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Nano-Hardness and Elasticity for Hydroxyapatite before and After of Immersing It into Simulated Body Fluid

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Abstract
Stoichiometric and anisotropic nanometer hydroxyapatite was made with co-precipitated method. The powder was compacted under uniaxial and isostatic press and lately sintered. Finally, the tablets were immersing in simulated body fluid during 1 day. The material loss hardness (68.20%), elasticity (64.93%) and plastic work (9.20%) only increasing elasticity work (30.06%).
Keywords: *Nanoindentation test, hardness, elasticity, hydroxyapatite, simulated body fluid.*

1. Introduction

The nano-indentation is a popular technique of surface analysis. Tests can be performed at variable displacements so that properties can be determined as a function changing substrate effect. Kumar & Wang have reported for αTCP(alpha- tricalcium phosphate), in house hydroxyapatite and spherical hydroxyapatite using nano-indentation test a hardness and elasticity of 2.56-6.76GPa and 71.2-122GPa, respectively, applying a force of 300mN[1].

Wang & Shaw reported inverse relation between hardness and grain size using ~50nm x ~15nm nano-rod synthesized hydroxyapatite via the wet precipitation process (surface 120m2/g, BET method). For example, sintering 850°C, grain size is 67nm and Knoop hardness is 4.3 GPa[2].

Other hand, Sun *et al* [3] have argued that HA powder without sintering immerged into simulated body fluid (SBF) suffers dissolution process and little precipitation while HA powder with sintering (T>900°C) immerged into SBF promotes the ability of bone-like apatite formation on surface.

Kanan *et al* [4] sintered bars after pressing hydroxyapatite powders HAP (1200°C), S-HAP1 and S-HAP2 at 1400°C. Reported Vickers hardness are 297.45 ± 9.53, 376.21 ± 8.18 and 414.65 ± 8.91. Synthesis of hydroxyapatites (HAP) with co-substituted essential trace elements (Na, Mg, K, Cl, and F) of natural bone was performed through aqueous precipitation. S-HAP1 with (Ca + Na + Mg + K)/P = 1.86 and S-HAP2 with 1.76. All powders with Ca/P = 1.67. Uniaxial pressing (80MPa). After debinding at 550°C for 2hrs, the bars were sintered at different temperatures, according to the phase stability, for 2h at a heating rate of 5°C/min.

Fathia *et al* [5] sintered at 600°C carbonated HA powder with low-crystallinity (20-30nm) via a simple sol-gel method. This HA showed dissolution/ resorption process into SBF like apatite-bone.

2. Experimental procedure

Hydroxyapatite was synthesized by co-precipitation method. A rod-like morphology with particle size of 20-50 nm x 100-200 nm, particle average size of 175.9nm with 1-modal, pore radii of 1.22nm and stoichiometric and anisotropic hydroxyapatite was

prepared by reacting calcium hydroxide and a phosphoric acid solution. Hydroxyapatite powder[6] was treated with heat at 680°C during 3 hrs in Ar atmosphere (HAcc). HAcc was submitted to uniaxial and isostatic compaction. The uniaxial press was a Daniels of Stroud Stryd glos No. 714987 RAM DIA 6". 5 g HAcc powder were compacted under 30Kgf/cm^2 during 90 seconds. Dade diameter was 1cm of steel 316L. Isostatic in cold compaction was made with a Yuken Model 334 press applied 400MPa during one cycle of four steps: Step 1: 0-100MPa, step 2: 101-200MPa, step 3: 201-300MPa and step 4: 301-400MPa. Time of step is 30 seconds.

Late, the tablets of diameter 1cm and thickness 5mm are sintered at 850°C in Ar atmosphere since 500°C during 1hr, heat up slop 5°C/min and heat slope down 6°C/min.

Finally, the tablets were immersed into SBF 1 day.

