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# Nanoscale arrangement of the healthy dentin and comparison with the dentino-genesis imperfecta-affected dentin

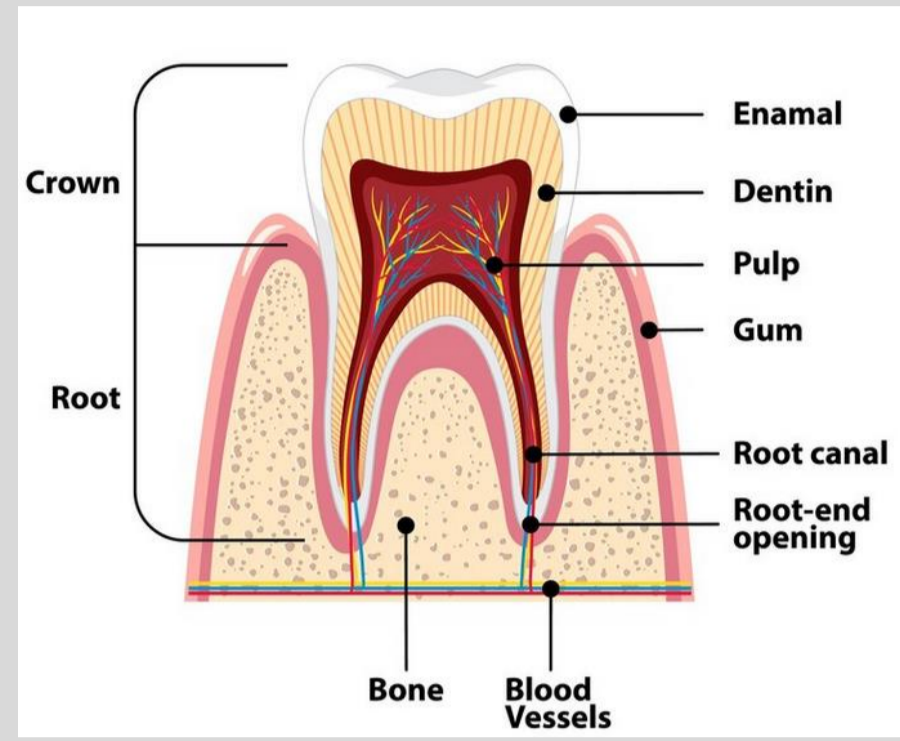
Maxime VALLET<sup>1,2</sup>, Margot Leclercq<sup>1</sup>, Margot Riou<sup>3,4</sup>, Elsa Vennat<sup>1</sup>

<sup>1</sup> Université Paris-Saclay, CentraleSupélec, ENS Paris-Saclay, CNRS, LMPS, France  
<sup>2</sup> Université Paris-Saclay, CentraleSupélec, CNRS, Laboratoire SPMS, France

<sup>3</sup> Université de Paris, Dental Faculty, Paris, France

<sup>4</sup> Centre de Recherche des Cordeliers, Université de Paris, Sorbonne Université, INSERM UMRS 1138, Molecular Oral Pathophysiology, Paris, France

## Introduction



Dentin is a complex biological hard tissue presenting a hierarchical organization at different scales as bones. At the nanoscale, dentin shows an organization involving a mineral phase made of HAP crystals and an organic phase mainly made of CFs.

### Dentinogenesis imperfecta (DI)

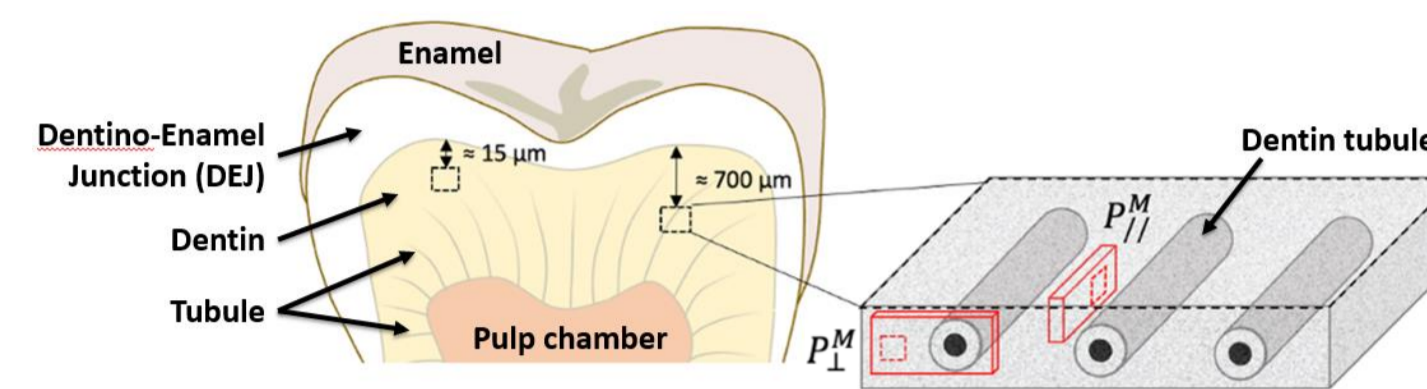
- Rare genetic disorder (1-5/10 000 births)
- Severe anomalous dentin



