視黃酸對成骨細胞分化的影響

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中文摘要:
視黃酸已被證實在調節細胞增生和分化中扮演重要角色. 許多證據也顯示成骨細胞的分化是經由一系列完整的基因表現來調控. 在分化過程中, 前驅細胞會壓抑本身的分化以維持一定數量的再生能力. 在本研究中我們發現視黃酸會增加轉錄因子Ets1之表現, 而此表現可能與成骨前區細維持其在生能力與防止分化有關.

Abstract:
There is extensive evidence that osteogenic cell differentiation is a multistep series of events mediated by an integrated cascade of gene expression. Retinoic acid has been reported as a potent regulator of cell proliferation and differentiation. Indeed, our preliminary data reveals that treatment of osteoblast-like PDL fibroblasts with retinoic acid increases the synthesis of EGF-receptor, which is only expressed in preosteoblast. This indicates that retinoic acid may act as a negative regulator for osteogenic cell differentiation. In this study, we have found that transcription factor Ets1 can be induced in MC3T3E1 cells by retinoic acid. Thus, the multiple functions of retinoic acid in bone cells are likely to be mediated in part by Ets1.

計劃緣由與目的:

It is well known that osteoblast differentiation and activities are regulated by numerous systemic hormones (Martin et al., 1987) and locally produced cytokines as well as growth factors (Krane et al., 1988). EGF and retinoic acid are both potent regulators in the control of cell growth and differentiation (Fisher et al., 1990). There is now extensive data to indicate that retinoic acid has the ability to modulate EGF and /or EGF receptors in several tissues. Most of the actions of retinoic acid are thought to result from changes in EGF-receptor gene expression which is probably caused via nuclear RARs and RXRs. Indeed, the transcripts of RAR-α and RAR-γ are constitutively expressed in osteoblast-like cells and the expression of RAR-β mRNA is induced by retinoic acid (Dolle et al., 1989; Tsukamoto et al., 1994). These observations suggest that retinoic acid may play an important role in regulation of osteoblast growth and

differntiation.

EGF-receptor has been found on both primary and certain clonal cells of osteoblastic lineage (Uneono et al., 1989; Bernier and Goltzman 1992). The expression of EGF-receptor on osteogenic cells, therefore, has been implicated in the regulation of osteoprogenitor cell proliferation (Yoneda 1996). We previously reported that a large number of EGF-receptor are expressed only on undifferentiated preosteoblasts and prechondrocytes. Interestingly, the number of EGF-recpetor on these cells falls dramatically as they differentiate into osteoblasts and chondrocytes.

Therefore, we hypothesize that retinoic acid may act as a negative regulator for osteogenic cell differentiation via modulation of EGF-receptor expression and transcriptional activation downstream its stimulation.

The purpose of this present study is to examine the effects of retinoic acid on osteoblast differentiation, and further to characterize possible gene expression involved in the process of regulation. By understanding the regulatory mechanisms of osteoblast differention and how progenitor cells remain as an undifferentiated phenotype, we may develop molecular based strategies to recruit and promote mitogenic growth of osteoprogenitor cells, and further increase bone-forming activity via the osteoprogenitor cell cycle.

結果與討論:

1. *Retinoic acid receptor expression in MC3T3E1 cells*

To see the expression profile of retinoic acid (RA) receptors in MC3T3E1 cells, we treated cells with all trans RA (atRA) for several time courses. Our results indicate that RARβ expression is atRA dependent and that RXRα and RXRβ transcript levels are not significantly altered after atRA treatment. However, we found that RARγ is moderately enhanced in the presence of atRA.

2.*Tanscription factor Ets1 mRNA expression is induced by retinoic acid*

Ets1 transcription factor is a oncoprotein acceleration cell growth and proliferation.

To examine whether RA inhibits preosteoblat MC3T3E1 differentiating into osteoblast by activating Ets1, we analyzed by Northern blot. We observed a increase in Ets1 mRNA levels after 4hr treatment of atRA. In the presence of atRA, this induction can be maintained for at least 14 days.

3. *Ets1 expression in bone differentiation*

The process of maturation of MC3T3E1 cells can be observed in three distinct phases, namely proliferation, differentioation and mineralization. We found that Ets1 is expressed in proliferating preosteoblatic phase. The expression pattern suggests that Ets1

may act by regulating genes involved in proliferation upon RA stimulation.

成果自評:

In this study, we have demonstrated that transcription factor Ets1 mRNA is induced by atRA in MC3T3E1 cells. This data also implies that retinoic acid may act as a negative regulator in osteoblast differentiation by enhancing Ets1.

However, the mechanism how Ets1 regulate preosteoblast in progenitor maintenance is still unclear. The relationship between RA and Ets1 should be further elucidated.

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