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Fibroblast Growth Factor-2 (Fgf-2) Role on Inner Hair Cell Regeneration and Tumor Growth with Regards to Regenerative Medicine – A Review

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Abstract: The present regenerative medicine needs more clear views on the use of various growth factors. This present study reviewed important facts on the use of Fibroblast Growth Factors-2 (Fgf-2). Fgf-2 role on regenerative medicine especially with regards to inner hair cell regeneration from different stem cell source is needs to be well understood, because its function and activity on specific cell differentiation and maintenance not fully understood. The above mitogen activity much speculated for tumor growth than angiogenesis as transforming and growth factor. Hence the present study discussed different experimental evidences to understand the role of Fgf-2 on specific cell type regeneration from different stem cell population. This is more important at present situation, since many clinical practices underway using the above with different stem cell sources on regenerative medicine.

Key words: Stem Cell, regenerative medicine, hair cells, tumor, fibroblast growth factors

INTRODUCTION

The organ of Corti composed of six distinct cell types arranged with different cellular patterns. On the above, the most interesting aspects is organ of Corti extends along the basal-to-apical axis with one row of inner hair cells and three rows of outer hair cells. The sensorineural deafness mainly caused by loss or damage to inner hair cells in many ways including infection, inflammation, accidental truma and etc., Many research works were carried out to understand the possible mechanism for regeneration of inner hair cells of mammalian origin. Many different ideas were placed for regeneration of inner hair cell from different types of stem cells, alongside understanding supplementary factors role, which has paramount importance. Fibroblast growth factor (Fgf) the much anticipated supplement for regeneration of hair cells from different stem cell sources. On the other hand many contrasting experimental results were also placed. This present work is dedicated to understand Fgf and its role on regenerative medicine to bring clear opinion on the use.

It is necessary to understand the role of Fgf on multiplication, differentiation and maintenance of specific cell types produced in *in vitro* and *in vivo* systems. Fgf is the multifunctional heparin-binding protein, characteristically mitogenic and angiogenic outside the nervous system. The distribution of Fgf and its receptors seems not for single function but argue for more complex action on the development of specific cell types both in *in vitro* and *in vivo*. The regulation of endogenous Fgf and its 1-3 different receptors during development and regeneration indicates different functions for these molecules (Baird, 1994). In normal tissue, Fgf-2 are present in basement membranes and in the sub-endothelial extracellular matrix of blood vessels. It stays membrane-bound as long as there is no signal peptide. During both wound healing process or tumor development, the heparan sulfate-degrading enzymes activates Fgf-2 and mediates angiogenesis. Recent evidence shown that, low level of Fgf-2 plays key role on the incidence of excessive anxiety and hair cell regeneration. In addition, Fgf-2 is the critical component for human embryonic stem cell culture medium as noted necessary for cells to remain undifferentiated but by which it does is poorly defined (Pereira *et al*., 2000). This study reviewed some experimental evidences produced using Fgf-2 on regenerative medicine and tumor growth studies.

DISCUSSION

Hair cell regeneration: Over the past few decades, it has been well demonstrated that, lower vertebrates and avian species has enormous capacity to regenerate new inner hair cells (Oesterle *et al*., 2000). Alongside certain degree of hair cell regeneration and repair also reported with mammalian inner ear structures. The importance of Fgf role on regeneration of inner hair cells from different stem cell sources has paramount importance when it comes with clinical application. Besides when it comes for clinical application, the regeneration and maintenance of such neural cell types is often more difficult. It is also noted from earlier studies, the implants are no long lasting. Hence current focus is on producing long lasting or permanent inner hair cells for clinical application. On this context many research considering use of Fgf. The Fgf-2 is the most potent mitogen when added with Insulin like growth factor-1 (IGF-1) in *in vitro*. This could be the

most potent physiological growth factor during regeneration of new hair cells on noise induced damage study (Lisa *et al*., 1997). In noise damaged avian cochlear epithelium resulted with up regulation of mRNA for Fgf receptor in the supporting cells and proliferation. This implicate to direct action on the inner ear supporting cells and induced proliferation (Lee and Cotanche, 1996; Corwin *et al*., 1995). Considered together, these experiments suggest that, Fgf-2 may be the candidate molecule regulating proliferation of the inner ear supporting cells. Similarly co-administration of Fgf-2 and Glial cell line-derived neurotrophic factor (Gdnf) provided long-term survival and Fgf-2 powerful action on promoting neurite regeneration (Wei *et al*., 2007). In addition, Fgf-2 promotes the proliferation of embryonic stem cells to produce inner ear precursors (Li *et al*., 2003a, b; Hu *et al*., 2005). This is in accordance with earlier observations implicating upon fgf-2 role on proliferation, differentiation and survival of developing inner hair cells (Leon *et al*., 1995; Zheng *et al*., 1997; Hossain and Morest, 2000; Varela-Nieto *et al*., 2004; Ladher *et al*., 2005).