## Method

Both permanent and deciduous teeth were prepared for the control samples. In contrast, a deciduous molar was chosen for DI ones exhibiting damage to type I collagen

FEI ThermoFisher Helios Nanolab 660 focused ion beam for localized sections

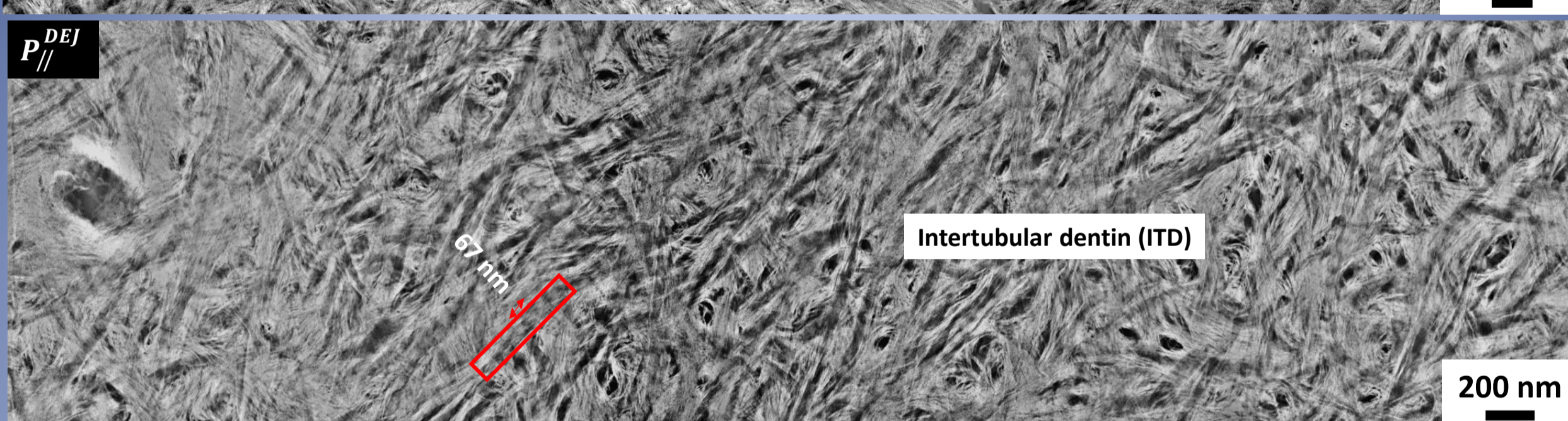
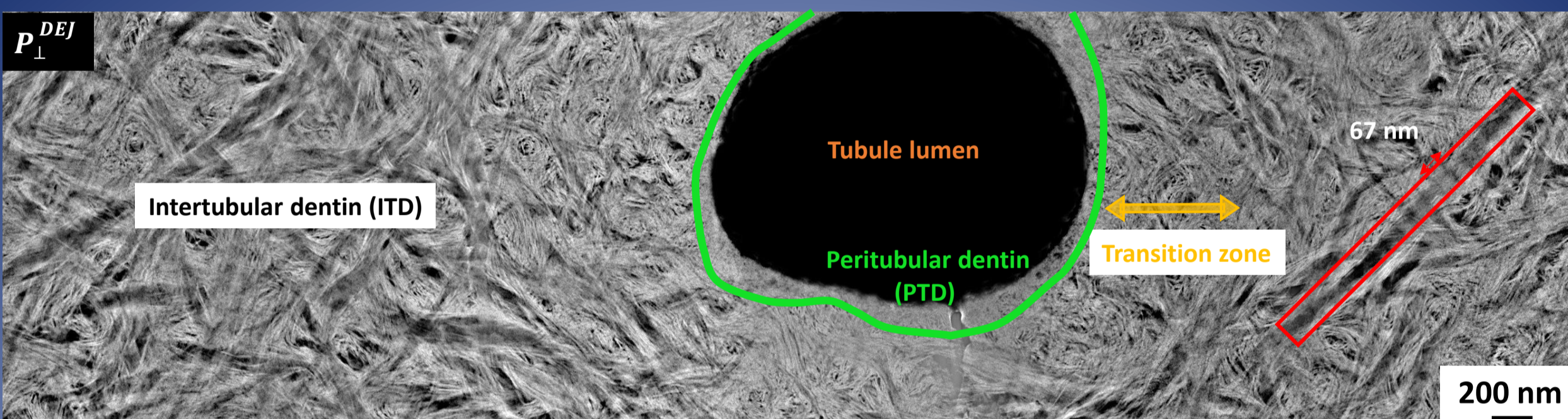


FEI ThermoFisher TITAN<sup>3</sup> G2 80-300 transmission electron microscope used in TEM and STEM mode

