Supplementary Material

Biological control of Ca-carbonate crystal microstructure and texture: adapting crystal morphology, orientation and arrangement to biomaterial function

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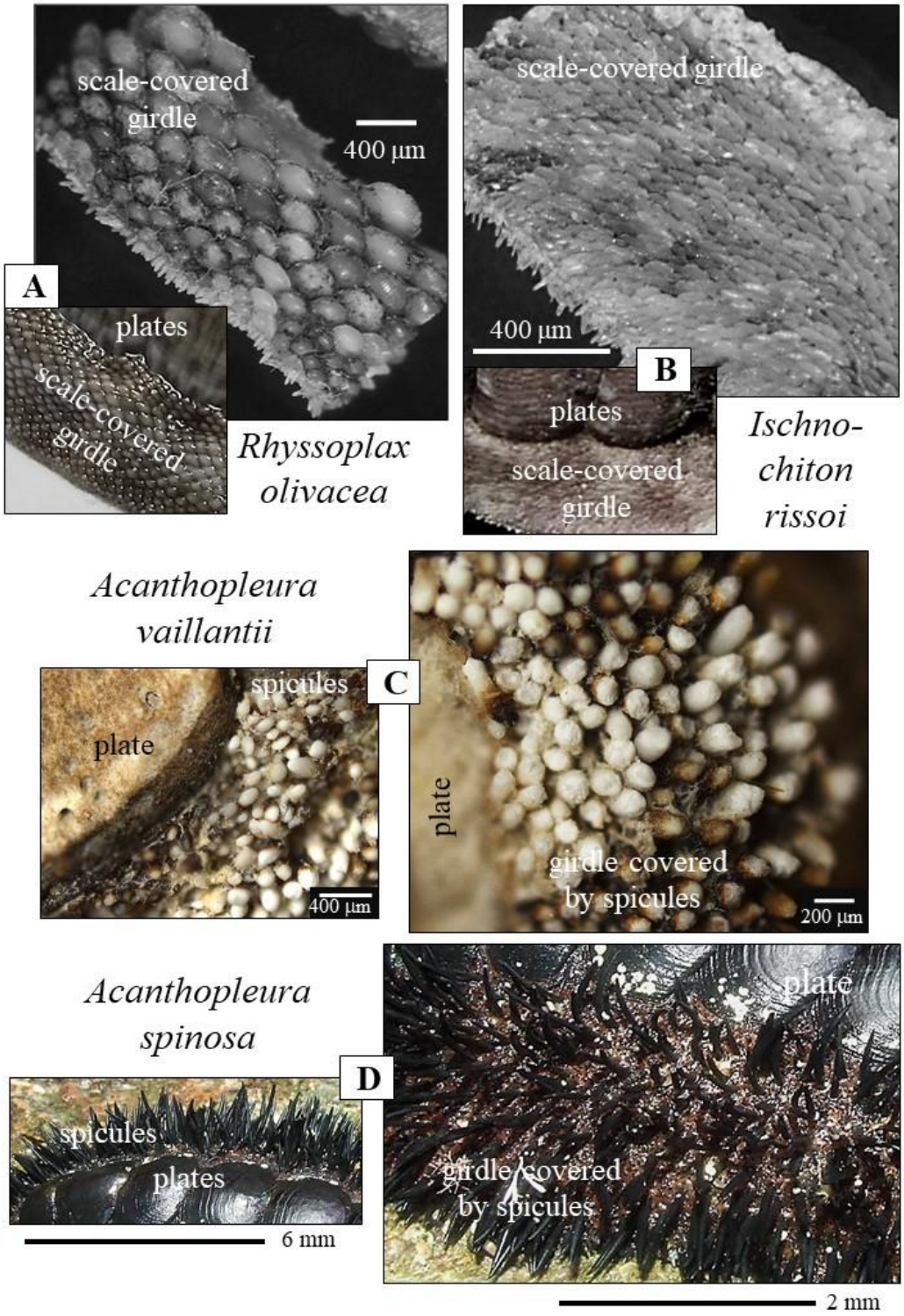