Hardness and elasticity measure was used CSM Instruments and software Indentation 4.16 applied to data of 24 zones or areas following random straight paths before immerge HAcc tablets. Each sample with 2mm of separation across tablet center, charge of 100mN and pauses each 10s, rates of charge and discharge of 200mN/min. Analogously, 14 zones for HAcc after immerged in SBF. The SBF was made following the method of Oyane A., *et al* [7].

Indentation software uses Power Law Method or Oliver & Pharr method (O&P). It recognizes the fact that the first portion of the unloading curve may not be linear, and can be described by a simple power law relationship[8].

3. Result and analysis

Nano-indentation technique was applied on sintered HAccs tablets before and after immerging into SBF. Figure 1 shows the results of the analysis with software Indentation 4.16 applied to data of 24 zones or areas following random straight paths before immerge HAccs tablets. Each sample with 2mm of separation across tablet center, charge of 100mN and pauses each 10s, rates of charge and discharge of 200mN/min.

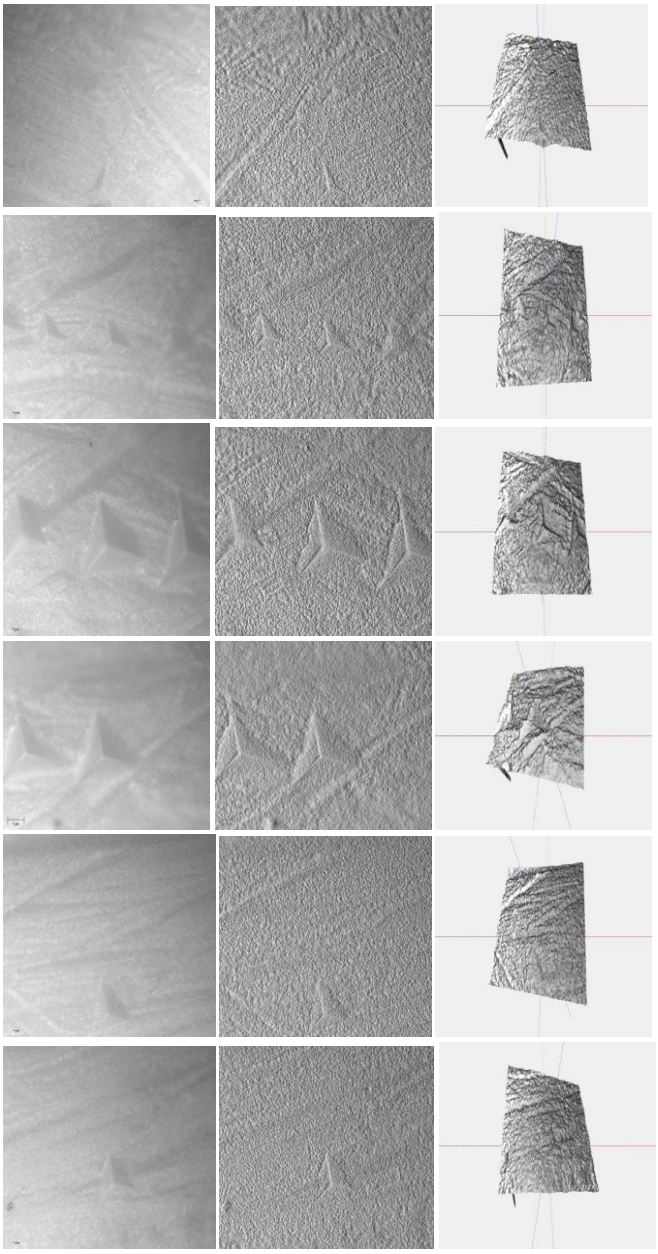

Figure 1: Mapping of the surface of the tablet of sintered HAccs in six zones (codes: 3, 6, 11, 13, 22, 24) in normal way, photo and topographically. It notices the heterogeneous ruggedness of the surface.