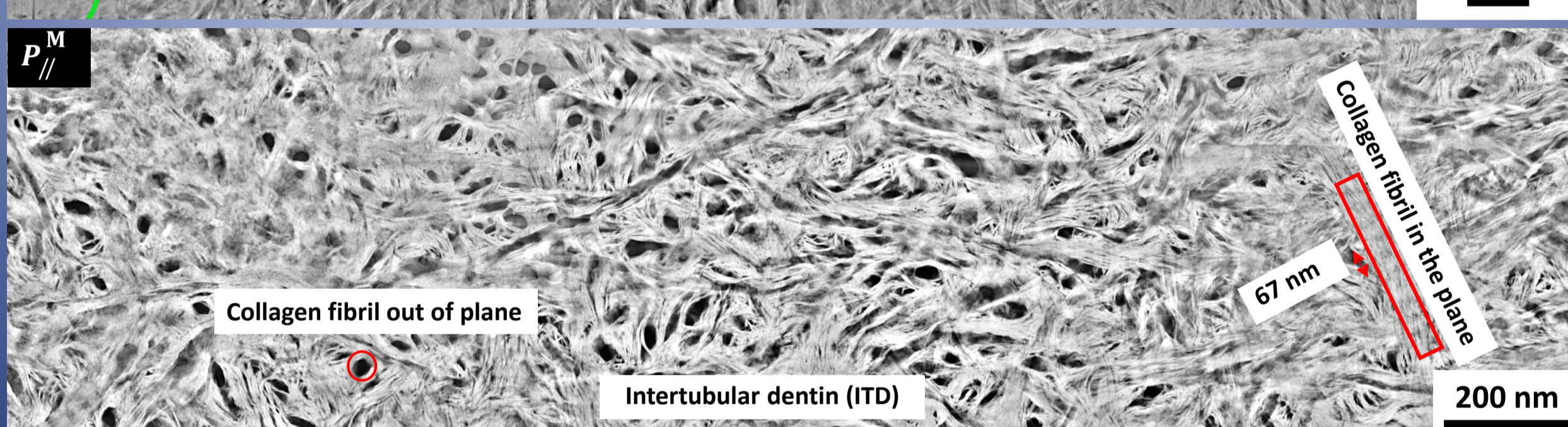
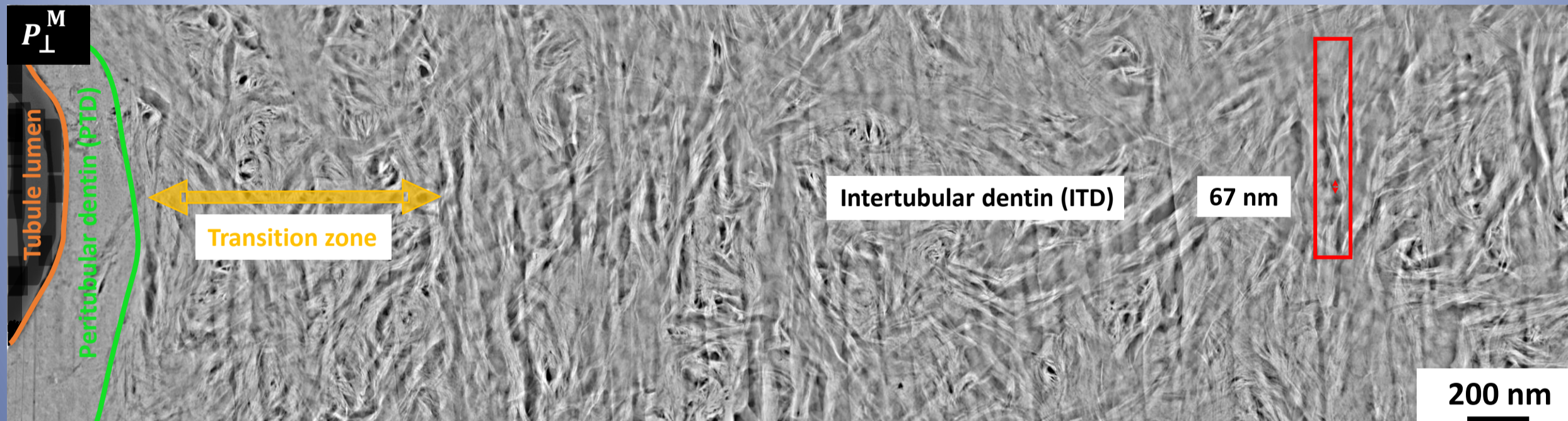
## Nanostructure of the healthy dentin [1]