Figure S1. Laser confocal microscopy micrographs depicting the top surface of the girdle of the investigated Polyplacophora species. When viewed from the top, we find for *Rhyssoplax olivacea* and *Ischnochiton rissoi* that scales are next to the plates and that spicules seam only the outermost rim of the girdle (A, B). In contrast, for *Acanthopleura vaillantii* and *Acanthopleura spinosa*, the cuticle adjacent to the plates is covered by spicules only (C, D).

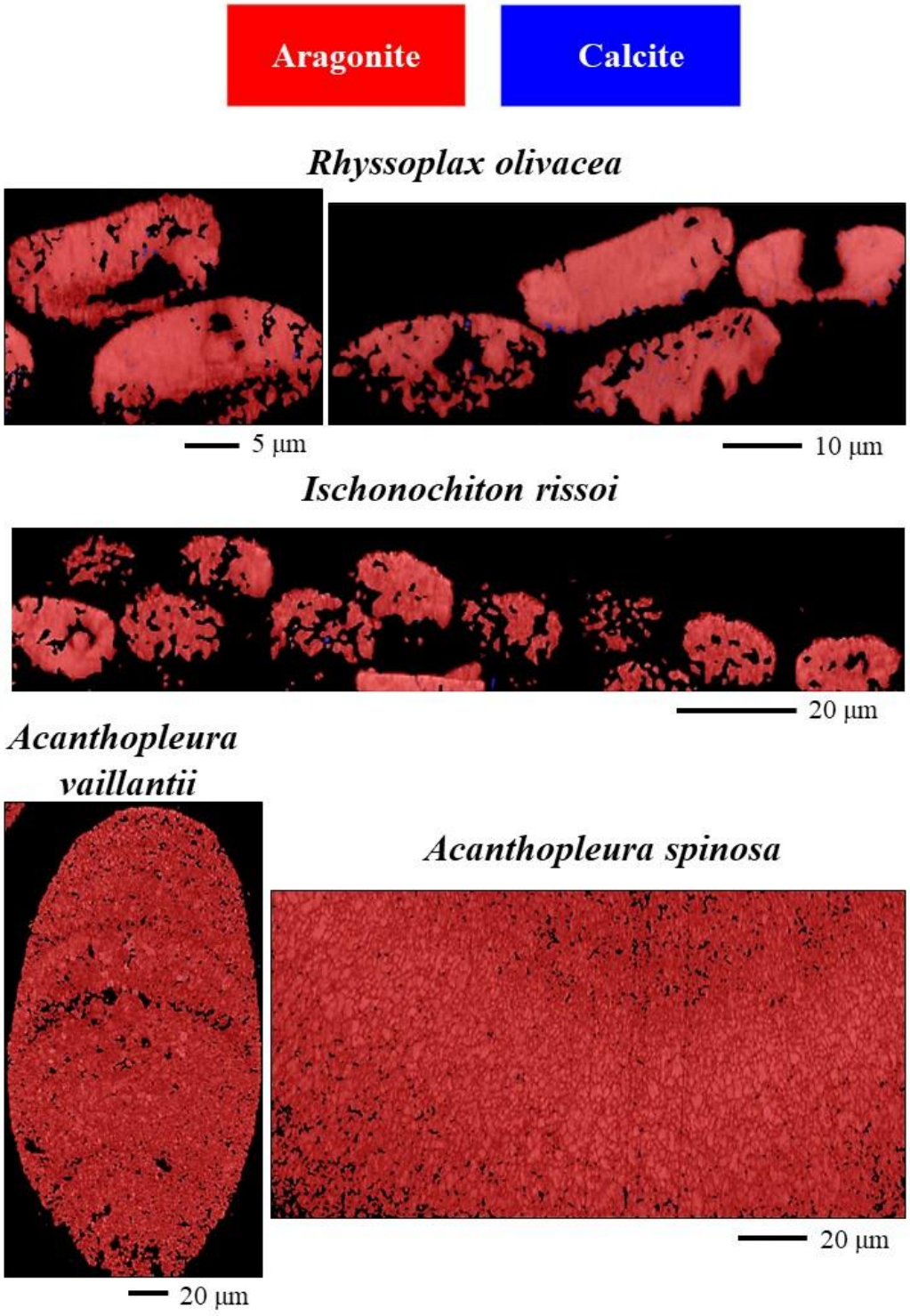


Figure S2. The Ca-carbonate phase that forms the spicules of *R. olivacea*, *I. rissoi*, *A. vailantii* and *A. spinosa*. Red: aragonite, blue: calcite.

