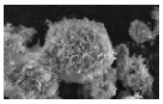
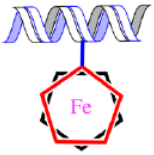
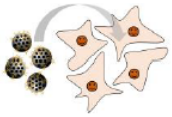
Synergy Symposium for NanoBioMedicine

4.-5.11.2019, Zagreb, Hrvatska

"Influence of albumine on calcium phosphate formation on TiO_2 nanotubes"

Ina Erceg, Atiđa Selmani, Ivana Panžić, Andreja Gajović, Maja Dutour Sikirić



11:20 – 11:30h	Emerik Galić, PhD Student "*In vitro toxicity assessment of chemically synthesized selenium nanoparticles*" (co-authors: Krunoslav Ilić, Tomislav Vinković, Ivana Vinković Vrček, Ivan Pavičić) University J. J. Strossmayer, Faculty of Agrobiotechnical Sciences, Osijek, Croatia
11:30 – 11:40h	Ina Erceg, PhD Student "*Influence of albumine on calcium phosphate formation on TiO_2 nanotubes*" (co-authors: Ivana Panžić, Atiđa Selmani, Andreja Gajović, Maja Dutour Sikirić) Ruđer Bošković Institute, Zagreb, Croatia
11:40 – 11:50h	Mateja Toma, PhD Student "*Organometallic derivatives of DNA bases*" (co-authors: Davor Šakić, Jasmina Lapić, Senka Djaković, Valerije Vrček) Faculty of Pharmacy and Biochemistry, University of Zagreb, Zagreb, Croatia
11:50 – 12:30h	Discussion
12:30h	Cooperative activities

INFLUENCE OF ALBUMINE ON CALCIUM PHOSPHATES FORMATION ON TiO_2 NANOTUBES

Ina Erceg[1], Atiđa Selmani[1], Ivana Panžić[2], Andreja Gajović[2], Maja Dutour Sikirić[1]

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[2]Laboratory for Energy Conversion Materials and Sensors, Division of Materials Physics, Ruđer Boskovic Institute

New implantation materials

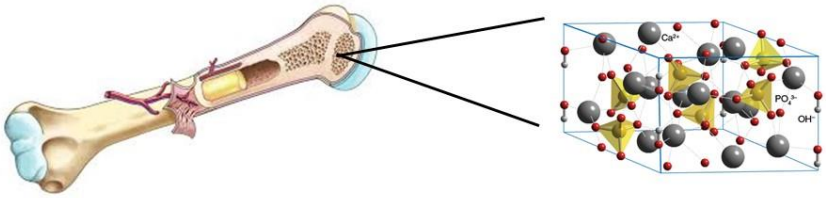
- Hard tissue chronical diseases are among the largest health issues in modern society
- Treatment of such diseases – implantation
- Multifunctional materials
- Calcium phosphate and inorganic nanomaterials nanocomposites

http://www.pamelaegan.com/bone-and-joint-pain
https://www.rheumatologynetwork.com
https://dentaltourism.rs

Moroni, A. Nandakumar, F. B. De Groot, C. A. van Blitterswijk and P. Habibovic, *J. Tissue Eng. Regen. Med.* **9** (2015) 745–759.

New implantation materials

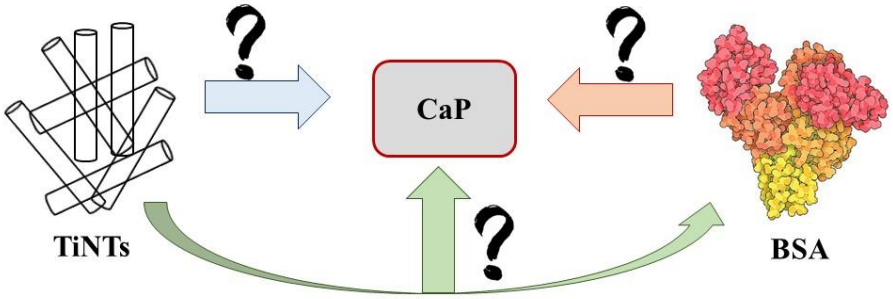
- Calcium Phosphates (CaPs) are main inorganic component of hard tissues
- "Biological apatite" - poorly crystallized nonstoichiometric hydroxyapatite or CaDHA doped with sodium, magnesium or carbonate ions
- CaPs have poor mechanical properties – doping with inorganic nanoparticles
- TiO_2 nanotubes (TiNT) acts as drug delivery systems



C. Canal and M. P. Ginebra, *J. Mech. Behav. Biomed. Mater.* **4** (2011) 1658–1671
E. Beltrán-Partida, et. al., *J. Nanobiotechnology*, **15** (2017) 10-15.

