

## SUPPLEMENTAL DATA

### The serotonergic 5-HT<sub>2B</sub> receptor controls Tissue Non-Specific Alkaline Phosphatase activity in osteoblasts via eicosanoids and phosphatidylinositol-specific phospholipase C

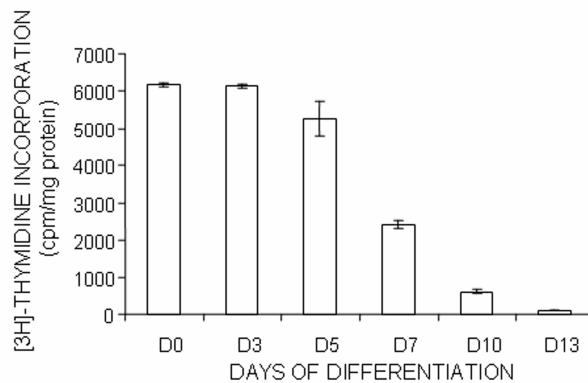
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#### Supplementary Experimental Procedures

*Thymidine incorporation* - [<sup>3</sup>H]thymidine (0.5  $\mu$ Ci; 1 Ci = 37 GBq) was added to C1 cells for 4 h at 37°C. The free radioactive thymidine was washed away in 5% trichloroacetic acid, and the incorporated radioactive thymidine in cell extracts was quantified by scintillation counting (1). Results were normalized to protein concentration.

#### References

1. Nebigil, C.G., Launay, J.M., Hickel, P., Tournois, C., and Maroteaux, L. (2000) *Proc Natl Acad Sci USA* **97**, 2591-2596.



**SUPPL. FIGURE 1: The reduction in C1 cell proliferation at the end stage of the osteogenic program mirrors the conversion of C1 cells to osteocytes-like cells.** Cell proliferation was determined during osteogenic differentiation of C1 cells by measuring [<sup>3</sup>H]thymidine incorporation. At indicated time, C1 cells were incubated with 0.5  $\mu$ Ci of [<sup>3</sup>H]thymidine for 4 h. Between days 10 and 13, when they convert into osteocytes, C1 cells cease dividing. Values are the means  $\pm$  sem of three independent experiments.