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RESEARCH ARTICLE
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Assessment Of Effects of Manipulation of Stem Cell Conditioning Media with Ethyl Gallate in Regenerative Strategies

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ABSTRACT

Introduction: In multicellular organisms , stem cells are undifferentiated or partially differentiated cells that can differentiate into various types of cells and proliferate indefinitely to produce more of the same stem cell . stem cell growth requires conditional media. The conditional media refers to the collection of proteins that contains a signal peptide and are processed via the endoplasmic reticulum and golgi apparatus through the classical secretion pathway.

Materials And Methods: Culture the fibroblasts stem cells using ethyl gallate stem cell conditioning medium. Incubated the stem cell at regular time intervals. The culture stem cell were assessed using scratch test assay.

Result: There was increased proliferation of stem cell in ethyl gallate stem cell conditioned media.

Conclusion: Ethyl gallate is a useful stem cell conditioning substance to increase stem cell growth.

Keywords: *stem cell, conditioning media, ethyl gallate, proliferation*

INTRODUCTION

Stem cells are bodies raw material cells from which all other cells with specialised functions are generated. Stem cells are the earliest type of cells in a cell lineage; they are found in both embryonic and adult organisms but they have very different properties in each. Stem cell hold great promise for cell therapy, tissue engineering and regenerative medicine as well as pharmaceutical and biotechnological application(Micromechanical Cues Converging on Fi...) Stem cell has many useful properties such as self renewal and potency.Stem cell use telomerase ,a protein that restore telemorse to protect their DNA and extend their cell division limit. It also has the ability to be totipotent ,pluripotent,multipotent, oligopotent and unipotent(Maeda et al. 2011).It is commonly known that stem cells are essential for replacing or healing damaged or ill tissues, however there are three things to take into account before using stem cells in clinical settings.

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The first is the non-intrusive, simple, and efficient way to harvest, handle, and multiply them. Accordingly, adipose derived stem cells are thought to be the easiest to work with among mesenchymal stem cells; their pluripotency, proliferative effectiveness, and low donor morbidity have all been adequately validated since they were originally reported in 2001. The ability to develop stem cells into the necessary cells and utilise them efficiently to create three-dimensional tissues is the second factor; in this case, tissue-specific scaffolds and signalling systems are crucial.(Ogawa 2006)An important achievement in regenerative medicine has been the identification and control of stem cells, which has also aided in the creation of clinical treatments based on tissue engineering. The substantial soft and hard tissue loss brought on by periodontitis requires the development of novel approaches, such as tissue engineering, due to the challenges associated with attaining predictable periodontal regeneration.(Maeda et al. 2011)

Ethyl gallate is a gallate ester obtained by the formal condition of gallic acid with ethanol. It is a plant metabolite and it is added to food as an antioxidant. The ethyl gallate was identified as the major constituent extracted from the roots of euphorbia fischeriana steud. It is also proved that ethyl gallate suppresses proliferation and invasion in human breast cancer cell tunes(Hu et al. 2021; Cui et al. 2015) the use of conditioned media divide of cells poses several benefits compared with the use of the cells (micler AM et al,2016)

Pluripotent stem cells (PSC) can differentiate into virtually any cell type in the body, making them attractive for both regenerative medicine and drug discovery.Pluripotent stem cells may be useful for treating wide variety of disease given their ability to differentiate, theoretically into every cell type in the body.(Micromechanical Cues Converging on Fi...; Dixon 2020) various studies on stem cell - denviolea conditioned media from various cell lines such as adipose derived stem cell and bone marrow.

Use of ethyl gallate as an additive to the condition media should increase the growth of stem cells culture.Traditionally, stem cells are grown as a single layer in two dimensional plastic culture plates of a required unfind are xenogeneic materials (Rasul chaudry et al,2017) since ethyl gallate has anti humarogenic property it will prevent the stem cell to turn into a cancer cell.

MATERIALS AND METHODS

The fibroblast cells were seeded in six well plates (density 1*106 cells/well) after being cultured and followed by 24hrs,48hrs&72hrs induction with dexamethasone respectively. After 48hrs cells were fixed with 4% pfa and stained with h&e vice versa.

The cells were stained with calce ,AM dye was added in the culture medium and incubated for 30 mins and washed with 1*pbs.Further,the cells were absorbed using inverted phase contrast fluorescence microscopy (evos-in vitrogen,USA

RESULTS

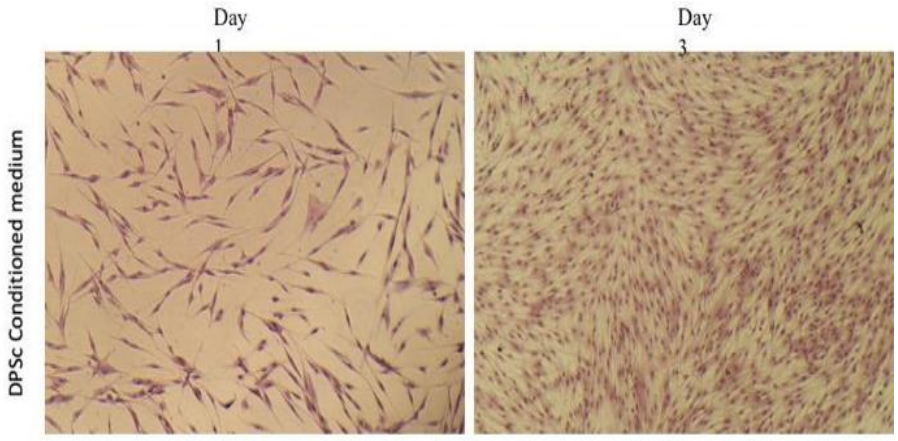

FIGURE 1: Represents the cell proliferation rate by DPC's conditioned medium on human gingival fibroblast cells.

