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Molecular structure characterization of extracted cellulose from different apple cultivars by transmission electron microscopy.

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Biopolymers have many applications in the food area and confer specific functional properties to both fresh food and processed products. Apples have cell wall biopolymers that play a pivotal role in fruit firmness. Different apple cultivars have variations in their physicochemical and microstructural properties that confer distinctive attributes to each cultivar. Among then, firmness plays an important role and as a consequence their textural properties have been studied intensively. It is commonly recognized that the cellular architecture of the apple mesocarp is strongly linked to fruit firmness. However, apple firmness and their quality attributes vary significantly with regards to ripening stage and cultivar. Previously, a complete and multifactorial analysis has been carried out. This includes the physicochemical, structural, and nanomechanical properties at macro, micro and nanometric scales. In particular the goal is to determine the features with the greatest influence on the firmness of selected apple cultivars (Golden Delicious, Granny Smith, Gala and Red Delicious). Physicochemical assays, microscopy techniques, image analysis, nanoindentation and spectroscopy have been used to characterize the properties of the four selected apples [1]. It has been determined that the cellular architecture, stiffness of cell walls and crystallinity index of cellulose fibers are the most important factors related to the firmness of apples. However, the cellulose structure of cell wall of apple mesocarp has not been characterized at the molecular level, so it is probable that its crystalline structure plays an important role in the structural and mechanical properties at different scales. Thus, in this work the cellulose purified from some apple cultivars has been investigated by transmission electron microscopy. Samples are prepared on C grids and low dose techniques are applied in order to preserve the genuine structure of the samples. Exit wave reconstruction techniques are applied in low dose conditions of around 10 e/Å2s in the TEAM 1 electron microscope of the MF-LBNL.

Figure 1 shows a phase image after EWR with 50 images. The sample has been extracted from the cultivar Golden Delicious apple. It clearly shows the lattice spacings of different crystallites with an average size of 4 nm, the particles have a varying orientation and different lattice spacings can be measured. The corresponding lattice in reciprocal space are shown in the upper right corner of the image. As expected this is a ring pattern. The image resolution is sufficiently high as to allow comparison to published results. Figure 2 shows the extracted biopolymer from the Gala Cultivar. The complete image is now formed by a single crystallite. All cultivars show these two variants i.e., nano and micro crystallites. Apparently such sizes depend upon the extraction technique and the milling that has been applied in this particular case. The right upper corner in Fig. 2 shows the corresponding diffraction pattern showing single crystallite characteristics. Here the d-spacing are also of interest as to determine the crystalline structure with a good special resolution. Electron microscopy is helpful to characterize the molecular structure of purified cellulose from apples, the provided information can be useful to develop bionanomaterials from apple waste.