Mean for each area was calculated. Late, for each set was calculated mean and standard deviation. Table 1 shows hardness and elasticity for all areas.

Table 1: Hardness and elasticity of sintered HAccs tablet.

HAccs area code	H_{IT} (O&P) MPa	HV (O&P) Vickers	E_{IT} (O&P) GPa	Welast pJ	Wplast pJ
1	2008.458	186.005	50.54	1248.675	4273.559
2	956.482	88.581	37.359	4742.882	18994.57
3	4170.035	386.191	80.578	12870.291	24519.916
4	1331.727	123.332	39.587	14440.784	65849.586
5	1394.079	129.107	41.349	14048.58	53408.273
6	1606.985	148.824	45.757	13742.479	48783.762
7	1512.566	140.08	43.687	13904.679	56567.688
8	1847.749	171.122	49.904	13749.042	49615.945
9	15277.107	1414.826	267.668	7214.993	28864.434
10	26103.078	2417.429	375.298	7250.824	21520.152
11	52429.531	4855.546	743.707	6595.688	15620.01
12	66585.836	6166.573	1085.086	5907.586	13619.011
13	60402.48	5593.927	908.732	6235.431	14690.003
14	2072.877	191.971	57.317	12921.148	38923.176
15	2090.619	193.614	53.51	13595.583	40794.441
16	3265.081	302.382	72.581	12968.737	38214.281
17	1723.455	159.611	47.813	13604.682	52487.344
18	2558.122	236.91	61.727	12987.537	42207.324
19	7144.622	661.67	128.545	9626.568	36695.402
20	1101.377	101.999	34.889	14566.141	63863.68
21	1049.98	97.24	35.052	14096.804	57317.586
22	1160.75	107.498	36.992	14298.34	59683.805
23	1461.02	135.306	42.237	13966.824	51104.266
24	1428.765	132.319	42.679	14038.279	53926.547
average	10861.7825	1005.91929	182.608083	11192.6074	39647.6984
standard desviation	19821.3062	1835.66873	297.099896	3934.54517	17929.3062

As it is observed, the sample is very heterogeneous due to the die marks, but there is a symmetrical local behavior, see figure 2. Then, if data are analyzed for zone sets and taking the average of the averages:

Table 2: Hardness and elasticity of sintered HAccs tablet, Set I (zones 1, 2, 3, 4, 5, 6).

SET I Haccs area code	H_{IT}(O&P) MPa	HV (O&P) Vickers	E_{IT} (O&P) GPa	Welast pJ	Wplast pJ
1	2008.458	186.005	50.54	1248.675	4273.559
2	956.482	88.581	37.359	4742.882	18994.57
3	4170.035	386.191	80.578	12870.291	24519.916
4	1331.727	123.332	39.587	14440.784	65849.586
5	1394.079	129.107	41.349	14048.58	53408.273
6	1606.985	148.824	45.757	13742.479	48783.762
average	1911.294	177.007	49.195	10182.282	35971.611
standard desviation	1159.227	107.357	16.078	5698.770	23600.888

Table 3: Hardness and elasticity of sintered HAccs tablet, Set II. (zones 7, 8, 9, 10, 11, 12, 13, 14, 15, 16).

SET II Haccs area code	H_{IT} (O&P) MPa	HV (O&P) Vickers	E_{IT} (O&P) GPa	Welast pJ	Wplast pJ
7	1512.566	140.08	43.687	13904.679	56567.688
8	1847.749	171.122	49.904	13749.042	49615.945
9	15277.107	1414.826	267.668	7214.993	28864.434
10	26103.078	2417.429	375.298	7250.824	21520.152
11	52429.531	4855.546	743.707	6595.688	15620.01
12	66585.836	6166.573	1085.086	5907.586	13619.011
13	60402.48	5593.927	908.732	6235.431	14690.003
14	2072.877	191.971	57.317	12921.148	38923.176
15	2090.619	193.614	53.51	13595.583	40794.441
16	3265.081	302.382	72.581	12968.737	38214.281
average	23158.692	2144.747	365.749	10034.371	31842.914
standard desviation	26682.349	2471.076	400.956	3611.527	15286.144

Table 4: Hardness and elasticity of sintered HAccs tablet, Set III (17, 18, 19, 20, 21, 22, 23, 14).

