Electronic Supplementary Information

Sustainable one-pot path to transform seashell waste calcium carbonate to osteoinductive hydroxyapatite micro-nanoparticles

Raquel Fernández-Penas[a], Cristóbal Verdugo-Escamilla[a], Carla Triunfo[b, c], Stefanie Gärtner[d], Annarita D´Urso[e], Francesca Oltolina[e], Antonia Follenzi[e], Gabriele Maoloni[f], Helmut Cölfen[d], Giuseppe Falini[b], Jaime Gómez-Morales[a*]

[a]*Laboratorio de Estudios Cristalográficos, IACT (CSIC-UGR), Avda. Las Palmeras, nº 4, 18100 Armilla (Spain).*

[b]*Department of Chemistry "Giacomo Ciamician", University of Bologna, via F. Selmi 2, 40126 Bologna (Italy).*

[c]*Fano Marine Center, The Inter-Institute Center for Research on Marine Biodiversity, Resources and Biotechnologies, viale Adriatico 1/N 61032 Fano (Italy).*

[d]*Physical Chemistry, Dpt. of Chemistry, University of Konstanz, Universitätsstrasse 10, Box 714, D-78457 Konstanz (Germany).*

[e]*Dipartimento di Scienze della Salute, Università del Piemonte Orientale, "A. Avogadro" Via Solaroli, 17, 28100 Novara (Italy).*

[f]*Plant Ascoli Piceno, Finproject S.p.A.,3100 Ascoli Piceno, Italy*

*Corresponding author: jaime@lec.csic.es (J.G.-M.)

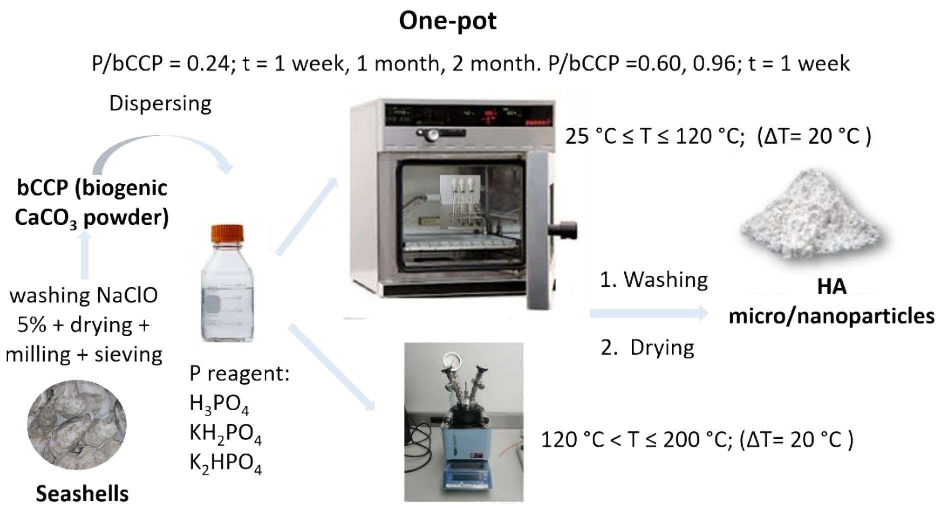

Figure S1. Experimental sketch for the one-pot hydrothermal transformation of seashells calcium carbonate particles (bCCP) to hydroxyapatite micro/nanoparticles.