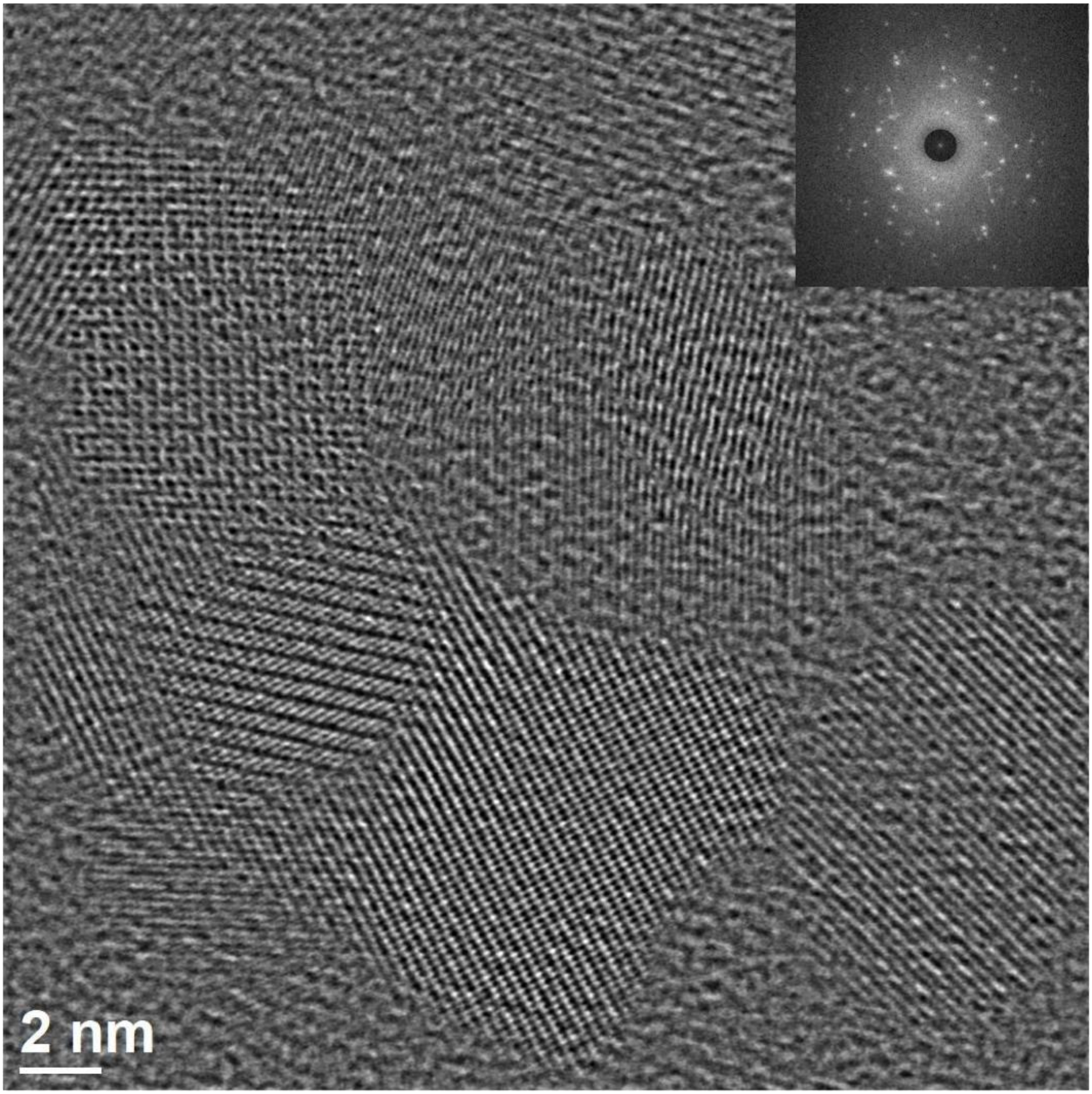

Figure 1. Figure 1. Phase image after a EWR procedure of 50 images in a focal series for cellulose of a Golden delicious apple cultivar. Different crystallites with nanometric sizes are shown together with the corresponding diffraction pattern.

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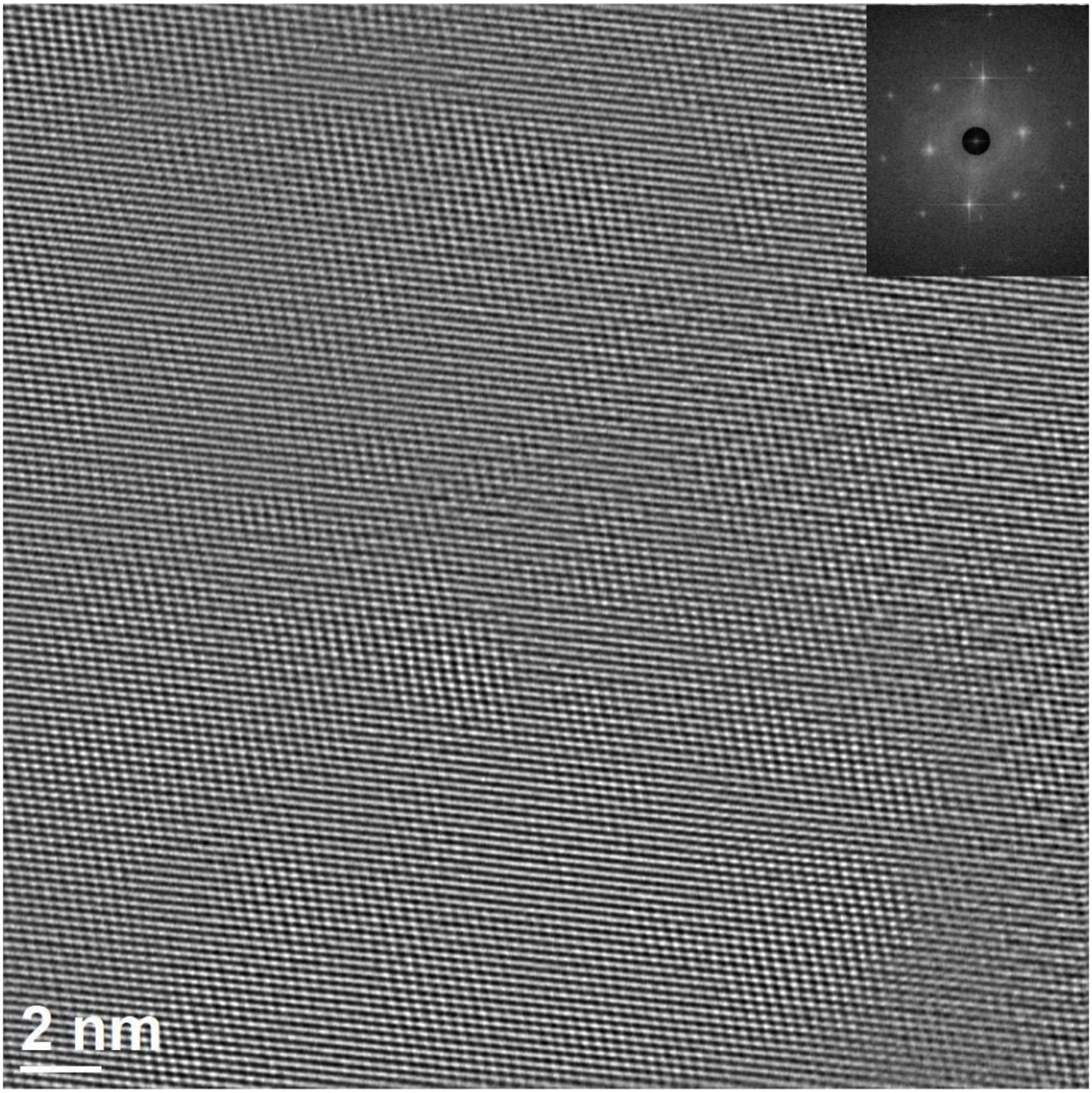

Figure 2. Figure 2. Phase image after a EWR procedure of 50 images in a focal series for cellulose of a Gala apple cultivar. Large crystallite with a micrometric sizes is shown together with the corresponding diffraction pattern.

References

1. L.E. Rojas-Candelas et al., Postharvest Biology and Tech. 171 (2021) 111342.

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