

# Responses to fluid flow during early osteogenic differentiation of mesenchymal stem cells

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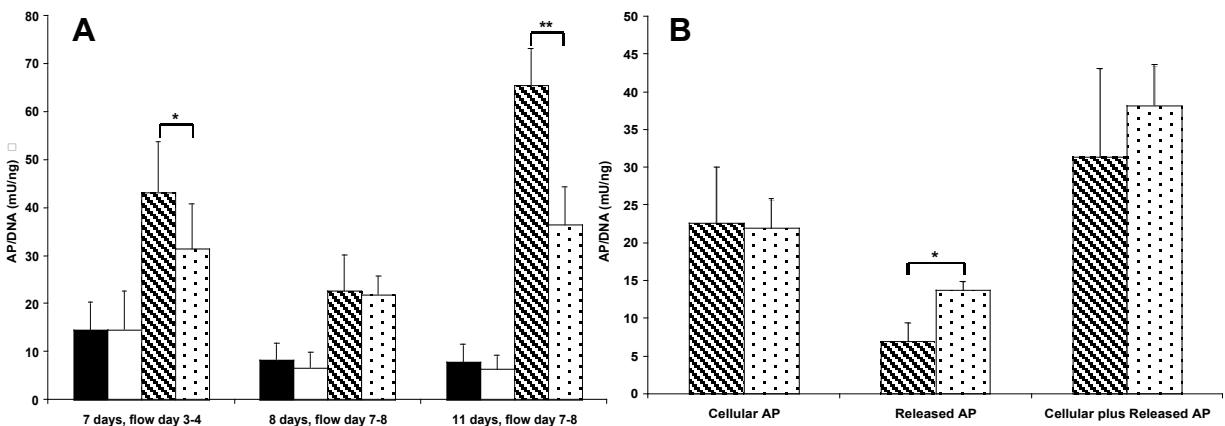
## Introduction

Human mesenchymal stem cells (hMSCs) are multipotent cells capable of self-renewal and differentiating into cells of mesenchyme origin, such as bone, cartilage, adipose, tendon, ligament and skin. Exogenous mechanical stresses can regulate an increasing amount of cellular components and mechanosensitive markers of multiple cell types including hMSCs.

## Methods

Human MSCs were cultured in either basal or 'osteogenic' medium (hMSC-Obs) for 3 days (group A) or 7 days (groups B and C). Both hMSCs and hMSC-Obs were subjected to fluid flow at 9 dyne/cm<sup>2</sup> for 24 hrs in a parallel plate flow chamber with flow loop. Following flow, groups A and B were returned to culture in basal or osteogenic medium, respectively, for 3 additional days, and then assayed for alkaline phosphatase (AP) activity and DNA content. Group C was assayed for cellular and released AP activity and DNA content immediately after flow treatment.

## Results



**Figure 1.** Alkaline phosphatase activity per cellular DNA. (A) Fluid flow-induced shear stress reduced sustained cellular AP/DNA at two time points but had no effect on immediate AP/DNA. (B) Fluid flow-induced shear stress reduced released AP/DNA at 8 days but had no effect on cellular or total AP/DNA. Black boxes are hMSC – no flow, white boxes are hMSC – flow, striped boxes are hMSC-Ob – no flow, dotted boxes are hMSC-Ob – flow. Values are mean +SEM of independent experiments: \*,  $p < 0.05$ ; \*\*,  $p < 0.01$ ; \*\*\*,  $p = 0.005$  ([A] 7 days,  $n = 4$ ; 8 days, hMSC  $n = 6$ , hMSC-Ob  $n = 7$ ; 11 days,  $n = 5$ ; [B] Cellular AP,  $n = 6$ ; released AP,  $n = 4$ ; cellular plus released AP,  $n = 4$ ).

## Discussion

This regime may inhibit early osteogenic differentiation of hMSCs, and help to understand development of early osteogenic differentiation.