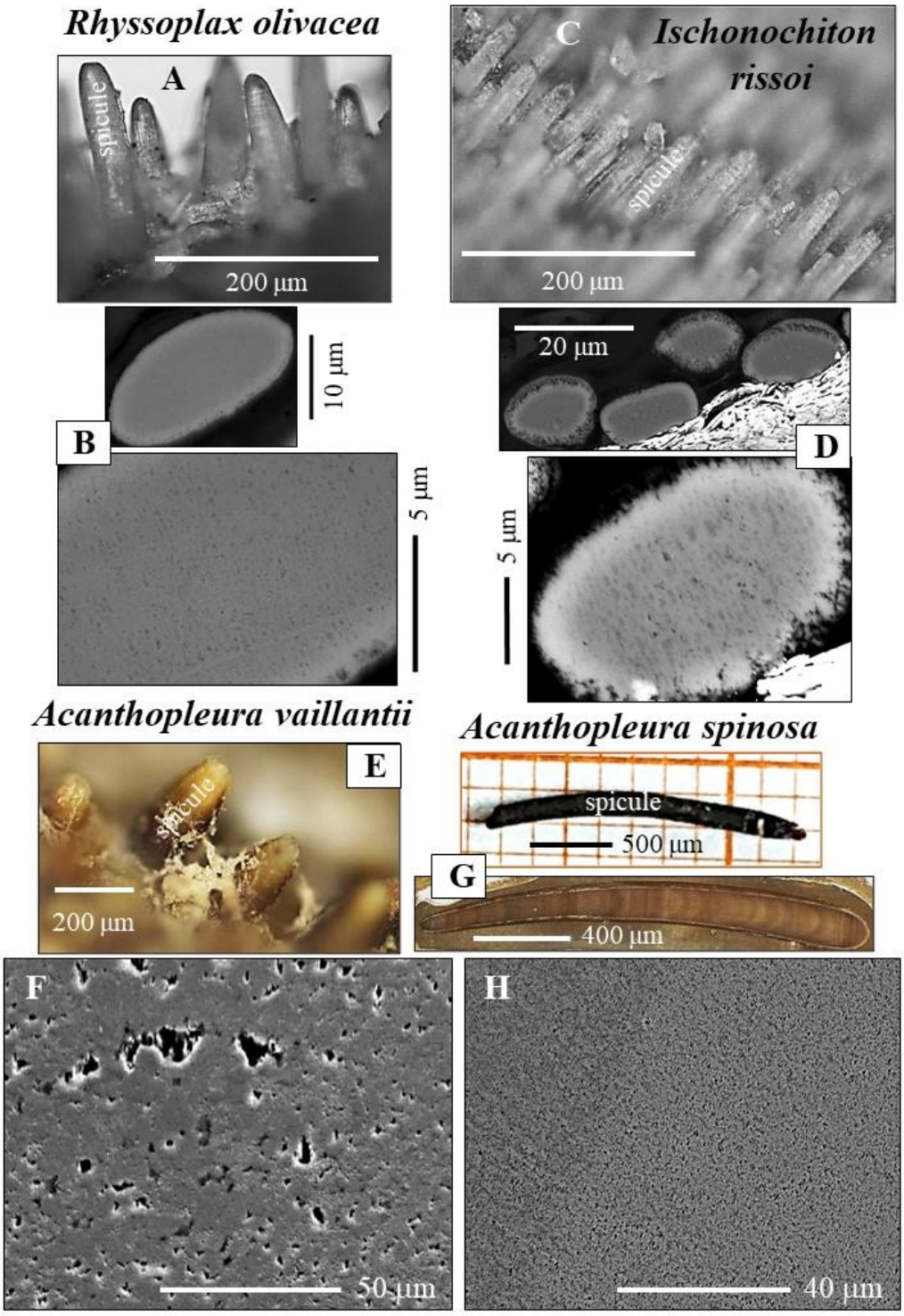


Figure S3. The porous nature of *Rhyssoplax olivacea*, *Ischnochiton rissoi*, *Acanthopleura vaillantii* and *Acanthopleura spinosa* spicules. Laser confocal (A, C, E) and SE images (B, D, F, H) visualizing internal structural features of the spicules. Even though being strongly mineralized, the spicules are porous (B, D, F, H), in particular, the spicules of *Acanthopleura vaillantii*. (G): light microscopy image of a spicule of *Acanthopleura spinosa*; well visible is a banded appearance of the spicule, possibly indicating growth lines and different episodes of growth.

