar E, Arkan H, Dedeoglu BG, Aksoz I, Polat E, Araz T, et al. Determination of effective miRNAs in wound healing in an experimental Rat Model. Cell Mol Biol (Noisy-le-grand). 2015;61(8):89.
4. Davies J, Davies D. Origins and evolution of antibiotic resistance. Microbiol Mol Biol Rev. 2010;74(3):417–33.
5. Lee YH, Bae SC. Association between functional CD24 polymorphisms and susceptibility to autoimmune diseases: A meta-analysis. Cell Mol Biol (Noisy-le-grand). 2014;61(8):97–104.
6. Wu X, Zhang J, Huang Q, Yang P, Chen J, Liu J. Role of Kruppel-like Factor 2 in Intracranial Aneurysm of the Rabbit Model. Cell Mol Biol (Noisy-le-grand). 2014;61(7):33–9.
7. Zheng L, Yang L, Wang Z, Chen C, Su Y. Protective effect of Esculin in adjuvant-induced arthritic (AIA) rats via attenuating pro-inflammatory cytokines and oxidative stress. Cell Mol Biol (Noisy-le-grand). 2014;61(7):1–5.
8. Shou J, Zheng JL, Gao WQ. Robust generation of new hair cells in the mature mammalian inner ear by adenoviral expression of Hath1. Mol Cell Neurosci. 2003;23(2):169–79.
9. Kawamoto K, Ishimoto S-I, Minoda R, Brough DE, Raphael Y. Math1 gene transfer generates new cochlear hair cells in mature guinea pigs in vivo. J Neurosci. 2003;23(11):4395–400.
10. Löwenheim H, Furness DN, Kil J, Zinn C, Gültig K, Fero ML, et al. Gene disruption of p27(Kip1) allows cell proliferation in the postnatal and adult organ of corti. Proc Natl Acad Sci U S A. 1999;96(7):4084–8.
11. Zheng JL, Gao WQ. Overexpression of Math1 induces robust production of extra hair cells in postnatal rat inner ears. Nat Neurosci. 2000;3(6):580–6.
12. Raphael Y. Cochlear pathology, sensory cell death and regeneration. Br Med Bull. 2002;63:25–38.
13. Huang Y, Sun Y, Wang Q. Encapsulation and in vitro release of erythromycin using biopolymer micelle. Cell Mol Biol (Noisy-le-grand). 2014;61(7):60–4.
14. Yao H, Han J, Wang J, Wang L, Gong C, Li L, et al. Amplification of rabbit hepatocyte growth factor and detection of its expression in COS-7 cell line. Cell Mol Biol (Noisy-le-grand). 2014;61(7):65–9.
15. Song W, Song C, Chen Y, Du M, Hu P, Liu A, et al. Polysaccharide-induced apoptosis in H22 cells through G2/M arrest and BCL2/BAX caspase-activated Fas pathway. Cell Mol Biol (Noisy-le-grand). 2014;61(7):88–95.
16. Luo Y, Li Q, Wang X, Yang F, Nong S, Zhu D. Molecular Characterization and TRAP Analysis of Gene in Dendranthema morifolium. Cell Mol Biol (Noisy-le-grand). 2014;61(7):119–22.
17. Wang Z, Chen J, Capobianco AJ. The Notch signaling pathway in esophageal adenocarcinoma. Cell Mol Biol (Noisy-le-grand). 2014;61(6):24–32.
18. Hsu YC, Hsieh YH, Liao CC, Chong LW, Lee CY, Yu YL, et al. Targeting post-translational modifications of histones for cancer therapy. Cell Mol Biol (Noisy-le-grand). 2014;61(6):69–84.
19. Coskunpinar E, Yildiz P, Aynaci E, Turna A, Oltulu YM, Hekimoglu E, et al. Investigation of some DNA repair genes association in non small cell lung cancer. Cell Mol Biol (Noisy-le-grand). 2015;61(8):57.
20. Burdon T, Smith A, Savatier P. Signalling, cell cycle and pluripotency in embryonic stem cells. Trends Cell Biol. 2002;12(9):432–8.

