

## (MAPKI) Mitogen Activated Protein Kinase Inhibitor

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### Studying the effect of Mitogen Activated Protein Kinase Inhibitor (MAPKI) on Mesenchymal Stem Cells in vitro

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**OBJECTIVES:** Multipotent mesenchymal stem cells (MSc) are known for a long period of time. These cells are adherent, clonogenic and fibroblastic and can be isolated from bone marrow stroma of postnatal organisms. Under appropriate conditions, these cells can give rise to the broad spectrum of fully differentiated connective tissues including cartilage, bone, adipose tissues and muscle cells. Upon isolating of cells in vitro, these cells tend to differentiate and it is very difficult to direct these cells into self-renewal in order to get more Multipotent MSc for medical purposes. In the previous study, the effect of leukemia inhibitory factor (LIF) on MSc was investigated and it was shown that in the presence of LIF, MSc have more proliferation ability and the self renewal ability of cells increases. Also it was shown that these cells tend to differentiate into bones. In this study the effect of Mitogen Activated Protein Kinase Inhibitor (MAPKI) on MSc was investigated. **METHODS:** Bone marrow stroma cells were extracted from 2-month-old mice. These cells were cultured in a medium containing 25 micro moles MAPKI. After formation of colonies, cells were stained by methylen blue. The activity of alkaline Phosphatase was also investigated. To study the effect of both LIF and MAPKI simultaneously on colony formation ability of MSc, cells were cultured in a culture containing 25 micro moles MAPKI and 500 U/ml LIF. The number and size of colonies was studied. **Results:** In the presence of MAPKI the number and the size of colonies (CFU-F) formed in the culture was significantly decreased. The number of CFU-ALP<sup>+</sup> in the culture containing MAPKI was decreased according to the decrease of CFU-F number. Adding MAPKI to a medium containing LIF neutralized the effect of LIF on MSc. **Conclusion:** The presence of MAPKI in the culture affects the number of colonies. This factor, despite LIF, does not affect the expression of alkaline phosphatase in Mesenchymal Stem Cells. LIF increases the self renewal and proliferative ability of cells. Adding MAPKI factor to a culture containing LIF neutralizes the proliferative effect of LIF.

**Key words:** MAPKI, MSc, Alkaline Phosphatase, LIF, proliferation.

(LIF) Leukemia Inhibitory Factor

MAPKI

(MAPKI) MAP Kinase Inhibitor

MAPKI

(Colony Forming Unite-Fibroblastic ) CFU-F

CFU-ALP<sup>+</sup>

CFU-F

LIF

MAPKI

.(MSc)

Alkaline Phosphatase (Self-Renewal)

UPSTREAM

LIF :

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LIF

JAK-STAT

(gp130-LIFR) LIF

Adult Stem Cells

Embryonic Stem Cells

( )

SHP-2

Pluripotent

MAPK

ERK2 ERK1

Multipotent

( )

( )

LIF

( )

(Self-renewal)

MAPKI

( )

(MSc)

( )

LIF

CFU

Colony Forming Unit

CFU

( )

Multipotent

( )

CFU

LIF

( )

MAPKI

LIF

Self-Renewal

( )

Mitogen (LIF) Leukemia Inhibitory Factor

(MAPKI) Activated Protein Kinase Inhibitor

(Self Renewal)

Totipotent Pluripotent

( )

LIF

G

)

10x

(

PBS 10x

) αMEM

) NFB 10% ( % FBS %

( NaH2PO4 Na2HPO4

18G αMEM

DMF % MX-PO4 )

/ Red Violet LB Salt / PH / TRIS-HCL (C57BL/6)

( (Genetic Background)

CFU

CFU-ALP<sup>+</sup>

( )

CFU-ALP<sup>+</sup> (

CFU-ALP<sup>-</sup>

CFU : (Trypan Blue)

CFU-F

) 1X

( 10X

)