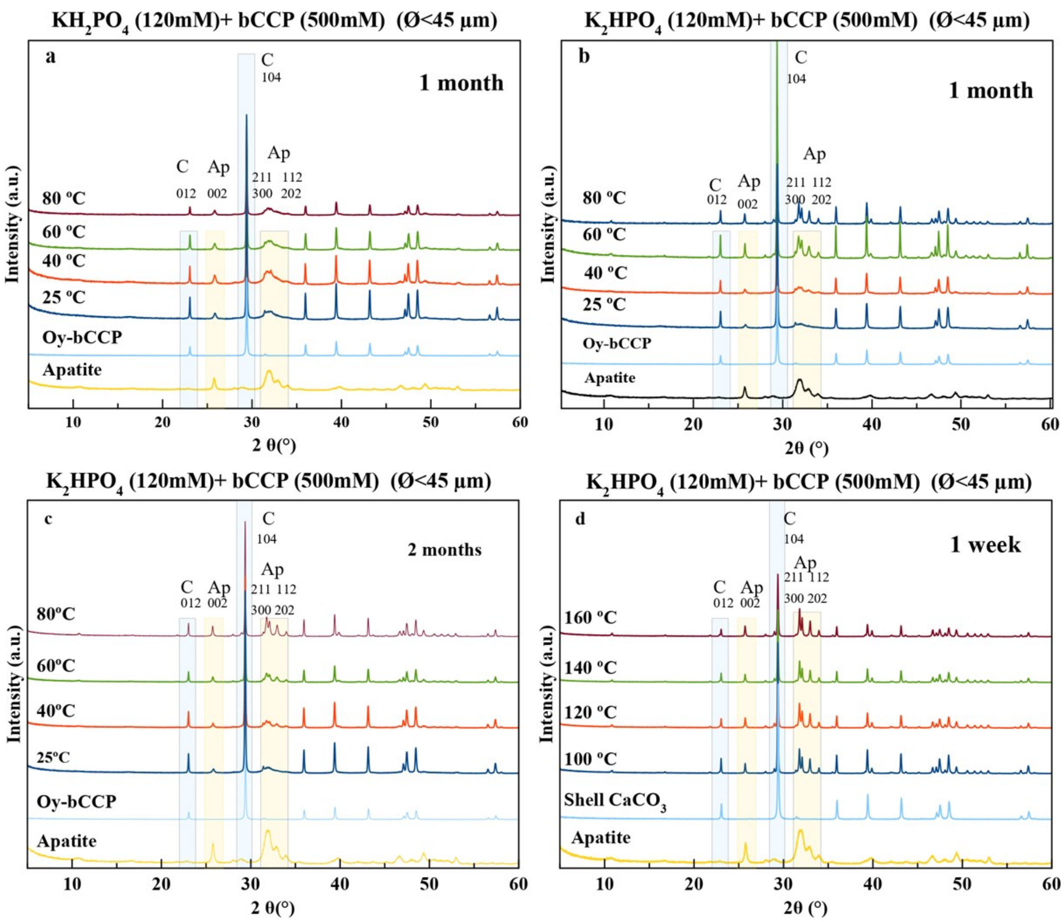

Figure S2. XRD patterns of precipitates obtained from aqueous suspensions with initial P/bCCP ratios 0.24 (defect of P) at 1 month, using KH_2PO_4 and bCCP size fractions Ø<45 µm at temperatures in the range from 25 °C to 80 °C.

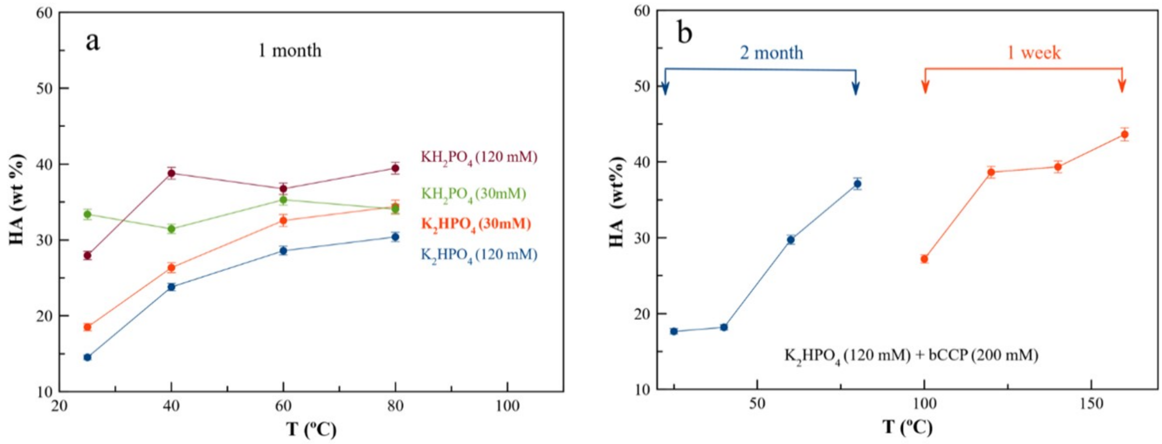

Figure S3. Evolution of apatite wt% with the temperature in experiments with initial P/bCCP ratios 0.24 (defect of P) between 25 °C to 80 °C for 1 month (a), between 25 °C to 80 °C for 2 month and between 100 °C to 180 °C for 1 week (b).