Alternatively, exogenously added Fgf-2 may not elicit proliferation in the intact mature utricles (Yamashita and Oesterle, 1995). This finding suggests that, Fgf-2 may have additive mitogenic effect and infers several other growth factors work in concert during regeneration of hair cells. For example, Fgf-2 and Tgf-b1 shown to work synergistically to regulate chondrogenesis during otic capsule formation (Gao and Macagno, 1988; Frenz *et al*., 1994). Hence it is quite clear that multiple growth factors contribute together for differentiation or regeneration of new hair cells. These factors works either in a sequential manner or at multiple cascading events. In support of this notion Kopke *et al*., (1996) study showed combination of Tgf, Igf-1, and retinoic acid facilitated new hair cell regeneration.

Autocrine or paracrine action of Fgf is another important phenomena on the new hair cell regeneration. Fgf-2 may exert its action via autocrine mechanism by providing their tropic support. Alternatively, paracrine action also postulated: Fgf-2 synthesized by hair cells could locally influence maintenance of neighboring hair cells and proliferation of supporting cells. Generally, degeneration of hair cells lead to release Fgf-2, which would, in turn, stimulate supporting cell proliferation. Besides Fgf-2 not having signal sequence and cell injury may be pawing the major way for mitogen release. Taken together autocrine or paracrine mode of action by Fgf is the most important on the proliferation and maintenance of the specific cell types *in vivo* or *in vitro*. Transdifferentiation and maintenance is the most intriguing aspect in regenerative medicine. Because transdifferentiation requires specific mitogenic action and along the autocrine or paracrine effect of the specific cell type.

Cancer: Growing body of evidences from many basic, pre-clinical and clinical research works in regenerative medicine promoted curiosity among the researchers and clinical practitioners on avoiding uncertainty over regenerative medicine therapy. In accord, cytokines used for regenerative medicine possess proinflammatory, pro-angiogenic, and immuno-regulatory activities and they are consistent with pathological alterations in neoplasm and play important role on promoting tumorogenesis. From many observations, the use of different supplements for regenerative medicine needs to be clearly defined especially growth and transforming factors. Those growth and transforming factors are the key on trans-differentiation and maintenence of the specific cell type. Fgf-2 is closely involved in angiogenesis and tumor growth of various cancers. Head and Neck Squamous Cell Carcinoma (HNSCC) cells taken from patient showed Fgf-2 the most commonly detected cytokine and implicated to tumor growth (Zhong *et al*., 1999). Fgf-2 and its receptor Fgfr-1 are implicated for tumor growth, as noted, the interplay between Fgf-2 and Platelet-derived growth factor-B (Pdgf-b) and Fgf-2 and vascular endothelial growth factor-3 (Vegfr-3) is most important cascading events in the initiation and maintenance of tumor. The study involved understanding the impact of Fgf-2 and Fgfr-1 on tumor cells and co-expression with Vegfr-3 or Pdgf-b demonstrated that Fgf-2 expression in tumor cells is a independent negative prognostic factor, and the co-expressions of Fgf-2 and Vegfr-3 and Fgfr-1 and Pdgf-b are strongly implicated for poor survival of tumor patients (Donnem *et al*., 2009). Likewise, over expression of Fgfr-1 correlated to liver metastasis and suggested that, the over expression of Fgfr-1 gene action may lead to liver metastasis in colorectal cancer. Further it is been assumed that Fgfr-1 gene may be the useful predictor of liver metastasis in patients with colorectal cancer (Lefevre *et al*., 2009; Sato *et al*., 2009). Similar study on the identification of patterns of protein expression of Fgf-2 and its receptors on lung carcinoma and their role in the early pathogenesis of Squamous Cell Carcinoma (SCC) showed high and frequent expression of Fgf-2, Fgfr1 on SCC and adeno-carcinoma of the lung tumor (Behrens *et al*., 2008). Study investigated over-expression of Fgf-2 isoforms on rat glioma cell line using tetracycline-regulated expression system demonstrates that, Fgf-2 has unique features in inhibiting glioma cell proliferation and cell-cycle arrest at the G2M and control over mitosis. This indicates that, Fgf-2 inhibits tumor growth in glioma cells by acting on cell-cycle progression (Lemiere *et al*., 2008). In addition PCR mRNA expression study for Igf-1, and Fgf-2 on human brain tumor revealed positive correlation between autocrine expression of Igf-1 and Fgf-2. This known to be involved with the progression of tumor and suggest mRNA of the above genes in the early stages of disease could be useful for prognostic purposes, and these genes can be considered as

