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Supporting Information

Differential biomolecular recognition by synthetic vs. biologically derived components in stone-forming process using 3D microfluidics

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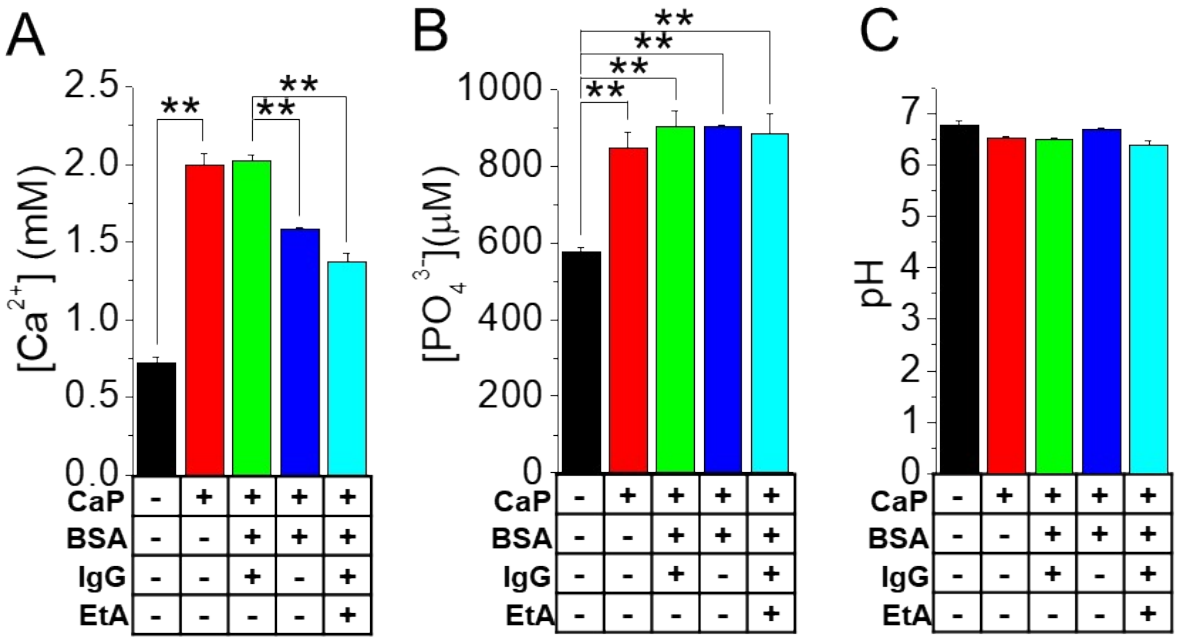

Figure S1. Microfluidic eluent solution electrolyte and pH measurements. Eluent measurements of **A**. Ca^{2+}, **B**. PO_4^{3-}, and **C**. pH measurements were performed after applying calcium phosphate (CaP), Bovine Serum Albumin (BSA), Immunoglobulin G (IgG), and/or Etidronic acid solutions in HK2 cell cultured microfluidic devices. Absence or presence of solution are indicated by – or +, respectively. Representative bar diagrams are in means + SD. **, P<0.01.

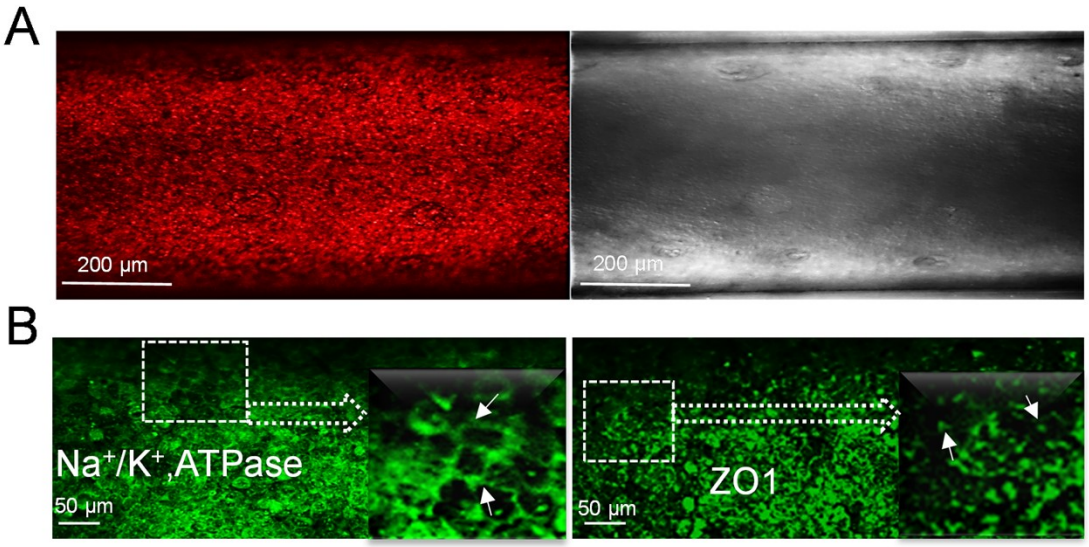

Figure S2. Representative confocal images of polarized HK2 cells cultured within the MF device of **A**. Propidium Iodide staining and corresponding DIC image, and **B**. Na^+/K^+-ATPase and ZO1 immunofluorescence labelling by staining with specific antibodies to those proteins as previously stated[34]. Insets images in **B** were magnified to show the spatial distributions of Na^+/K^+-ATPase and ZO1proteins, ensuring the monolayer formation.

