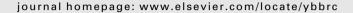


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## Biochemical and Biophysical Research Communications





Letter to the Editor

## Letter to the Editor on "Effects of BMP-2 and vitamin D3 on the osteogenic differentiation of adipose stem cells"

The induction of osteogenic differentiation of adipose stem cells (ASCs) is one of the hot topics in the field of bone tissue engineering. For such purpose, bone morphogenetic protein-2 (BMP-2) and Vitamin  $D_3$  (VD<sub>3</sub>) are two promising exogenous growth factors. Our group is also working on this topic. It is exciting to see that the publication of "I. Song et al., Effects of BMP-2 and vitamin D3 on the osteogenic differentiation of adipose stem cells, Biochem. Biophys. Res. Commun. 408 (1) (2011) 126-131." systematically demonstrated the respective and synergistic effects of BMP-2 and VD<sub>3</sub>. However, there were some inconsistencies in the results of the synergistic effects of these two growth factors. From Fig. 1 in this publication, we can estimate the folds of alkaline phosphatase (ALP) and mineralization that were induced by osteogenic media (OM) including 50 ng/ml BMP-2 or 10<sup>-7</sup> M VD<sub>3</sub> in comparison with that by OM alone. From Fig. 2, we can estimate the folds of ALP and mineralization that were induced by OM including 50 ng/ml BMP-2 and  $10^{-7}$  M VD<sub>3</sub> in comparison with that by OM including only  $10^{-7}$  M VD<sub>3</sub>. From Fig. 3, we can estimate the folds of ALP and mineralization that were induced by OM including 50 ng/ml BMP-2 and 10<sup>-7</sup> M VD<sub>3</sub> in comparison to OM including only 50 ng/ml BMP-2. By normalizing these data to that of OM alone, we could find big discrepancy between the data of the ALP and mineralization induced by 50 ng/ml BMP-2 and  $10^{-7}$  M VD<sub>3</sub> (ALP: 3.08 vs 8.51; 8.41 vs 27.26, Mineralization: 4.87 vs 8.20) from Fig. 2 and Fig. 3 (Table 1). Such inconsistencies in ALP data also existed within Fig. 4, when normalizing the data to that of OM alone in Fig. 1. The discrepancies in actual folds of ALP and mineralization were too significant to be ignored.

In addition, Fig. 2A demonstrated no significant mineralization (Alizarin red staining) of ASCs after a 14-day treatment of OM including  $10^{-7}$  M VD $_3$  without 50 ng/ml BMP-2 (the second column and third row in Fig. 2A). However, Fig. 4A demonstrated significant mineralization after a 14-day treatment of OM including  $10^{-7}$ M VD $_3$  without 50 ng/ml BMP-2 (the second column and second row in Fig. 4A). Consequently, the results are too confusing and inconsistent to make a definite conclusion. An explanation should be clarified for these inconsistent results. If such inconsistent result was due to the facts that ASCs may come from different donors, a discussion and precaution should be stated in the section of Discussion.

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Available online 18 September 2012

**Table 1**Discrepancies in alkaline phosphatase activity (ALP) and mineralization of adipose stem cells that were induced by osteogenic medium (OM) with BMP-2 and VD<sub>3</sub>.

		VD <sub>3</sub> (10 <sup>-7</sup> M)	BMP-2 (50 ng/ml)	ALP		Mineralization	
				7 days	14 days	14 days	24 days
Estimated folds from Fig. 1		-	-	1	1	1	1
, and the second	A	-	+	2.35	2.95	2.1	1.94
	В	+	-	2.43	3.95	2.94	1.59
Estimated folds from Fig. 2		+	-	1	1	1	1
	С	+	+	3.5	6.9	2.79	7.9
Estimated folds from Fig. 3		-	+	1	1	1	1
	D	+	+	1.31	2.85	2.32	6.1
Normalization to OM without BMP-2 or $VD_3$		-	-	1	1	1	1
	normalized D = $A \times D$	+	+	3.08	8.41	4.87	11.83
	normalized $C = B \times C$	+	+	8.51	27.26	8.20	12.56

<sup>\*</sup>DOI of original article: http://dx.doi.org/10.1016/j.bbrc.2011.03.135