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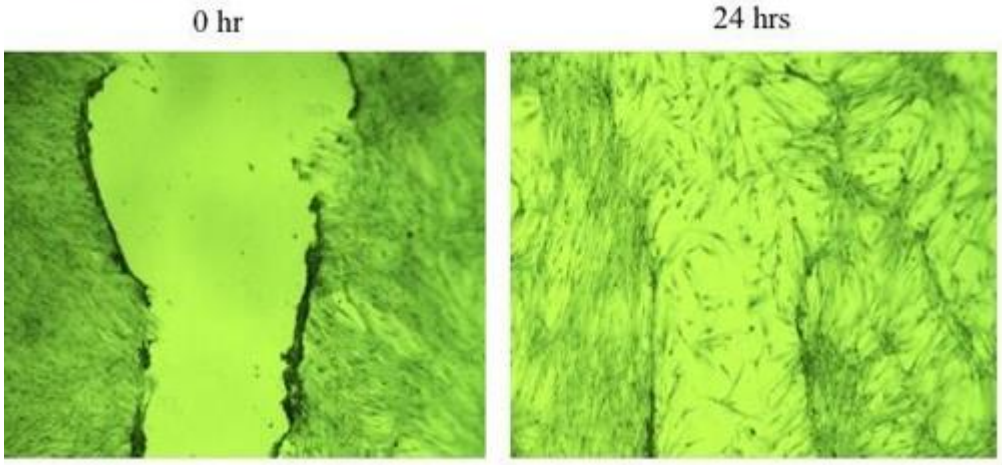

FIGURE 2: Representative images from in vitro scratch wound healing assay demonstrating that cell migration is significantly accelerated in the presence of conditioned media of 24hrsWhen compare To scale 100M Images were obtained A inverted phase Contrast microscope.

DISCUSSION

There are several ways to improve stem cell proliferation.A healthy conditioning medium plays a major role in proliferation of stem cell.Altering the condition media will alter the growth of the stem cells.The stem cells also sense the physical constants of their microenvironment under appropriate conditions. They are able to self renew indefinitely(Manipal et al. 2014)

The ability of stem cells to self-renew and produce various cell types opens up new possibilities for the regeneration of damaged tissues and the treatment of illness. Adult mesenchymal stem/stromal cells (MSCs) have been discovered in a number of oral and maxillofacial tissues in the field of dentistry.(Zheng and Cheung 2012) This indicates that the oral tissues are a rich source of stem cells, and oral stem and mucosal cells are anticipated to provide an ideal source for genetically reprogrammed cells like induced pluripotent stem (iPS) cells. Furthermore, as stem cell and tissue engineering therapies in dentistry continue to garner growing clinical attention, oral tissues are anticipated to be both a source and a therapeutic target for stem cells.(Paz et al. 2018)

Traditional dental procedures call for the employment of specialised, to the tissue-adapted materials, which have questionable durability and efficacy. Since they can guarantee physiologically superior structural and functional outcomes, stem cell-based treatment techniques could present an alluring alternative in dentistry.(Shruthi 2012; Mitsiadis et al. 2015) These treatments require enough of these particular stem cell types to be implanted. Dental mesenchymal stem cells are simple to isolate and can grow in vitro while still being stem cells. Dental mesenchymal stem cells have the potential to support pulp and periodontal regeneration, as shown by in vivo research carried out on small and large animals, but they also present new, significant problem(Shruthi 2012)

Through this study, it is revealed that culture medium influence cell fate and acts not only as a.Feeder, but also a instructor.Culture of stem cell includes a lot of complications.Inclusive high infection NSK,Stem cell forming a tumour is also highly possible once It is used in the body.Use of ethyl gallate as the conditioning media produces the NSK of stem cell transforming into a Tumor.One hallmark of embryonic stem cell is that they cause particular type of tumour called teratoma(Malhotra 2016).

Ethyl Gallate Inhibits proliferation Indicates breast cancer cells in a dose and time dependent.(Cui et al. 2015)

In this study 2 petri dish samples of stem cells were maintained. One petri dish served as a control and the petri dish contained ethyl gallate conditioning medium.It was clearly proved that the petri dish containing ethyl gallate proliferated more compared to the control.(Ethyl Gallate 831-61-8 2004) This idea was further assessed using images from invitro scratch wound healing

assay.The assay showed the migration of stem cell and increase in number of stem cells was also clearly noted.Cell migration can be measured in vitro using the simple, affordable, and well-established in vitro scratch assay. The fundamental procedure entails making a "scratch" in a monolayer of cells, taking pictures of the cells as they migrate to fill the scratch at various points throughout their migration, then comparing the pictures to determine how quickly the cells are migrating. The in vitro scratch experiment is superior to previous approaches in that it may be used to study the impact of cell-matrix and cell-cell interactions on cell migration, imitate in vivo wound healing, and allow imaging of live cells during migration to see intracellular events if desired(Goetsch and Niesler 2011).

The research was done using fibroblast stem cells. And the study was done to come to a concrete conclusion. The mechanism by which fibroblast feeder layer provides a microenvironment essential for stem cell maintenance are Fully characterised. There have been studies to prove that administration of plant - derived small molecules could improve stem cell proliferation .It is reported that gaule acid causes neural crest cells to differentiate(Hu et al. 2021).Modification of the conditioning media reveals significant improvement in the growth characteristics of stem cells. Improved proliferation is clearly evident in modified media . Gallic acid modification proves to be an effective method in simulating growth of stem cells

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