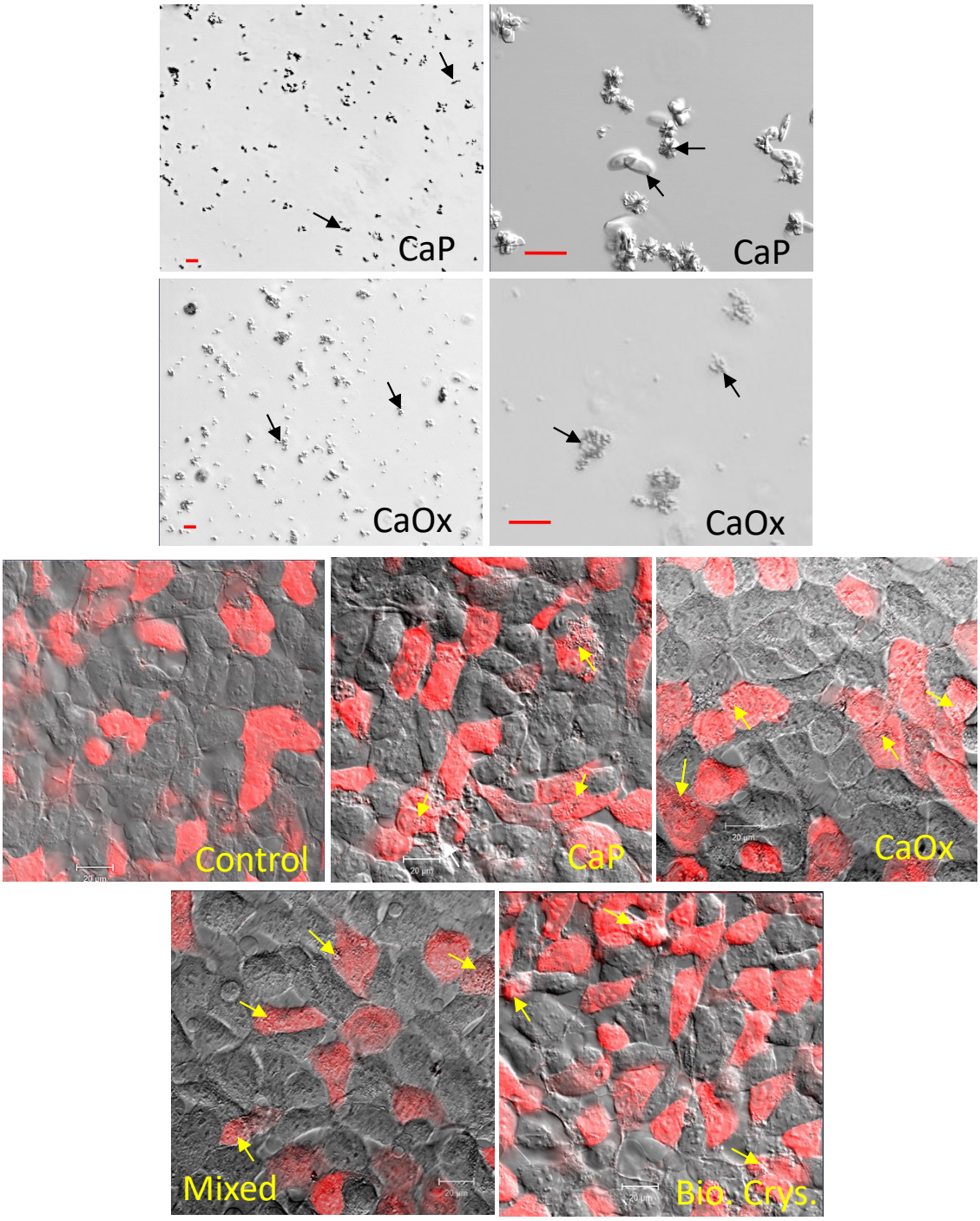

Figure S3. Crystal and cellular images were captured with representative confocal images. DIC images of **A**. calcium oxalate (CaOx) and **B**. calcium phosphate (CaP) synthetic crystals formed in 1x HBSS. Black arrows indicate respective crystals. Scale bars = 5 µm. DIC images of cultured HK2 cellular monolayer in **C**. Control and **D**. CaP, CaOx and Mixed crystal internalized conditions were captured. Cultured HK2 polarized monolayer was counterstained with wheat germ agglutinin

(WGA) Alexa Fluor 594 Conjugate (Thermo-Fisher Scientific) to illuminate the cytoplasm for contrasting to show that crystals are internalized inside the cells in **E**. Control (Control + WGA), **F**. CaP (CaP + WGA), **G**. CaOx (CaOx + WGA), **H**. Mixed (Mixed + WGA) crystal internalization conditions. Black arrows indicate internalized crystals. Scale bars = 20 μm.