SET III Haccs area code	H_{IT} (O&P) MPa	HV (O&P) Vickers	E_{It} (O&P) GPa	Welast pJ	Wplast pJ
17	1723.455	159.611	47.813	13604.682	52487.344
18	2558.122	236.91	61.727	12987.537	42207.324
19	7144.622	661.67	128.545	9626.568	36695.402
20	1101.377	101.999	34.889	14566.141	63863.68
21	1049.98	97.24	35.052	14096.804	57317.586
22	1160.75	107.498	36.992	14298.34	59683.805
23	1461.02	135.306	42.237	13966.824	51104.266
24	1428.765	132.319	42.679	14038.279	53926.547
Average	2203.511	204.069	53.742	13398.147	52160.744
standard desviation	2054.785	190.296	31.474	1596.062	8966.098

From tables II, III and IV:

Center (set II), $H_{IT\text{-}C}$ 23,158.6924 ± 26,682.3485 [MPa], HV_C 2,144.747 ± 2,471.0759 [Vickers], $E_{IT\text{-}C}$ 365.749 ± 400.956 [GPa], $W_{elast\text{-}C}$ 10,034.3711 ± 3611.5270 [pJ] and $W_{plast\text{-}C}$ 31,842.9141 ± 15286.1437 [pJ].

Shores (sets I and III), $H_{IT\text{-}I}$ 1,911.2943 ± 1,159.2265 [MPa], $H_{IT\text{-}III}$ 2,203.5114 ± 2,054.7849 [MPa]; HV_I 177.0067 ± 107.3571 [Vickers], HV_{III} 204.0691 ± 190.2955 [Vickers]; $E_{IT\text{-}I}$ 49.195 ± 16.0778 [GPa], $EI_{T\text{-}III}$ 53.7418 ± 31.4744 [GPa]; $W_{elast\text{-}I}$ 10,182.2818 [pJ] ± 5698.7701, $W_{elast\text{-}III}$ 13,398.1469 [pJ] ±

1,596.0624 and; $W_{plast-I}$ 35,971.611 ± 23,600.8878 [pJ], $W_{plast-III}$ 52,160.7442 ± 8,966.0982 [pJ].
It would give local properties of homogeneity of the compressed and sintered material. The zone of major compression is the center of the tablet diminishing towards the shores. The difference between the center and shores is one order of magnitude for H_{IT}, HV and E_{IT}. W_{elast} is for both center and shores similar. W_{plast} is smaller for center than shores.