### Collagen fibrils (CFs) distribution

#### HAADF-STEM images of dentin near enamel



#### HAADF-STEM images of middle dentin

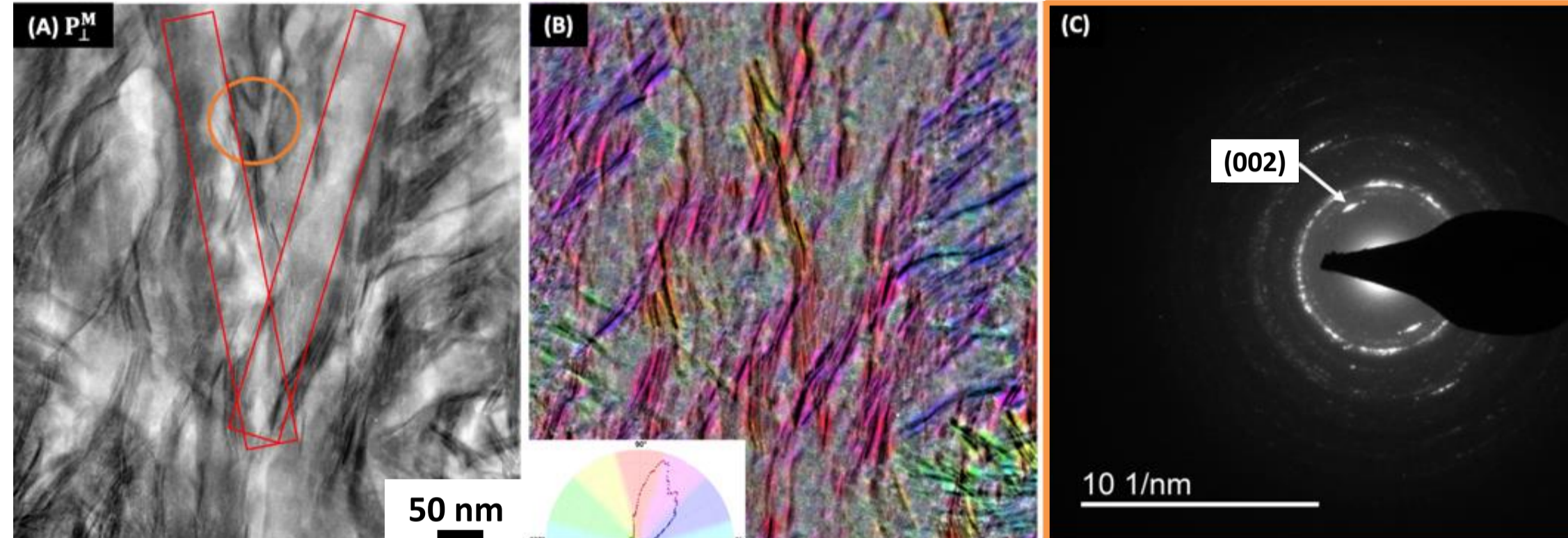


**Collagen fibrils (CFs)**  
1- Near enamel: CFs are roughly isotropic  
2- Middle dentine: CFs mainly lie in planes perpendicular to the tubule axis and create a woven structure

**Transition zone between PTD and ITD**  
Mineralized collagen fibrils are running parallel to the tubule axis, unlike in the rest of ITD. It might explain that cracks initiated in tubules do not propagate in ITD

### Hydroxyapatite (HAP) cristal organization

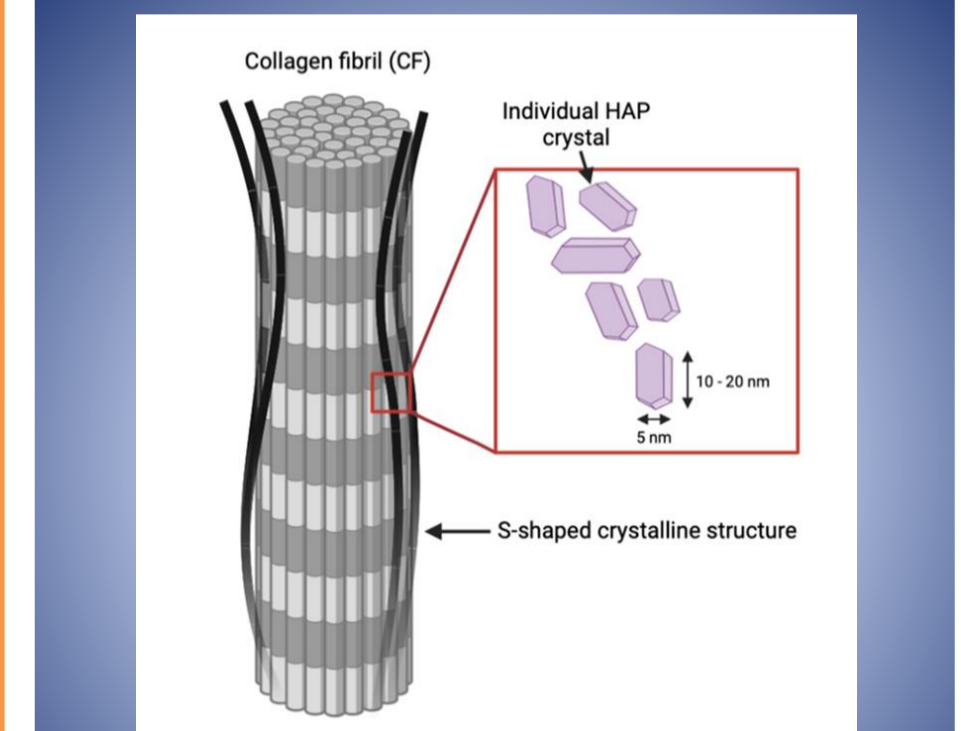
#### BFTEM image zoomed on some collagen fibrils