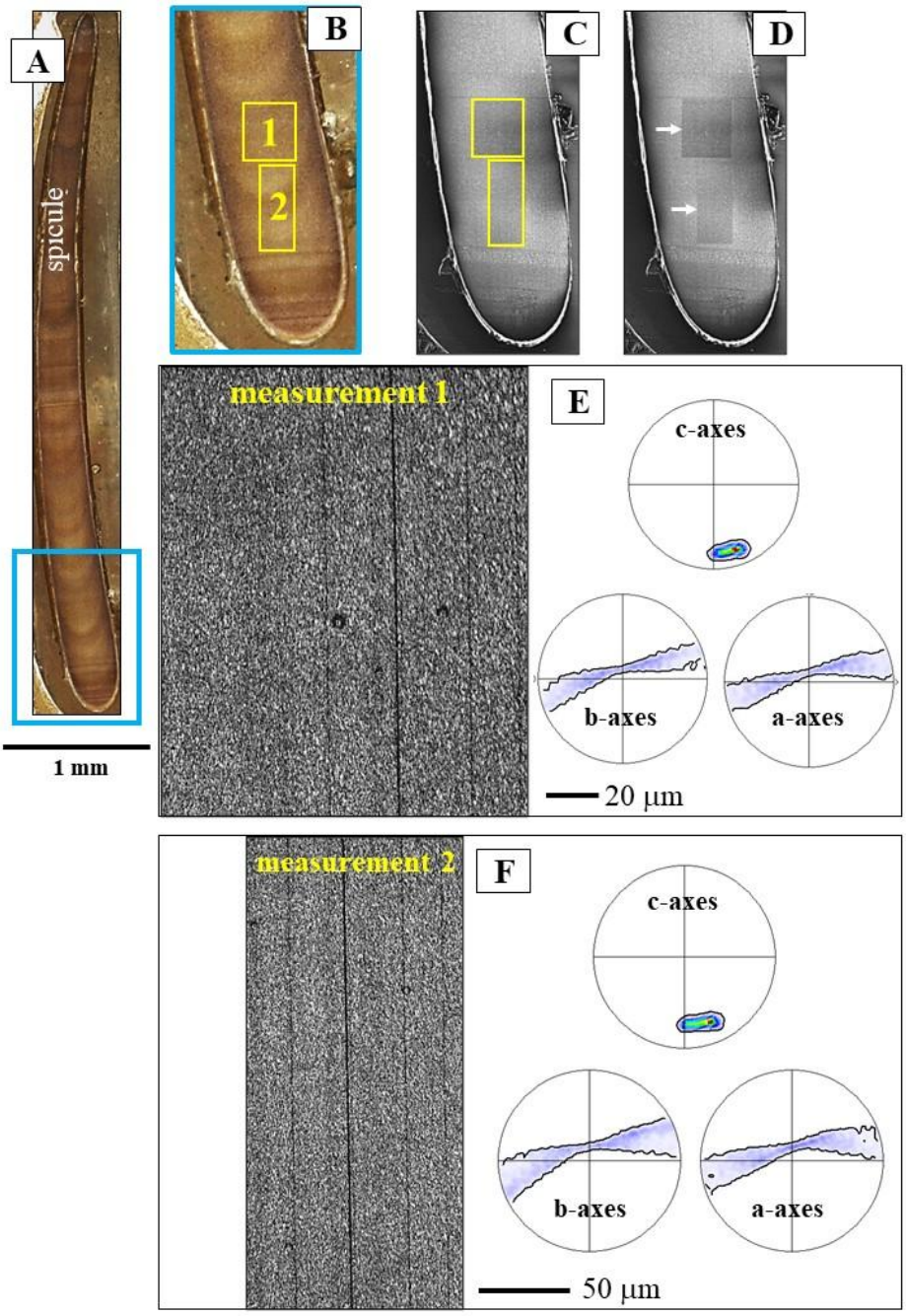


Figure S4. The banded nature of *Acanthopleura vaillantii* spicules (A). EBSD scans (B, C, D, E, F) do not show any structural or texture difference for a scan performed on a darker or lighter spicule region (B, E, F). The blue rectangle in (A) indicates the spicule portion shown in (B). The yellow rectangles in (B) and (C) indicate the positions of the two EBSD measurements conducted on the spicule. White arrows in (D) point to the scanned regions; the micrograph shown in (D) was taken subsequent to the EBSD measurement. Measurement 2 scans over lighter and darker bands, measurement 1 scans predominantly over a lighter, not banded, spicule portion. (A) and (B) are laser confocal microscopy, (C) and (D) are BSE images. (E) and (F) show EBSD band contrast measurement images together with pole figures, the latter depicting aragonite crystallographic axes orientations of measurements 1 and 2 (B). We do not see any structural difference for the observed bands (A, B), or for lighter or the darker spicule regions.