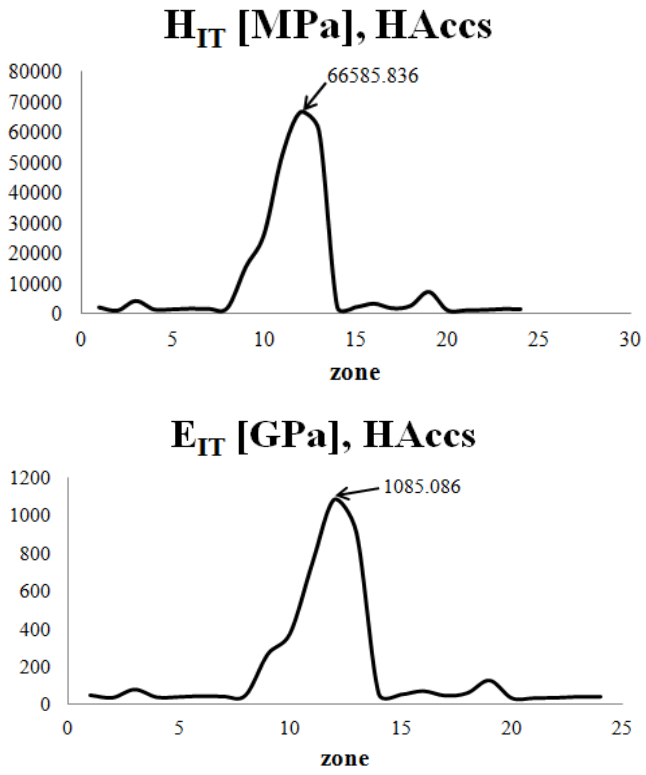

Figure 2: Nano-hardness and elasticity per zones from shore to shore on tablet radial before it was immerged into SBF.

The work or energy of plasticity and elasticity diminish from the shores to the center, which means that the tablet would recover under compression rapidly in the center to do the function of support.

In the other hand, extrapolating Wang & Show hardness curve fitting to HAccs with heat treatment without sintering, HknoopHAccnsW&S = ~4.3GPa (surface 32.53 m^2/g, BET test[6]). Analogously, HAccs without heat treatment and without sintering, $H_{knoop}HAscns^{W\&S}$ = ~4.2GPa (surface 68.665 m^2/g, BET test [6]).

Figure 3 shows mapping of the surface of the tablet of HAccs after immerging into simulated body fluid one day. The surface was scanned into 13 zones from shore to center to opposite shore.