The HAP minerals (in black) surrounding collagen fibrils (red rectangles) Color-coded map of the BF showing the angle distribution of minerals Diffraction pattern of the orange circle in figure (a)

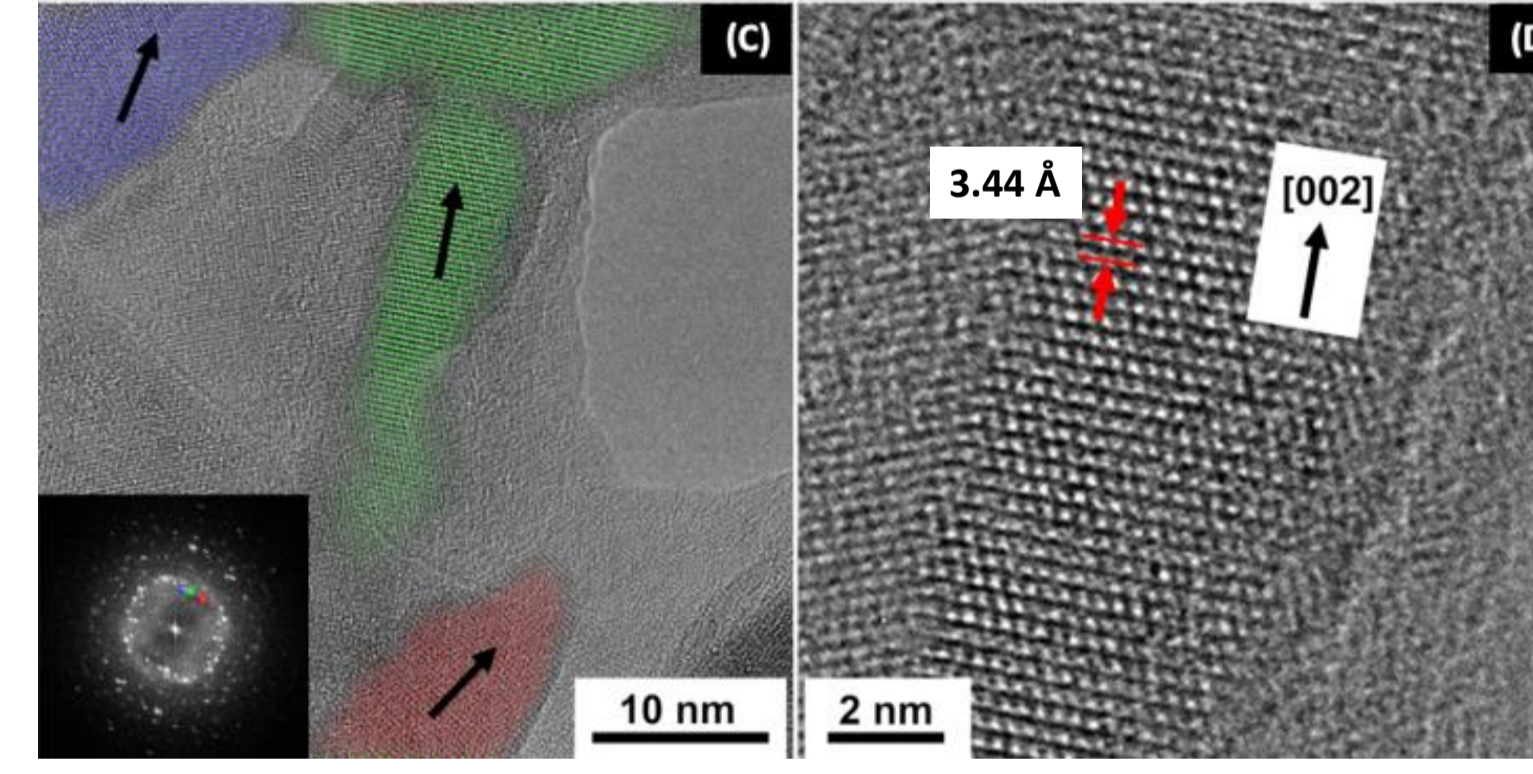
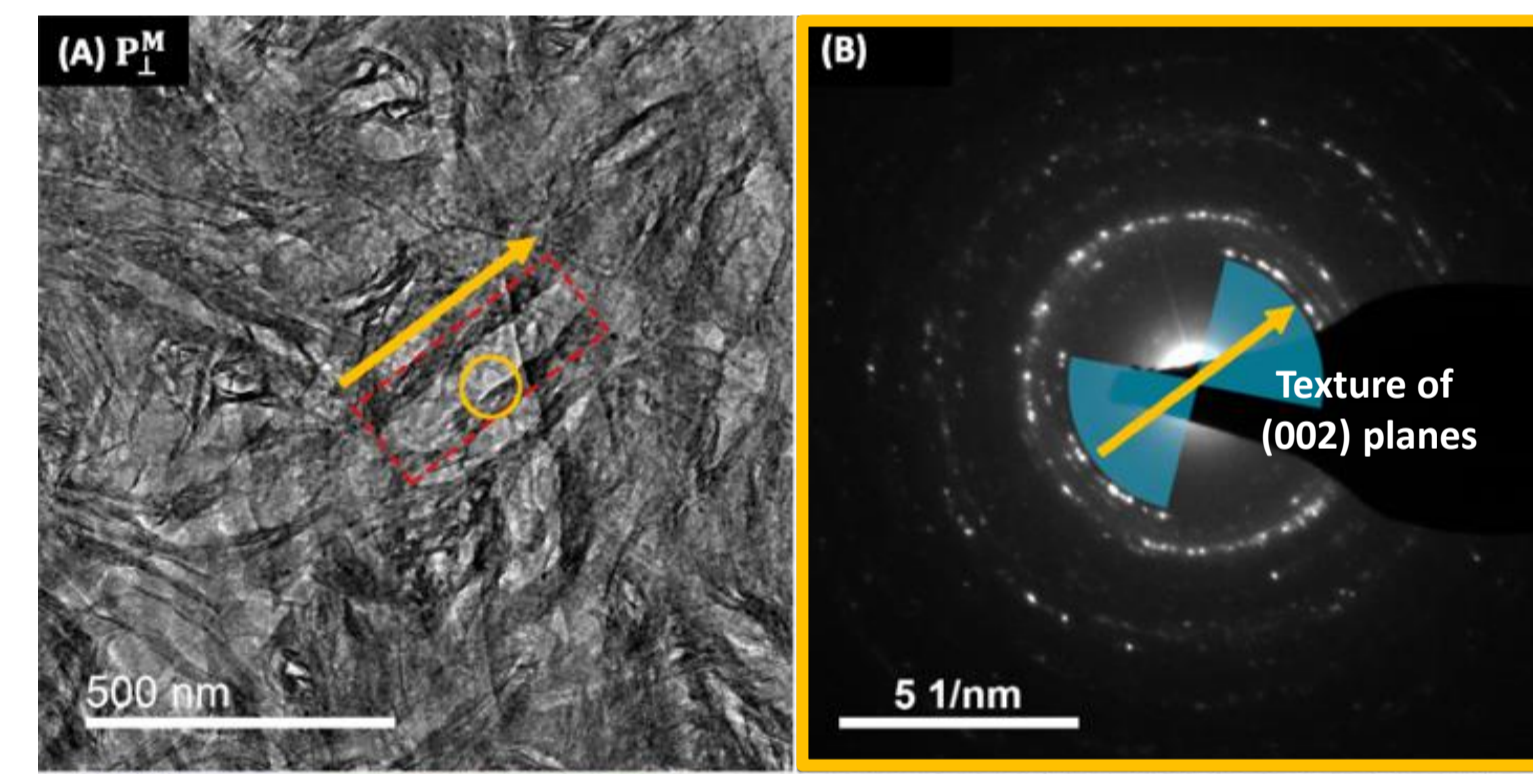
HAP minerals are strongly oriented around the collagen fibrils. The [001] direction is mainly lying parallel to the collagen fibrils as see in the diffraction pattern.