21. Henningson CT, Stanislaus MA, Gewirtz AM. 28. Embryonic and adult stem cell therapy. J Allergy Clin Immunol. 2003;111(2 Suppl):S745-53.
22. Graven SN, Browne J V. Auditory Development in the Fetus and Infant. Newborn Infant Nurs Rev. 2008;8(4):187–93.
23. Kandel ER, Schwartz JH, Jessell TM. Principles of Neural Science. Vol. 4, Neurology. 2000. 1414 p.
24. Hu Z, Wei D, Johansson CB, Holmström N, Duan M, Frisén J, et al. Survival and neural differentiation of adult neural stem cells transplanted into the mature inner ear. Exp Cell Res. 2005;302(1):40–7.
25. Pujol R, Lavigne-Rebillard M. Development of neurosensory structures in the human cochlea. Acta Otolaryngol. 1992;112(2):259–64.
26. Hall JW. Development of the ear and hearing. J Perinatol. 2000;20(8 Pt 2):S12–20.
27. Morris BH, Philbin MK, Bose C. Physiological effects of sound on the newborn. J Perinatol. 2000;20(8 Pt 2):S55–60.
28. D'Andrea P, Veronesi V, Bicego M, Melchionda S, Zelante L, Di Iorio E, et al. Hearing loss: Frequency and functional studies of the most common connexin26 alleles. Biochem Biophys Res Commun. 2002;296(3):685–91.
29. Trautwein PG, Sininger YS, Nelson R. Cochlear implantation of auditory neuropathy. J Am Acad Audiol. 2000;11(6):309–15.
30. Glastonbury CM, Davidson HC, Harnsberger HR, Butler J, Kertesz TR, Shelton C. Imaging findings of cochlear nerve deficiency. Am J Neuroradiol. 2002;23(4):635–43.
31. Holley MC. Application of new biological approaches to stimulate sensory repair and protection. Vol. 63, British Medical Bulletin. 2002. p. 157–69.
32. Izumikawa M, Minoda R, Kawamoto K, Abrashkin K a, Swiderski DL, Dolan DF, et al. Auditory hair cell replacement and hearing improvement by Atoh1 gene therapy in deaf mammals. Nat Med. 2005;11(3):271–6.
33. Parker MA, Cotanche DA. The Potential Use of Stem Cells for Cochlear Repair. Vol. 9, Audiology and Neuro-Otology. 2004. p. 72–80.
34. Snyder EY, Yoon C, Flax JD, Macklis JD. Multipotent neural precursors can differentiate toward replacement of neurons undergoing targeted apoptotic degeneration in adult mouse neocortex. Proc Natl Acad Sci U S A. 1997;94(21):11663–8.
35. Yandava BD, Billinghurst LL, Snyder EY. "Global" cell replacement is feasible via neural stem cell transplantation: evidence from the dysmyelinated shiverer mouse brain. Proc Natl Acad Sci U S A. 1999;96(12):7029–34.
36. Timothy Himes B, Liu Y, Solowska JM, Snyder EY, Fischer I, Tessler A. Transplants of cells genetically modified to express neurotrophin-3 rescue axotomized Clarke's nucleus neurons after spinal cord hemisection in adult rats. J Neurosci Res. 2001;65(6):549–64.
37. Teng YD, Lavik EB, Qu X, Park KI, Ourednik J, Zurakowski D, et al. Functional recovery following traumatic spinal cord injury mediated by a unique polymer scaffold seeded with neural stem cells. Proc Natl Acad Sci U S A. 2002;99(5):3024–9.
38. Mi R, Luo Y, Cai J, Limke TL, Rao MS, H??ke A. Immortalized neural stem cells differ from nonimmortalized cortical neurospheres and cerebellar granule cell progenitors. Exp Neurol. 2005;194(2):301–19.
39. Parker MA, Anderson JK, Corliss DA, Abraria VE, Sidman RL, Kook IP, et al. Expression profile of an operationally-defined neural stem cell clone. Exp Neurol. 2005;194(2):320–32.