FBS % ) αMEM (

(

) Standard Deviation

(Excel

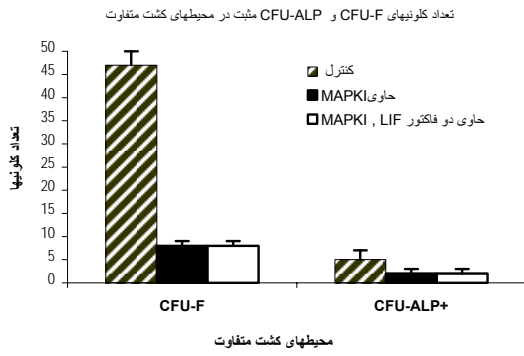
CO2

MAPK1

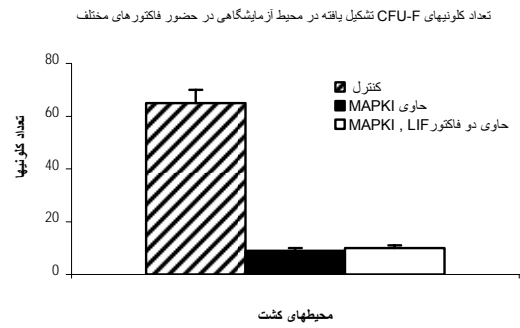
LIF

CFU-F (Gibco-BRL 1x10<sup>5</sup> U/μg) U/ml

( )



(MAPKI) MAPKI



CFU-ALP<sup>+</sup> CFU-F :  
 MAPKI CFU-ALP<sup>+</sup> CFU-F  
 LIF MAPKI  
 CFU-F ±  
 ±  
 CFU-F ± MAPKI  
 MAPKI LIF  
 MAPKI MAPKI LIF  
 ) MAPKI ( ( )  
 LIF MAPKI ( )  
 ) ( )

تعداد کلونیهای CFU-F تشکیل یافته در محیط آزمایشگاهی در حضور فاکتورهای مختلف

تعداد کلونیهای

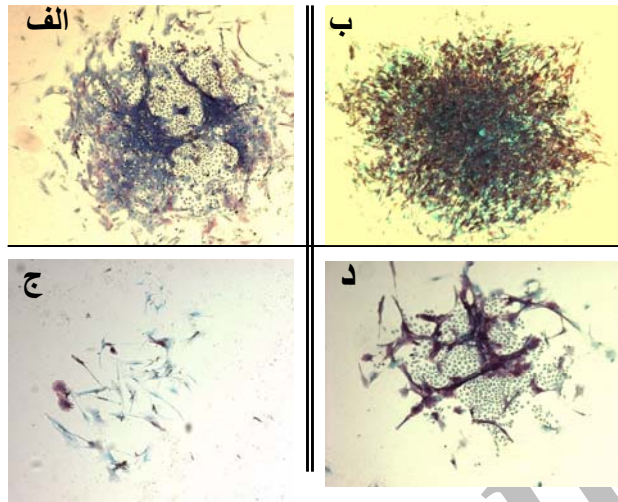
محیطهای کشت

کنترل  
 حاوی MAPKI  
 حاوی دو فاکتور MAPKI, LIF

CFU-F :  
 MAPKI  
 LIF  
 CFU-F  
 CFU-F  
 MAPKI  
 MAPKI  
 MAPKI + LIF ±  
 ±  
 MAPKI

(Colony Forming Unit Alkaline Phosphatase positive) CFU-ALP<sup>+</sup>

( )



الف) MAPK1 ( ) LIF  
 ب) MAPK1 ( ) LIF  
 ج) MAPK1 ( ) LIF  
 د) MAPK1 ( ) LIF



Multipotent MSc  
 In Vivo

(Self-Renewal)

( )

LIF

( )

LIF

(Self-Renewal)

MAPK1

(Age Dependent Osteoporosis)

MSc

( )

LIF

( )

MAPK1

LIF (Upstream)

MSc

( )

( )

MAPK1

MSc

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