potential targets for therapeutic approaches against brain tumors (Ru *et al*., 2008; Baritaki *et al*., 2009). Furthermore Fgf-2 and angiogenin enzymes direct contribution to HeLa cell proliferation more appraised its role on tumor growth (Yang *et al*., 2009).

Similarly many studies on the role of Fgf-2 importance on tumor growth brought stronger association between those above. A study on tumor-associated macrophage role on angiogenesis and tumor progression demonstrated that, rat prostate tumor cells injected to prostate gland of immuno-competent Copenhagen rats showed with increased endothelial proliferation and tumor. When the tumors examined by PCR arrays showed many factors promoting monocytes recruitment, angiogenesis, and tissue remodeling including chemokine ligand 2, Fgf- 2, matrix metalloproteinase 9, interleukin 1beta, interferon gamma, and transforming growth factor beta were found highly up regulated (Halin *et al*., 2009). Tumor associated macrophages induced cyclooxygenase-2-dependent secretion of Fgf-2, Vegf-A and increased angiogenesis in human basal cell carcinoma is reported (Tjiu *et al*., 2009). Similarly neuroblastoma cells transplanted on Matrigel (gelatinous protein mixture secreted by mouse tumor cells and used as a substrate for cell culture) showed Vegf-A, Fgf-2, angiopoietin-1 (ANG-1), hypoxia inducible factor-2alpha (Hif-2alpha) appearance after 4 days and thus implicated to angiogenic activity of neuroblastoma cells (Mangieri *et al*., 2009). Recently, Chikazawa *et al*. (2008) and Li *et al*. (2009) were demonstrated the relation between Fgf-2 expression play important role on tumor growth angiogenesis. Subsequently, study demonstrated decreased serum level of angiogenic cytokines after radiotherapy in patients with cancer has predictive value for tumor growth and Fgf-2 like cytokines (Ria *et al*., 2008).

CONCLUSION

In recent times observation procured from basic, pre-clinical and clinical research on regenerative medicine strongly advocated researchers to look for avoiding uncertainty over regenerative medicine therapy using stem cells and supplements. Cytokines, which are used for regenerative medicine, possess proinflammatory, pro-angiogenic, and immuno-regulatory activities and they are consistent with pathological alterations in neoplasm and play important role behind trans-differentiation of specific cell type and promoting tumorogenesis. The use of different supplements for regenerative medicine needs to be clearly defined for their role on transdifferentiation of specific cell type, especially using growth and transforming factors. Those growth and transforming factors are the key on trans-differentiation and maintenence of the specific cell type. But the uncertainty is plausible over the use of supplements like growth and differentiation factors for regenerative medicine. The use of Fgf-2 on the above is the one much speculated for tumor growth, since the use of the aove growth factor is more frequent in the regenerative medicine theraphy. Hence their role noted on both transdifferentiation of inner hair cells from different stem cell sources and various tumor growth creating uncertainty over the use of Fgf-2 on clinical application. The exact role of Fgf-2 alone or with other factors in *in vivo* or with *in vitro* may differ from one another. Since, similar situation of defined environment cannot be produced in *in vivo*, hence the possibility of contribution of other factors available with *in vivo* or natural environment may take the application into different route and will also create undesirable effect. Hence the use of Fgf-2 needs to be clearly defined for a particular situation along with other factors available in *in vivo* and supplemented along with co-culture system.

ABBREVIATIONS

Fgf-2 = Fibroblast growth factors-2
Fgfr-1 = Fibroblast growth factor receptor-1
Igf-1 = Insulin like growth factor-1
Gdnf = Glial cell line-derived neurotrophic factor
Tgf-b1 = Tissue growth factor-b
HNSCC = Head and Neck Squamous Cell Carcinoma
Pdgf-b = Platelet-drived growth factor-b
Vegfr-3 = Vascular endothelial growth factor-3
SCC = Squamous Cell Carcinoma
Vegf-A = Vascular endothelial growth factor-A
Hif-2alpha = hypoxia inducible factor-2alpha

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