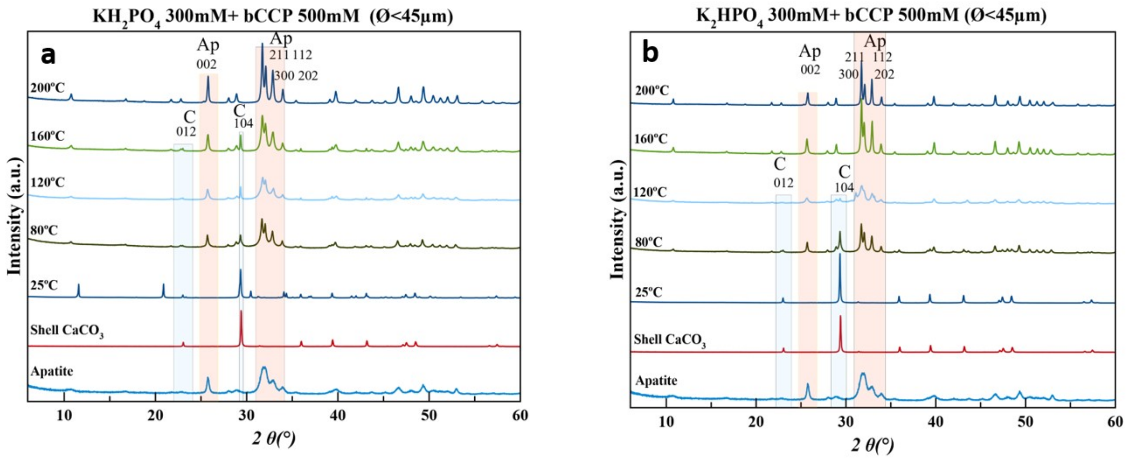

Figure S4. XRD patterns of precipitates obtained from aqueous suspensions with initial P/bCCP ratios 0.6 (stoichiometric composition respect to HA) at 1 week using bCCP size fraction Ø<45 µm and KH_2PO_4 (a) or K_2HPO_4 (b) at different temperatures in the range from 25 °C to 200 °C.

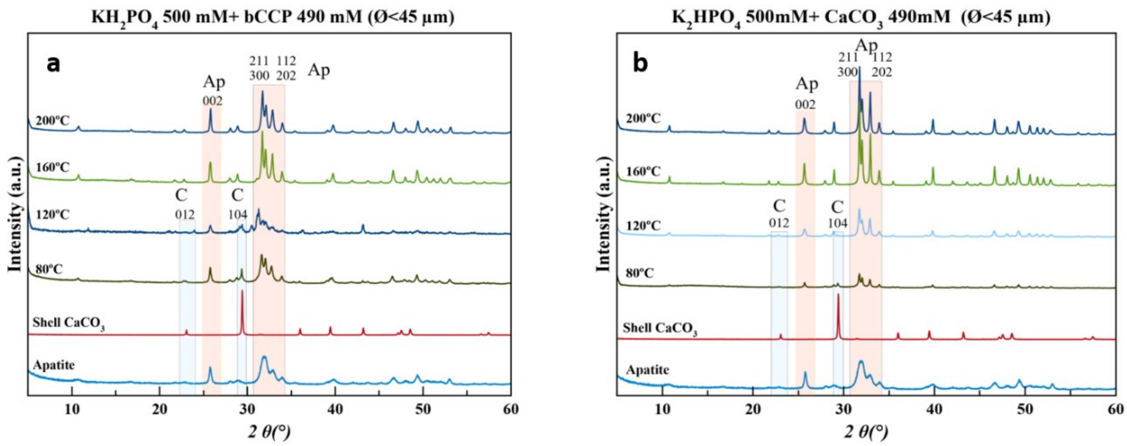

Figure S5. XRD patterns of precipitates obtained from aqueous suspensions with initial P/bCCP ratios 0.96 (excess of P) at 1 week using bCCP size fraction Ø<45 µm and KH_2PO_4 (a) or K_2HPO_4 (b) at different temperatures in the range from 25 °C to 200 °C.