40. Abrams RM, Gerhardt KJ. The acoustic environment and physiological responses of the fetus. J Perinatol. 2000;20(8 Pt 2):S31-36.
41. Li H, Liu H, Heller S. Pluripotent stem cells from the adult mouse inner ear. Nat Med. 2003;9(10):1293–9.
42. Gillespie LN, Clark GM, Bartlett PF, Marzella PL. BDNF-induced survival of auditory neurons in vivo: Cessation of treatment leads to accelerated loss of survival effects. J Neurosci Res. 2003;71(6):785–90.
43. McGuinness SL, Shepherd RK. Exogenous BDNF rescues rat spiral ganglion neurons in vivo. Otol Neurotol. 2005;26(5):1064–72.
44. Pettingill LN, Richardson RT, Wise a. K, O'Leary SJ, Shepherd RK. Neurotrophic Factors and Neural Prostheses: Potential Clinical Applications Based Upon Findings in the Auditory System. IEEE Trans Biomed Eng. 2007;54(6):1138–48.
45. Roehm PC, Hansen MR. Strategies to preserve or regenerate spiral ganglion neurons. Curr Opin Otolaryngol Head Neck Surg. 2005;13(5):294–300.
46. Miller JM, Chi DH, O'Keeffe LJ, Kruszka P, Raphael Y, Altschuler RA. Neurotrophins can enhance spiral ganglion cell survival after inner hair cell loss. Int J Dev Neurosci. 1997;15(4–5):631–43.
47. Staecker H, Kopke R, Malgrange B, Lefebvre P, Van de Water TR. NT-3 and/or BDNF therapy prevents loss of auditory neurons following loss of hair cells. Neuroreport. 1996;7(4):889–94.
48. Wise AK, Richardson R, Hardman J, Clark G, O'leary S. Resprouting and survival of guinea pig cochlear neurons in response to the administration of the neurotrophins brain-derived neurotrophic factor and neurotrophin-3. J Comp Neurol. 2005;487(2):147–65.
49. Gillespie LN, Clark GM, Marzella PL. Delayed neurotrophin treatment supports auditory neuron survival in deaf guinea pigs. Neuroreport. 2004;15(7):1121–5.
50. Fayad JN, Linthicum FH. Multichannel cochlear implants: relation of histopathology to performance. Laryngoscope. 2006;116(8):1310–20.
51. Parolini O, Soncini M, Evangelista M, Schmidt D. Amniotic membrane and amniotic fluid-derived cells: potential tools for regenerative medicine? Regen Med. 2009;4(2):275–91.
52. Dunnett SB, Björklund a, Lindvall O. Cell therapy in Parkinson's disease - stop or go? Nat Rev Neurosci. 2001;2(5):365–9.
53. Miura M, Sando I, Hirsch BE, Orita Y. Analysis of spiral ganglion cell populations in children with normal and pathological ears. Ann Otol Rhinol Laryngol. 2002;111(12):1059–65.
54. Nadol JB, Young YS, Glynn RJ. Survival of spiral ganglion cells in profound sensorineural hearing loss: Implications for cochlear implantation. Ann Otol Rhinol Laryngol. 1989;98(6):411–6.
55. Zimmermann CE, Burgess BJ, Nadol Jr JB. Patterns of degeneration in the human cochlear nerve. Hear Res. 1995;90(1–2):192–201.
56. Spoendlin H. Factors inducing retrograde degeneration of the cochlear nerve . PubMed Commons. Ann Otol Rhinol Laryngol Suppl 1984 Jul-Aug;11276-82. 1984;6431887.
57. Leake P a, Hradek GT. Cochlear pathology of long term neomycin induced deafness in cats. Hear Res. 1988;33(1):11–33.
58. Fritzsch B, Tessarollo L, Coppola E, Reichardt LF. Neurotrophins in the ear: their roles in sensory neuron survival and fiber guidance. Prog Brain Res. 2004;146:265–78.
59. Stankovic K, Rio C, Xia A, Sugawara M, Adams JC, Liberman MC, et al. Survival of adult spiral ganglion neurons requires erbB receptor signaling in the inner ear. J Neurosci. 2004;24(40):8651–61.