#### Model proposed by Reznikov et al. [2] for cortical bones



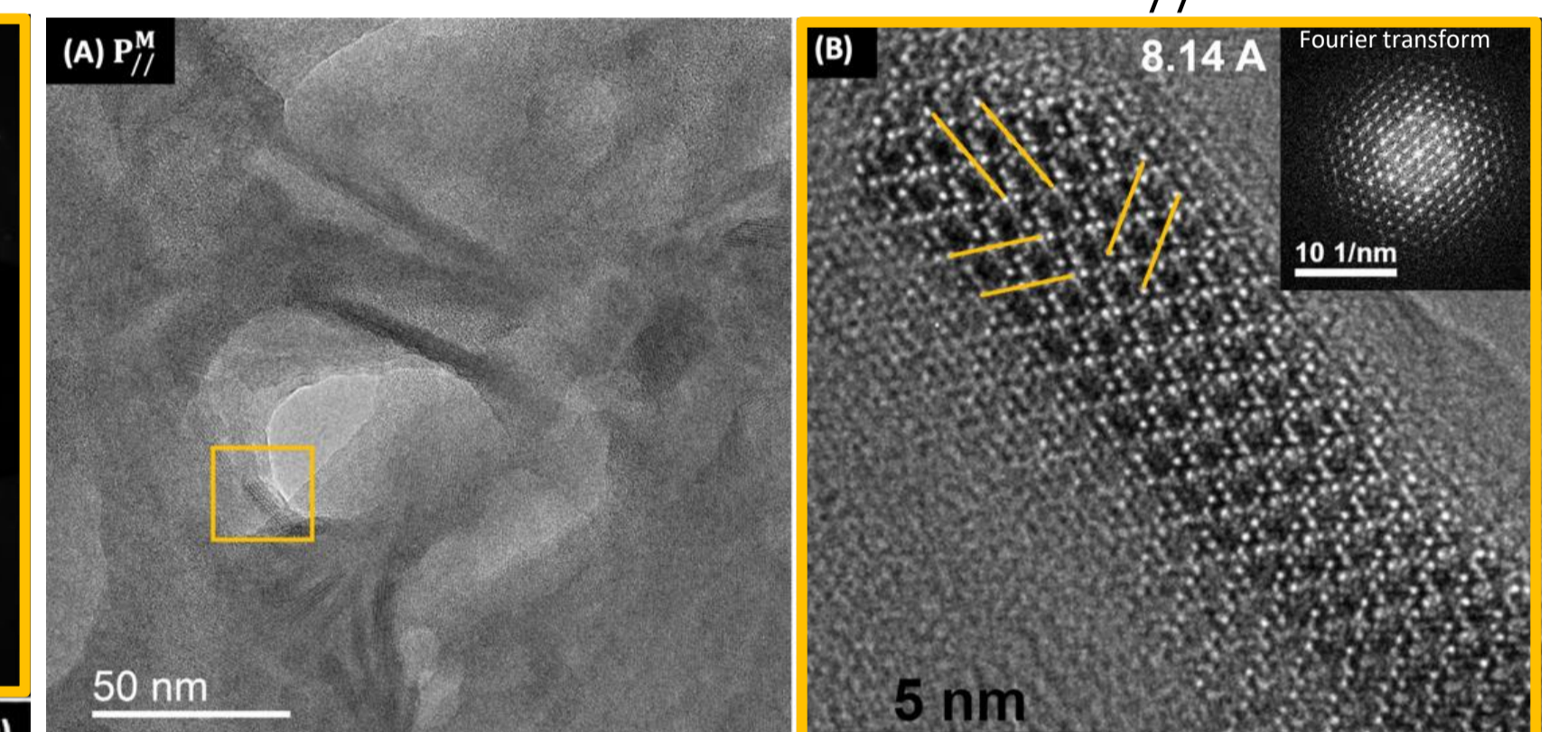
Organization of merged individual HAP crystals with various orientation around adjacent CFs and forming a S-shaped crystalline structure