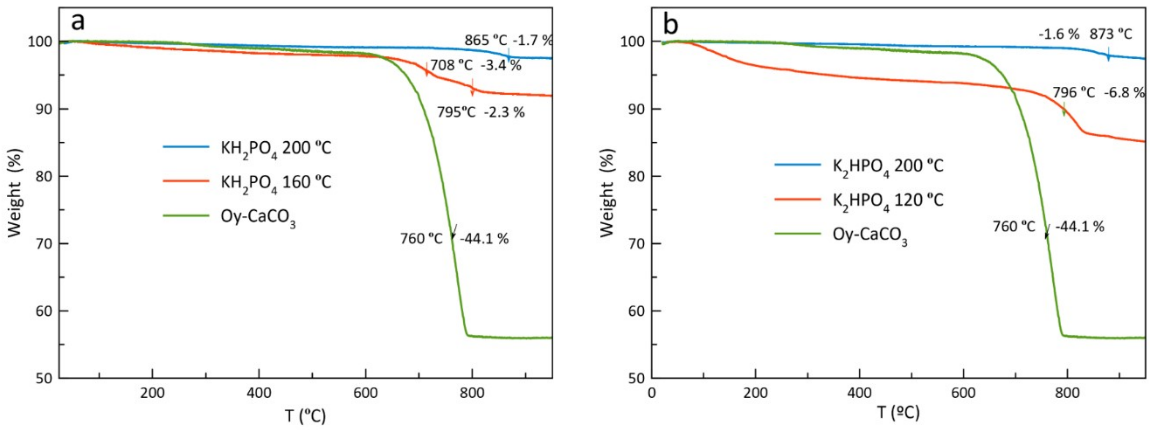

Figure S6. TGA of Oy-$CaCO_3$ and CO_3-apatites obtained by one-step hydrothermal conversion of Oy-$CaCO_3$ at 160 °C and 200 °C using KH_2PO_4 (a) and at 120 °C and 200 °C using K_2HPO_4 (b) at P/$CaCO_3$ molar ratios 0.6. Arrows indicate the temperature of the endothermic peak associated to loss of CO_2 by carbonate decomposition from the CO_3-apatite samples.

Table S1. Initial and final pHs of aqueous suspensions in the experiments using initial KH_2PO_4/bCCP and K_2HPO_4/bCCP ratios 0.6 and 0.96.

	pH_0	25°C	40°C	60°C	80°C	100°C	120°C	140°C	160°C	180°C	200°C
KH_2PO_4/bCCP (0.6)	5.64	8.51	7.17	9.55	8.54	9.41	10.76	9.12	9.27	9.58	9.97
K_2HPO_4/bCCP (0.6)	8.68	9.20	9.45	9.62	9.66	10.26	10.65	9.37	9.69	9.80	9.94
KH_2PO_4/bCCP (0.96)	5.82	8.85	7.68	8.05	7.08	10.96	7.39	8.52	7.14	9.56	9.95
K_2HPO_4/bCCP (0.96)	8.51	9.18	9.28	6.67	10.12	9.62	9.46	9.45	9.27	9.56	9.71