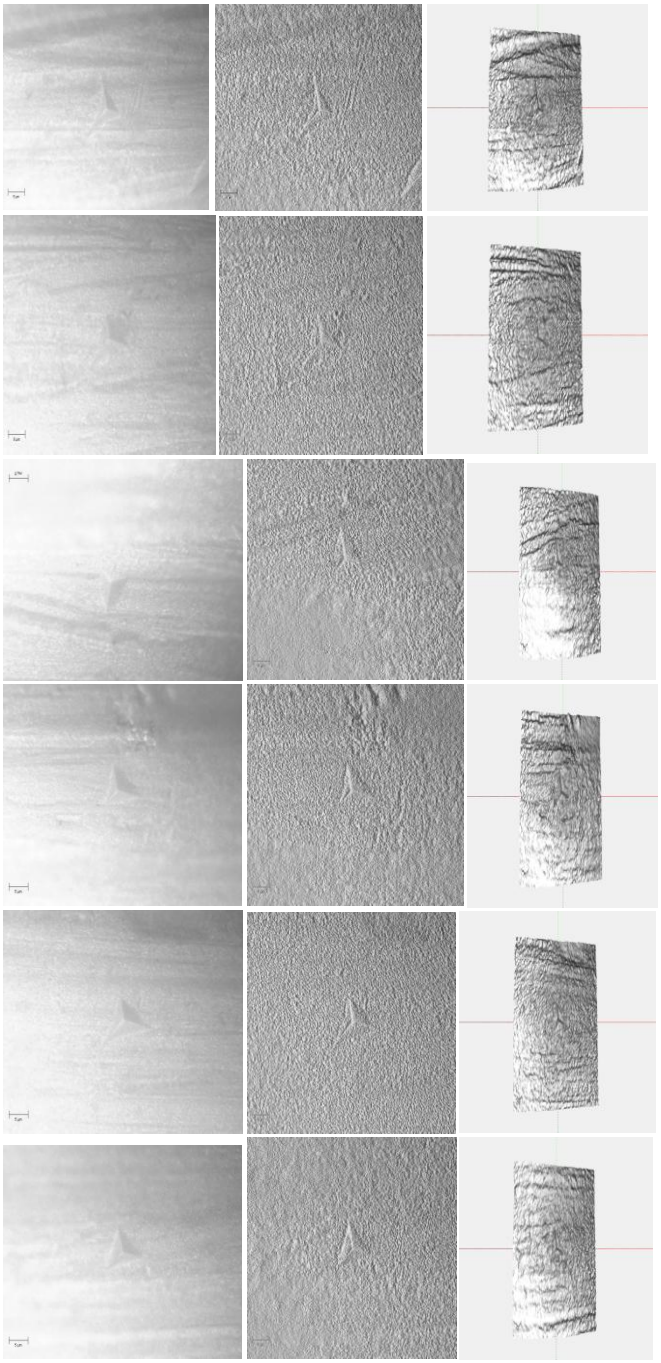

Figure 3: Mapping of the surface of the tablet of sintered HAccs after immerging into simulated body fluid one day in six zones (codes: 1, 4, 8, 10, 12, 13) in normal way, film and topographically. It notices the heterogeneous ruggedness of the surface.

Table 5: Hardness and elasticity of sintered HAccs tablet after immerging into SBF.

HAccs SBF area code	H_{IT} (O&P) MPa	HV (O&P) Vickers	E_{IT} (O&P) GPa	Welast pJ	Wplast pJ
1	2819.556	261.122	58.176	14866.777	34004.063
2	3792.386	351.216	52.566	17945.014	29598.307
3	1273.873	117.975	38.237	14359.133	47973.715
4	1551.869	143.72	36.932	16386.402	48525.012
5	3610.826	334.402	58.526	15680	40040.043
6	4252.036	393.785	74.04	13621.606	31195.736
7	3212.534	297.516	71.976	12180.998	37747.969
8	8564.53	793.169	137.643	11115.193	20634.311
9	2655.72	245.949	54.921	15181.857	36342.941
10	3813.664	353.187	64.489	15454.485	27357.281
11	3110.783	288.092	58.787	14833.493	38325.68
12	2797.192	259.05	63.234	13061.274	41436.785
13	3453.833	319.863	62.909	14557.204	34819.352
average	3454.523	319.927	64.034	14557.187	36000.092
standard desviation	1756.919	162.710	24.648	1781.796	7825.969

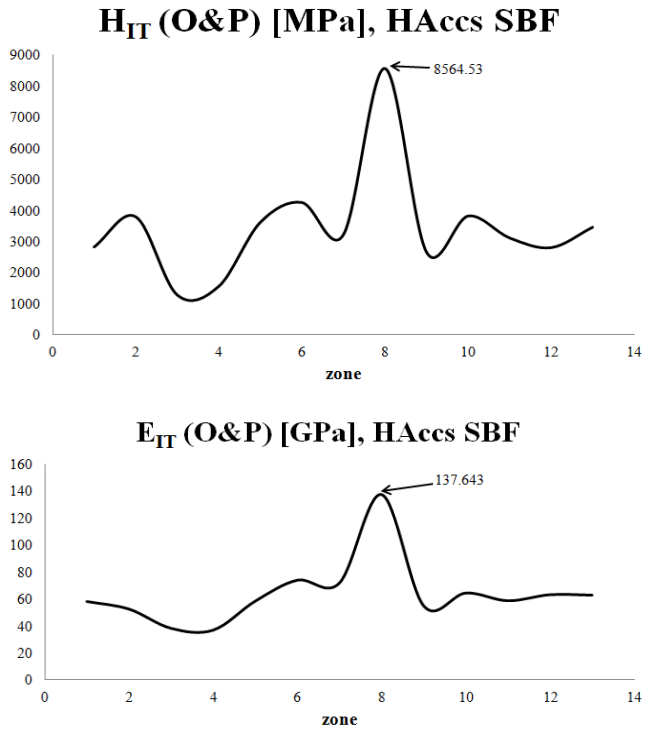

Figure 4: Nano-hardness and elasticity per zones from shore to shore on tablet´s radial after it was immerged into SBF.