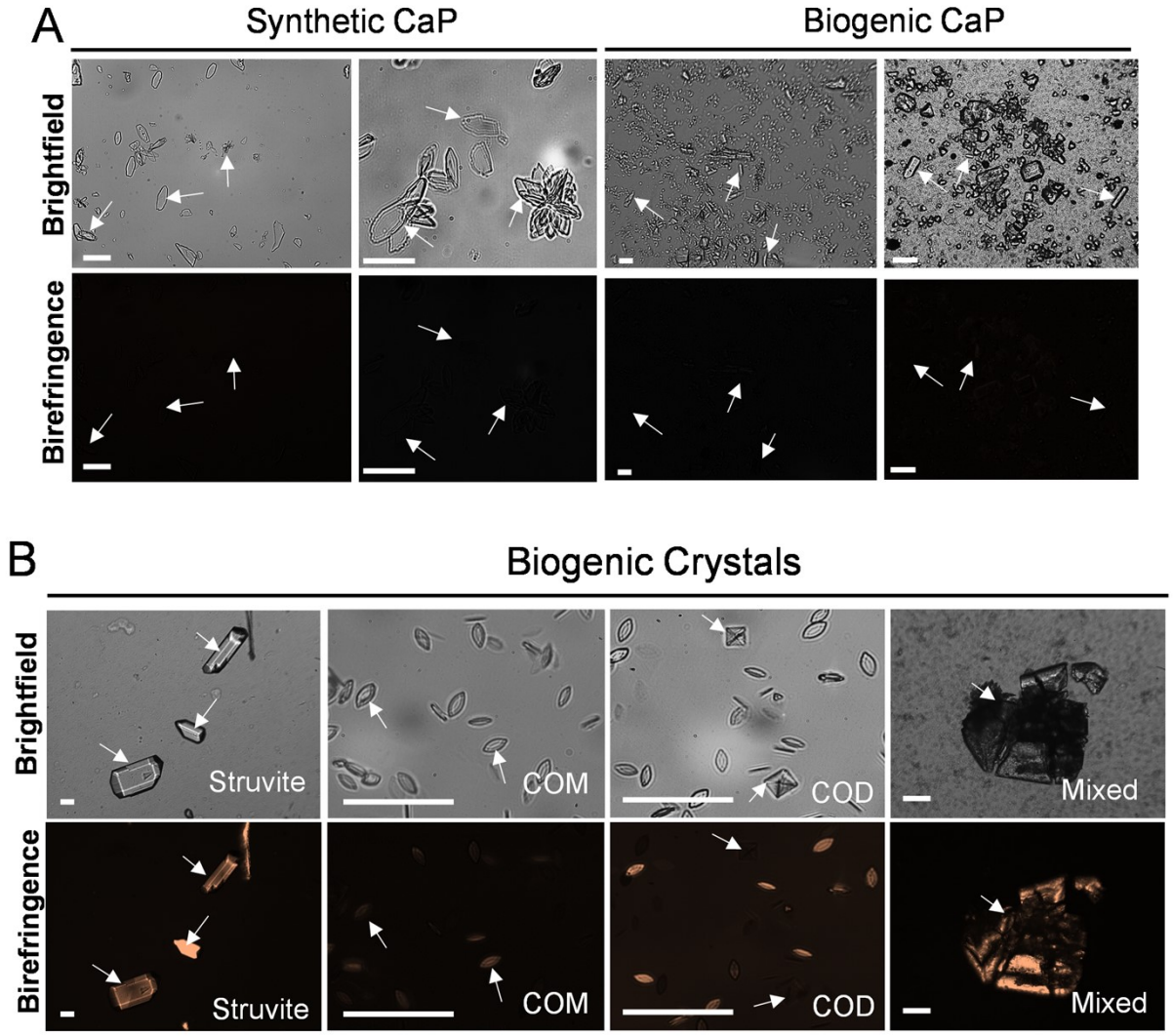

Figure S4. **A**. Brightfield and birefringence images of synthetic and biogenic calcium phosphate (CaP) crystals were captured. Brushite crystals were identified with white arrows in **A**. **B**. Struvite, calcium oxalate monohydrate (COM), calcium oxalate dehydrates (COD), and mixed crystal images were captured and identified with white arrows, respectively in **B**. Biogenic CaP, COM,

COD, and Mixed crystal images were obtained from biogenic urine crystals of TRPC3-/- mice treated with CaG. Biogenic struvite crystal images were taken from urine crystals of TRPC3-/- mice treated with 0.08% acetazolamide orally for 4 weeks. Scale bars=12.5 μm.