pH accuracy ≤ 0.02 pH

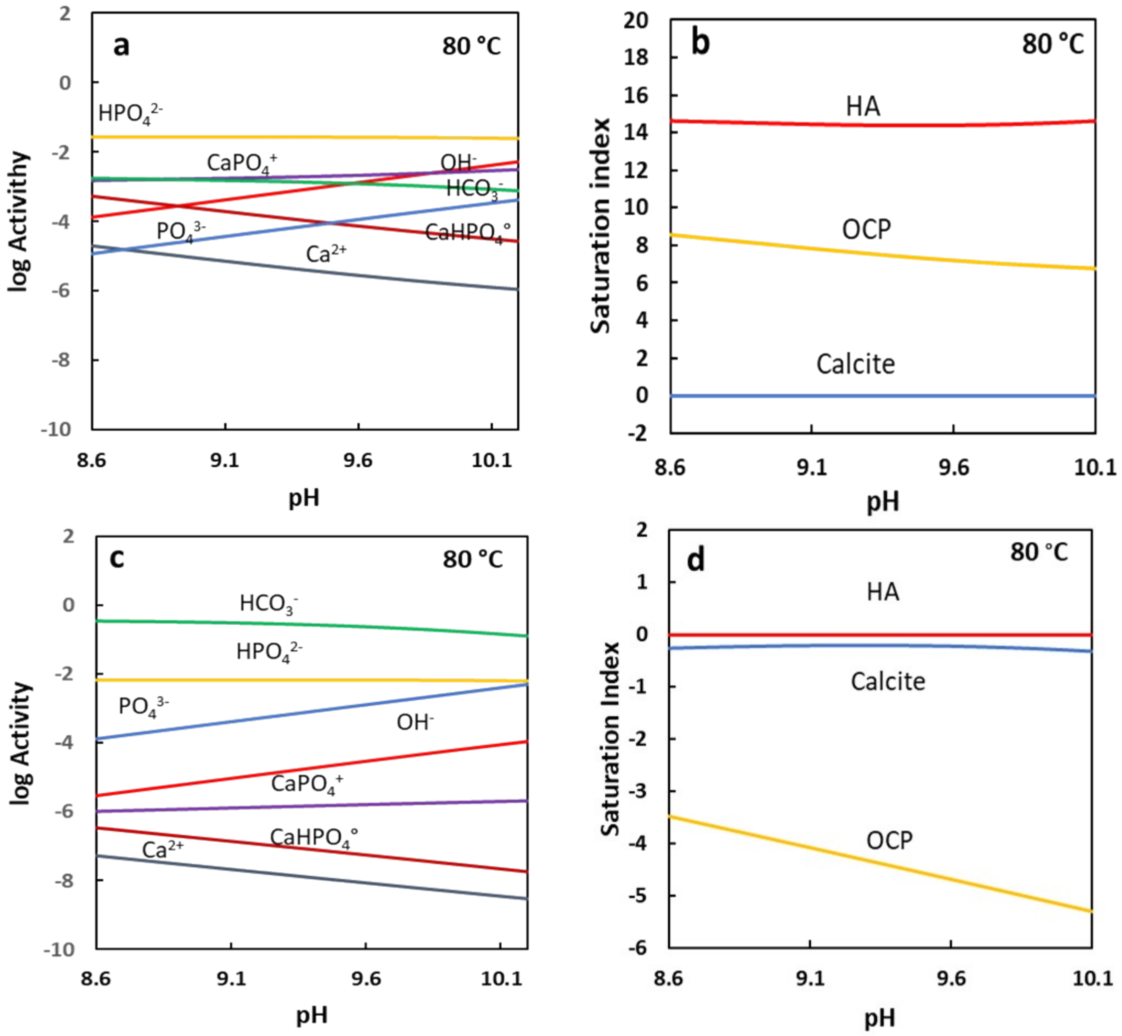

Figure S7. Simulation results with Visual MinteQ 3.1 [1] of the experiment P/$CaCO_3$ 0.96 using K_2HPO_4 as P reagent in which full conversion was obtained at 80 °C. (a) Speciation vs pH considering calcite as a finite solid; (b) Saturation indexes (S.I.) vs pH of hydroxyapatite (HA), octacalcium phosphate (OCP) and $CaCO_3$ (calcite) considering calcite at finite solid (S.I.=0); (c) speciation vs pH after full conversion giving rise to calcite=0 mol/l and HA=0.1 mol/l; (d) Saturation indexes vs pH of HA, OCP and calcite, showing the mother solution in equilibrium with HA (S.I.=0), and subsaturated respect to calcite and OCP (S.I. < 0). In the simulation the “activity” units are mol/l and S.I.= log IAP-log K_{sp}, being IAP the ionic activity product, and K_{sp} the solubility product.

The simulation shows the evolution of the aqueous activities of Ca^{2+}, HCO_3^-, HPO_4^{3-}, PO_4^{3-}, OH^-, $CaPO_4^+$ and $CaHPO_4^{\circ}$ versus pH (Fig. S6a) as well as of S.I. respect to HA and OCP (Fig. S6b) considering both calcite as the calcium source (S.I.=0) and no precipitation of a new phase. It is shown the slow increase of PO_4^{3-}, OH^- and $CaPO_4^+$ activities with pH while the activity of the other species decreases, and the higher S.I (HA) compared to S.I.(OCP) indicating the preferred precipitation of HA at these pHs. Once the experiment finishes the amount of calcite is 0 and that of HA is 0.1 mol/l (full conversion). The activity of all species in the remaining solution is now generally lower than in Figure S6a expected for HCO_3^- and PO_4^{3-}. Under this condition, the solution remains undersaturated with respect to OCP and calcite, and just saturated (S.I.=0) with respect to HA (Fig. S6d).

