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Letter to the Editor

Rebuttal to “Letter to the Editor on ‘Effects of BMP-2 and vitamin D3 on the osteogenic differentiation of adipose stem cells’”

We thank the group of Dr. Wu for their interest in our study “Effects of BMP-2 and vitamin D3 on the osteogenic differentiation of adipose stem cells”. They raised a question on the inconsistency in the synergistic effects of two factors, VD3 and BMP-2. We would like to emphasize that it is not reasonable to calculate the estimated fold from ASCs by multiplying normalized values as Dr. Wu suggested. We used primary cell from several individuals. Therefore the control condition was necessary for every experiment, and the data were represented as ratio over the control. In addition, even the primary cells from the same patient can behave differently depending on the reagents, especially FBS as well as BMPs according to different lots. Considering that experiments in Figs. 1–4 were performed several months apart, there is a good possibility that cells can respond differently depending on the timing of experiments. That is why the controls are needed every time, and normalized D ($A \times D$) can be different from normalized C ($B \times C$). For example, the ALP levels of a control sample can vary according to different lot of reagents and timing of culture, and its response to VD3 or BMP-2 can also vary. So we think that the general trend in the value (the ratio over the control) and statistical analysis matter here. One difference in the method between Figs. 2 and 3 is that while VD3 was first treated followed by BMP-2 treatment in Fig. 2, the reverse was done in Fig. 3. However,

it is a matter of seconds (1–2 min at the longest) and we do not believe that the order of treatment significantly affected the results. Of course, we agree with Dr. Woo in that ASCs from different donors can show variable response to an identical culture condition, which may explain the difference in Alizarin red staining between Figs. 2A and 4A.

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