#### HRTEM image of HAP crystals in P<sub>⊥</sub><sup>M</sup>



The diffraction pattern (b) taken on the side of a single CF (a) again shows a preferred orientation of HAP crystals with their [001] direction roughly aligned with the CF one. (c) shows three crystals highlighted by choosing their (002) reflections. (d) Zoom on a single crystal with the (002) interplane distance of 3.44 Å

#### HRTEM image of HAP crystals in P<sub>∥</sub><sup>M</sup>



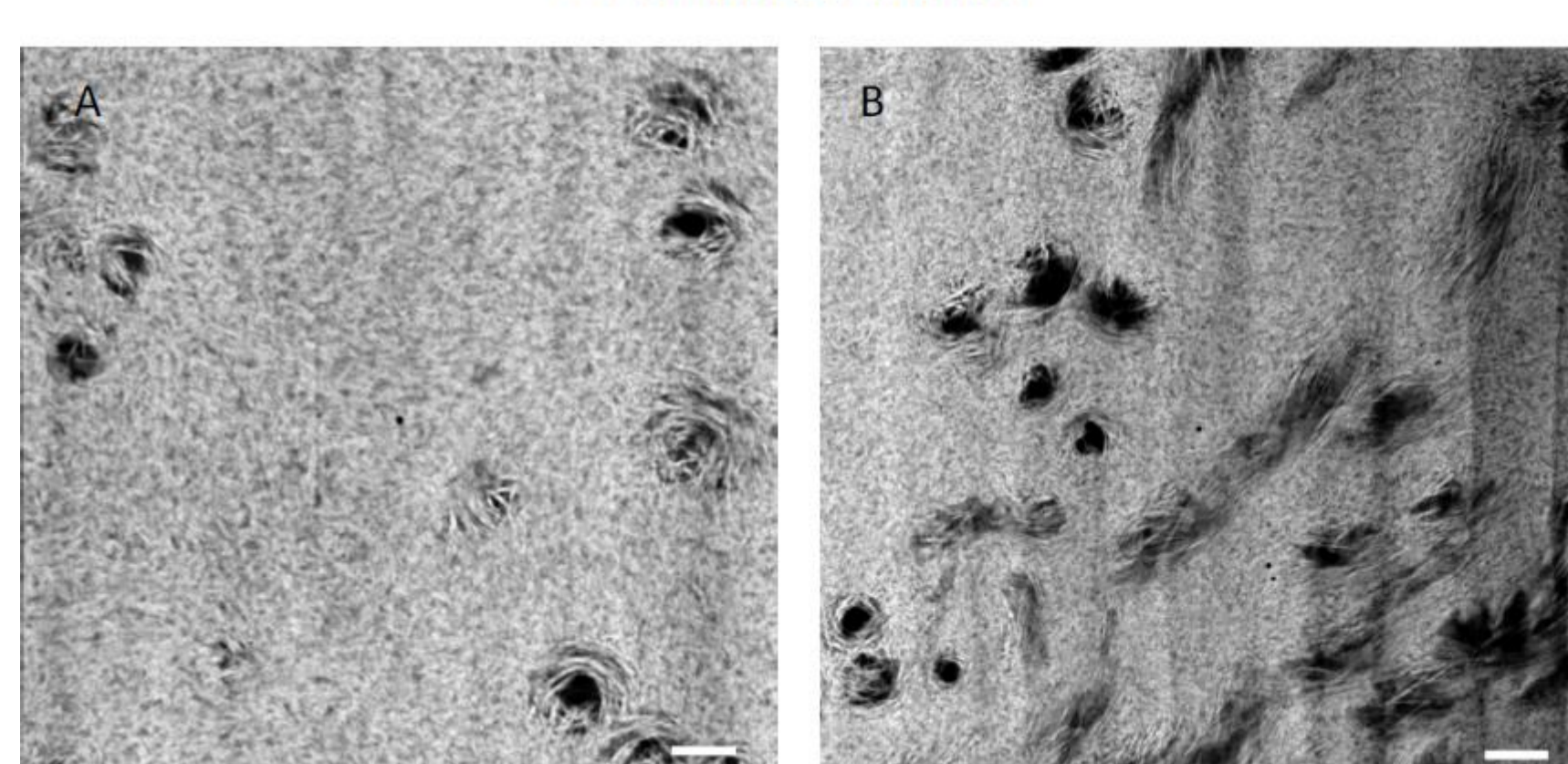
Atomic arrangement of a single HAP crystal seen normal to the c-axis. Periodicities of 8.14 Å characteristic of basal interplane distances are observed in P<sub>∥</sub><sup>M</sup>