1 J. P. Gustafsson, “Visual MINTEQ 3.1, freeware chemical equilibrium model,” 2013., adapted from MINTEQA2 (CEAM, EPA, 1999).

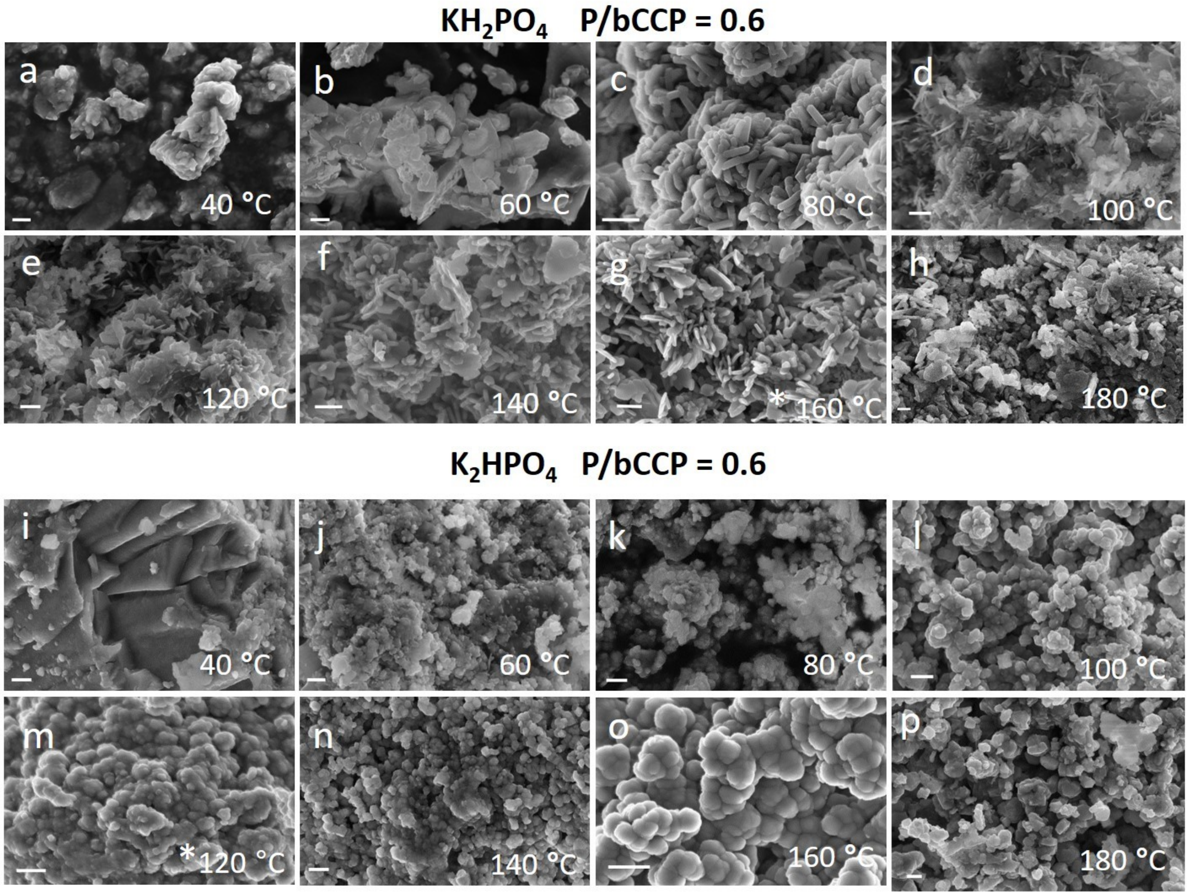

Figure S8. SEM pictures of precipitates obtained at different temperatures (40 °C -180 °C) in experiments of transformation of bCCP to hydroxyapatite in suspensions with stoichiometric composition (P/bCCP=0.6) for 1 week, using KH_2PO_4 (a-h) and K_2HPO_4 (i-p). The (*) denotes the minimum temperature at which the full transformation takes place.