## **Supplemental Materials**

STIM1 a calcium sensor promotes the assembly of an ECM that contains Extracellular vesicles and factors that modulate mineralization

Yinghua Chen, Rahul Koshy, Elizabeth Guirado & Anne George\*

Brodie Tooth Development Genetics & Regenerative Medicine Research Laboratory Department of Oral Biology University of Illinois at Chicago Chicago, IL 60612, USA

\* To whom correspondence should be addressed. Anne George Department of Oral Biology, 312-413-0738 FAX: 312-996-6044 E-mail: anneg@uic.edu

- 1. Supplement Figure S1: Localization of STIM1 and Orai1 in the mandibles of wild type and STIM1 KO mice:
  - A. Expression of STIM1 in the differentiating odontoblasts, alveolar bone and dental pulp cells in 2-month old WT and STIM1 KO mouse heads. Boxes indicated as 1 and 2 have been magnified and the corresponding images are shown in the panel. P=pulp; OD=odontoblasts, Den = dentin and AB=alveolar bone.
  - B. Quantification of the immunohistochemical positive signals in odontoblasts (Od), pulp= (P) and alveolar bone (AB). Statistical significance: \*\*p<0.01.</li>
  - C. Colocalization of STIM1 (green) and Orai1 (Red) in the dental pulp of day5 wild type mice.
- 2. Supplement Figure S2: Gene expression analysis at day 0 in transgenic DPSCs:

Gene expression analysis of the "early" and "late" differentiation markers expressed in control and genetically modified DPSCs at day 0. Statistically significant differences are indicated. \*: p<0.05, \*\*: p<0.01.

3. Supplement Figure S3: Characterization of the crown and root dentin by micro-computed tomographic analyses:

Dentin thickness, Dentin volume fraction (BV/TV) and tissue mineral density (TMD) are shown for 1, 2 & 3 month WT and STIM1 knockout

mice. Statistically significant differences are indicated. \*: p<0.05, \*\*: p<0.01.

## 4. Supplement Figure S4: Characterization of the root dentin by $\mu\text{CT}$

Representative micro-CT image of the first molar in 1, 2 & 3 months WT and STIM1-KO mice (n=6). The regions contoured for measuring dentin thickness were illustrated as red circles. Note reduced dentin thickness in 1, 2 and 3M STIM1-KO mice. Statistical significance: \*: p< 0.05.

## 5. Supplement Figure S5: Live-cell imaging showing changes in spatial distribution of STIM1:

Time-lapse imaging to show the spatial distribution of CFP-STIM1 in DPSCs in response to ER  $Ca^{2+}$  depletion by 500ng/ml of DMP1.



Supplemental Fig S1A-B: Localization of STIM1 and Orai1 in the mandibles of wild type and STIM1 KO mice:



## Supplemental Fig S1C: Localization of STIM1 and Orai1 in the mandibles of 5 day wild type mice



Supplement Figure S2: Gene expression analysis at day 0 in transgenic DPSCs



Supplement Figure S3: Characterization of the crown and root dentin by micro-computed tomographic analyses



Supplement Figure S4: Characterization of the root dentin by  $\mu$ CT



Supplement Figure S5: Live-cell imaging showing changes in spatial distribution of STIM1

Movie