60. Tan J, Shepherd RK. Aminoglycoside-induced degeneration of adult spiral ganglion neurons involves differential modulation of tyrosine kinase B and p75 neurotrophin receptor signaling. Am J Pathol. 2006;169(2):528–43.
61. Ylikoski J, Pirvola U, Moshnyakov M, Palgi J, Arumäe U, Saarma M. Expression patterns of neurotrophin and their receptor mRNAs in the rat inner ear. Hear Res. 1993;65(1–2):69–78.
62. Aiyelabegan HT, Zaidi SSZ, Fanuel S, Eatemadi A, Ebadi MTK, Sadroddiny E. Albumin-Based Biomaterial for Lungs Tis-

sue Engineering Applications. Int J Polym Mater Polym Biomater. 2016;(just-accepted).
63. Beiranvand S, Eatemadi A, Karimi A. New Updates Pertaining to Drug Delivery of Local Anesthetics in Particular Bupivacaine Using Lipid Nanoparticles. Nanoscale Res Lett. 2016;11(1):1–10.
64. Daraee H, Eatemadi A, Abbasi E, Fekri Aval S, Kouhi M, Akbarzadeh A. Application of gold nanoparticles in biomedical and drug delivery. Artif cells, nanomedicine, Biotechnol. 2016;44(1):410–22.
65. Daraee H, Etemadi A, Kouhi M, Alimirzalu S, Akbarzadeh A. Application of liposomes in medicine and drug delivery. Artif cells, nanomedicine, Biotechnol. 2016;44(1):381–91.
66. Eatemadi A, Darabi M, Afraidooni L, Zarghami N, Daraee H, Eskandari L, et al. Comparison, synthesis and evaluation of anticancer drug-loaded polymeric nanoparticles on breast cancer cell lines. Artif cells, nanomedicine, Biotechnol. 2016;44(3):1008–17.
67. Eatemadi A, Daraee H, Karimkhanloo H, Kouhi M, Zarghami N, Akbarzadeh A, et al. Carbon nanotubes: properties, synthesis, purification, and medical applications. Nanoscale Res Lett. 2014;9(1):1–13.
68. Eatemadi A, Daraee H, Zarghami N, Melat Yar H, Akbarzadeh A. Nanofiber: synthesis and biomedical applications. Artif cells, nanomedicine, Biotechnol. 2016;44(1):111–21.
69. Ghafarzadeh M, Eatemadi A, Fakhravar Z. Human amniotic fluid derived mesenchymal stem cells cause an anti-cancer effect on breast cancer cell line in vitro. Cell Mol Biol. 2016;2016(6):102–6.
70. Mellatyar H, Akbarzadeh A, Rahmati M, Ghalhar MG, Etemadi A, Nejati-Koshki K, et al. Comparison of inhibitory effect of 17-DMAG nanoparticles and free 17-DMAG in HSP90 gene expression in lung cancer. Asian Pac J Cancer Prev. 2014;15(20):8693–8.
71. Mohammadian F, Eatemadi A. Drug loading and delivery using nanofibers scaffolds. Artif cells, nanomedicine, Biotechnol. 2016;1–8.
72. Namdari P, Daraee H, Eatemadi A. Recent Advances in Silicon Nanowire Biosensors: Synthesis Methods, Properties, and Applications. Nanoscale Res Lett. 2016;11(1):406.
73. Seidi K, Eatemadi A, Mansoori B, Jahanban-Esfahlan R, Farajzadeh D. Nanomagnet-based detoxifying machine: an alternative/complementary approach in HIV therapy. J AIDS Clin Res. 2014;2014.
74. Kandel ER, Schwartz JH, Jessell TM. Principles of Neural Science. Vol. 3, Neurology. 2000. 1414 p.
75. Rask-Andersen H, Bostr??m M, Gerdin B, Kinnefors A, Nyberg G, Engstrand T, et al. Regeneration of human auditory nerve. In vitro/in video demonstration of neural progenitor cells in adult human and guinea pig spiral ganglion. Hear Res. 2005;203(1–2):180–91.