Comparing all zones of the tablet before and after it was immerged into SBF: $H_{IT\text{-}before}$ 10,861.7825 ± 19,821.3062 [MPa], $H_{IT\text{-}after}$ 3,454.5232 ± 1,756.9187 [MPa], (%Δ = 31.80%); HV_{before} 1,005.9193 ± 1,835.6687 [Vickers], HV_{after} 319.9266 ± 162.7098 [Vickers], (%Δ = 31.80%); $E_{IT\text{-}before}$ 182.6081 ± 297.0999 [GPa], $E_{IT\text{-}after}$ 64.0335 ± 24.6481 [GPa], (%Δ = 35.07%); $W_{elast\text{-}before}$ 11,192.6074 ± 3,934.5452 [pJ], $W_{elast\text{-}after}$ 14,557.187 ± 1,781.7956 [pJ], (%Δ = 130.06%) and, $W_{plast\text{-}before}$ 39,647.6984 ± 17,929.3062 [pJ], $W_{plast\text{-}after}$ 36,000.092 ± 7,825.9688 [pJ], (%Δ = 90.80%).

H_{IT}, HV, E_{IT} and W_{plast} after tablet was immerged are smaller than before immerging. Inversely, W_{elast} is lightly bigger after immerging than before it. The maximum H_{IT} before and after SBF are 66,585.836MPa and 8,564.53MPa, respectively. Analogously, for E_{IT} 1,085.086GPa and 137.643 GPa. This is because of dissolution/resoption process.

4. Conclusions

Nanometer hydroxyapatite (HAccs) immersed into simulated body fluid one day loss hardness (68.20%) and elasticity (64.93%) only increasing elasticity work (30.06%). HAccs is used as refill material but is not to charge bone substitute. There are 87.14% and 87.31% of decreasing of Hit and E_{IT}, respectively. The Wang & Show hardness curve fitting to grain size less 67 nm is expected non-linear owing to low-crystallinity and carbonated apatitic structure (sintering at 600°C) [5]. Comparing with Kannan´s results the hardness could improve applying cold isostatic compaction and sintering green piece after.

5. Acknoledgements

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References

1. R. Roop Kumar and M. Wang; "Modulus and hardness evaluations of sintered bioceramic powers and functionally graded bioactive composites by nano-indentation technique"; Material Science and Engineering A338, pp. 230-236, 2002.

2. J. Wang & L.L. Shaw; "Nanocrystalline hydroxyapatite with simultaneous enhancements in hardness and toughness", Biomaterials Vol. 30, pp. 6565-6572, 2009.
3. Sun R., Li M., Lu Y, Sun R., Wang A., Immersion behavior of hydroxyapatite (HA) powders before and after sintering, Materials Characterization 56, pp. 250-254, 2006.
4. S. Kannan, A. F. Lemos, and J. M. F. Ferreira; "Synthesis and Mechanical Performance of Biological-like Hydroxyapatites"; Chem. Mater. Vol. 18, pp. 2181-2186, 2006.
5. Fathia M.H., Hanifia A., Mortazavi V., Preparation and bioactivity evaluation of bone-like hydroxyapatite nanopowder, Journal of Materials Processing Technology 2 0 2, pp. 536-542, 2008.
6. D. E. Ledesma-Carrión; *"Modification on the synthesis process of hydroxyapatite"*; Asian Journal of Science and Technology; Vol. 6, Issue 04, pp. 1311-1315, April, 2015; ISSN: 0976-3376.
7. Oyane A., Kim H.-M., Furuya T., Kokubo T., Miyazaki T., Nakamura T., Preparation and assessment of revised simulated body fluids, Journal of Biomedical Materials Research 65A, pp. 188-195, 2003.
8. CSM Instruments; Indentation software User´s manual; version 0.1.0 (2008) pp.94-95. Rue de la Gare 4 CH-2034 Peseux (Switzerland) T +41 32 557 56 00 F +41 32 557 56 10 e-mail: info@csm-instruments.com website: www.csm-instruments.com

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