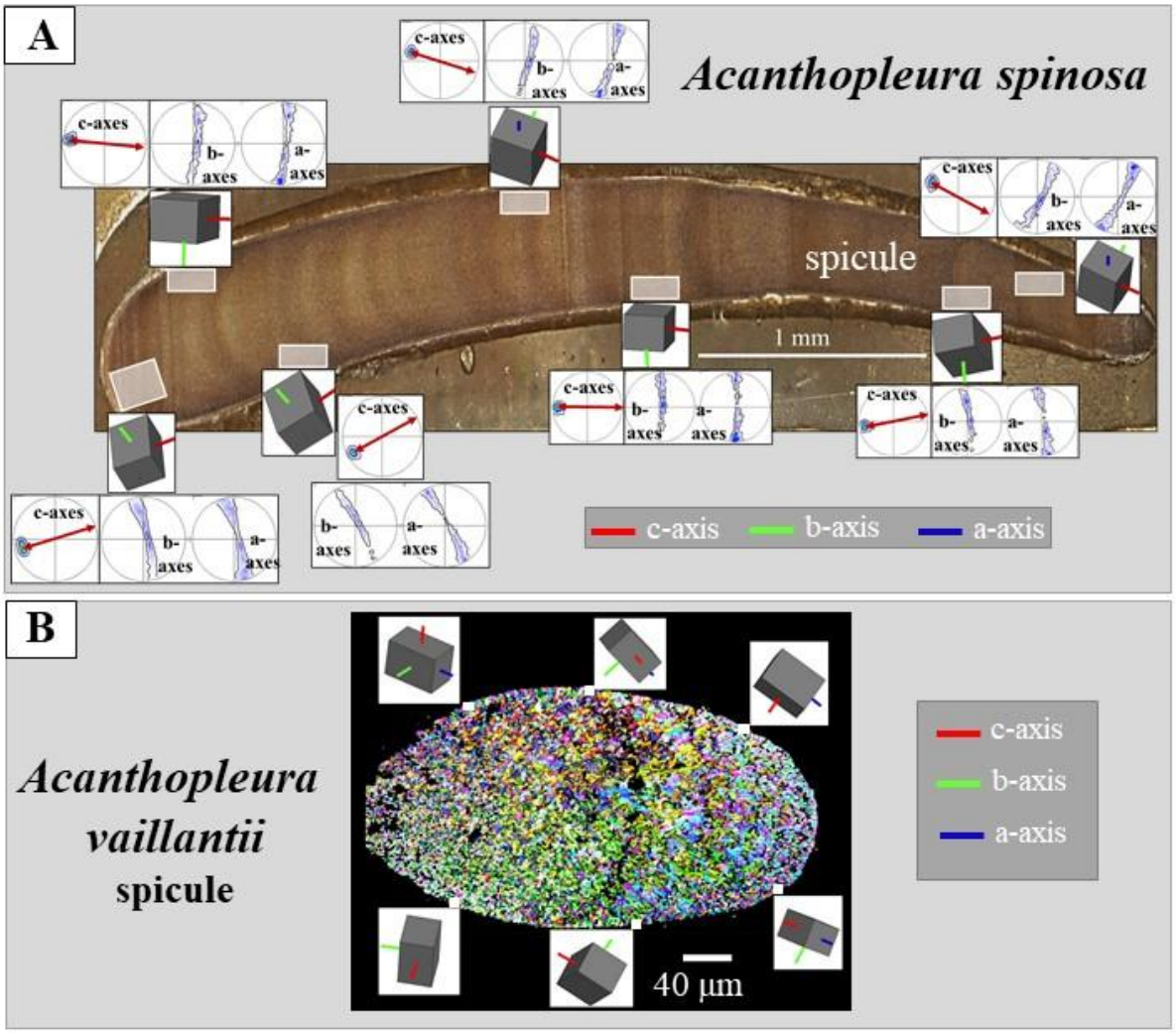


Figure S5. Difference in mode of crystal organization and aragonite c-axis orientation between *Acanthopleura spinosa* (A) and *Acanthopleura vaillantii* (B) spicules. The spicules are cut longitudinally. Crystal orientation in *Acanthopleura vaillantii* spicules is random, hence, aragonite c-axes orientation within the spicules is almost random (B). In contrast, the crystals that form *Acanthopleura spinosa* spicules are assembled with an axial texture, aragonite c-axes orientations are directional. However, aragonite c-axes are always at an angle to the outer surface of the spicule and rotate with its curvature. (A): Laser confocal microscopy image. (B): EBSD measurement depicting crystal orientation in color; the used color-code is given in Fig. 7C.