1. Single HAP crystals in all samples
  - Width 4-5 nm
  - Length few tens of nanometers
2. Elongated mainly along their [001] c-axis but were also seen elongated in the other crystalline directions (a, b axis).
3. Formation of a S-shape crystalline structure composed of merged single HAP crystals

## Dentinogenesis Imperfecta (DI)-affected dentin

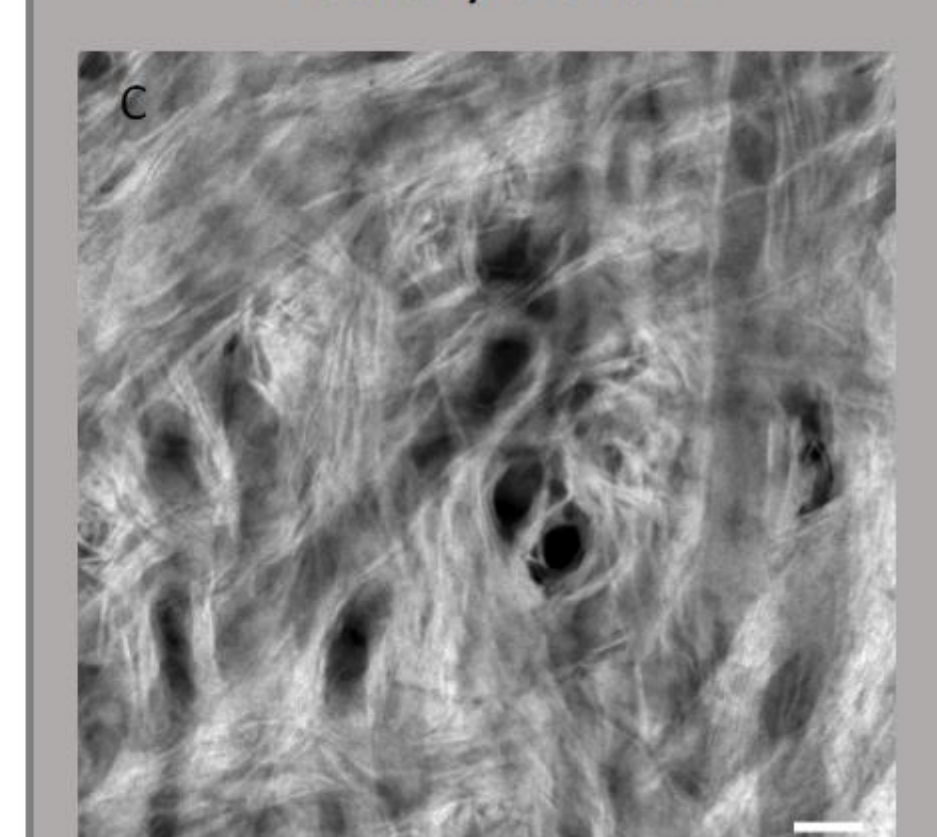
### HAADF-STEM images of healthy and DI-affected dentin at the same scale (50 nm)

#### DI-affected dentin



- 1- Few or no collagen fibrils with varying orientation planes in DI-affected dentine
- 2- Hypermineralization of the dentin: more than 90%

#### healthy dentin



Mineralization of the healthy dentin around 55% in volume has been evaluated by Vennat et al. 2009 [3]

In DI-affected dentin samples, a drastic decrease in organic content is locally observed associated with a different organization of the remaining collagen fibrils



Modification of the mechanical properties due to a high mineral content:

- Harder
- Stiffer
- More brittle

## Conclusions

- Isotropic distribution of CFs close to the enamel than in the middle dentin
- Organization of HAP in the [001] direction around CFs forming S-shaped crystalline structure
- Low mineralization structures reduce the propagation of cracks
- Hypermineralization of DI-affected dentin at the origin of a significant increase of the brittleness