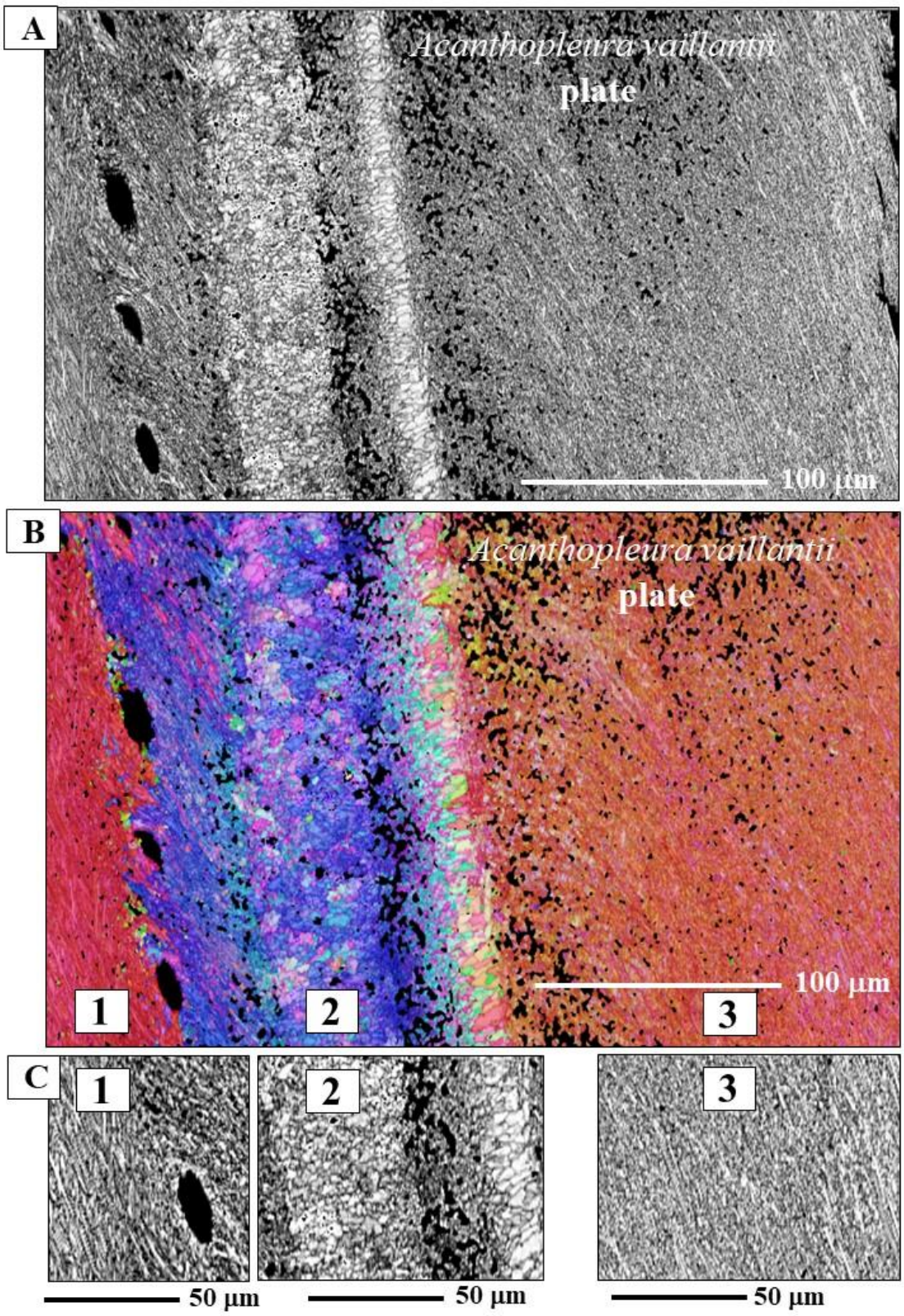


Figure S6. EBSD scan, taken over the entire cross-section of a plate of *Acanthopleura vaillantii*. Enlargement of the map shown in Fig. 10A, B. (A and C): EBSD band contrast measurement image, (B): crystal orientation given color-coded; the used color-code is shown in Fig. 10B. Numbers 1 to 3 indicate different regions of the plate: 1 equals plate position 1 in Fig. 10C; 2 equals plate position 2 in Fig. 10D; 3 equals plate position 3 in Fig. 10E. The different regions of the plate show different crystal sizes, orientations, degrees of twinning and presence of other misorientations (see also Fig. 10).