Aim of the study

- Investigation of the effect of TiNT on properties of formed CaP solid phase at conditions closed to physiological in the presence of Bovine Serum Albumine (BSA)
- BSA is model molecule for Human Serum Albumine (HAS)

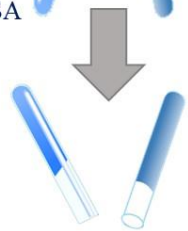


Materials and Methods

Anionic solution
a) Na_2HPO_4
b) Na_2HPO_4 + TiNT
c) Na_2HPO_4 + BSA
d) Na_2HPO_4 + TiNT/BSA

Cationic solution
$CaCl_2 \cdot 2H_2O$

Solution	Concentration
$CaCl_2 \cdot 2H_2O$	4 mmol dm^{-3}
Na_2HPO_4	4 mmol dm^{-3}
TiNT	50 or 100 mg dm^{-3}
BSA	50 or 100 mg dm^{-3}



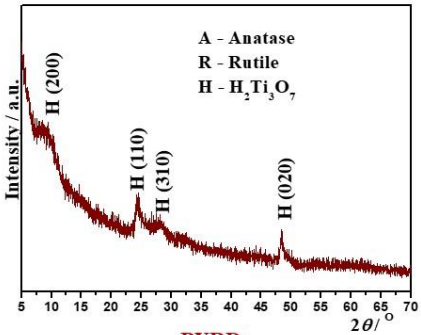
25 °C
$pH_{int} \approx 7.4$
60 min

time

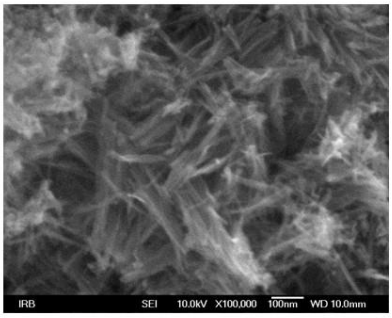
- Potentiometry
- Fourier Transform Infrared Spectroscopy (**FTIR**)
- Powder X-ray diffraction (**PXRD**)
- Scanning Electron Microscopy (**SEM**)
- Transmission Electron Microscopy (**TEM**)

TiNT Synthesys and Characterization

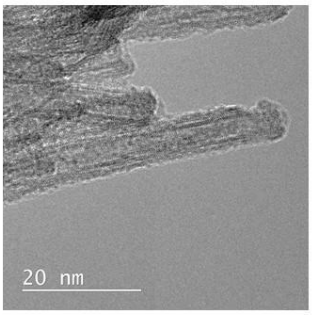
- TiNTs were prepared using a hydrothermal method similar to that described by Kasuga and co-workers



PXRD



SEM



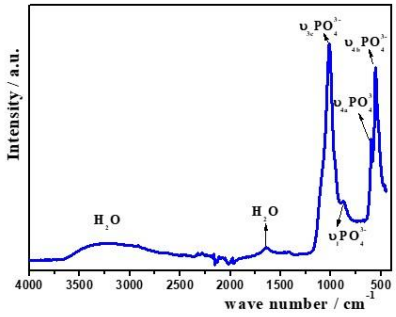
TEM

- TiNTs are mixture of rutile, anatase and $H_2Ti_3O_7$
- Average length 100 nm and diameter 5-10 nm

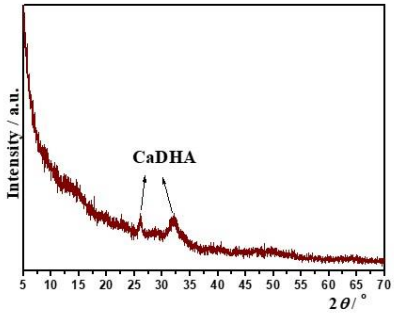
T. Kasuga, M. Hiramatsu, A. Hoson, T. Sekino, K. Niihara, *Adv. Mater.* **15** (1999) 1307.

Control System (CS)

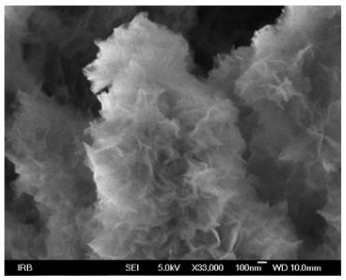
- System without TiNT or BSA: $Na_2HPO_4 + CaCl_2$



FTIR



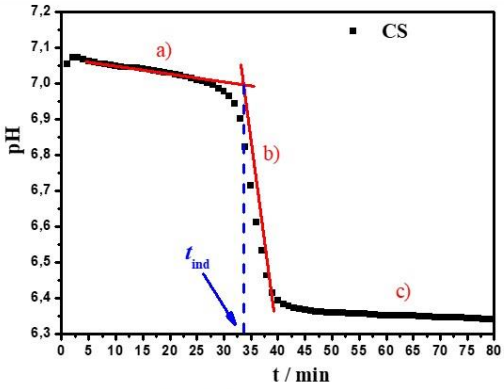
PXRD



SEM

- Calcium deficient hydroxyapatite (CaDHA, $Ca_{10-x}(HPO_4)_x(PO_4)_{6-x}(OH)_{2-x}$, $0 < x < 2$)
- Leaf-like morphology

CaP/TiNT nanocomposites Characterization – Induction time



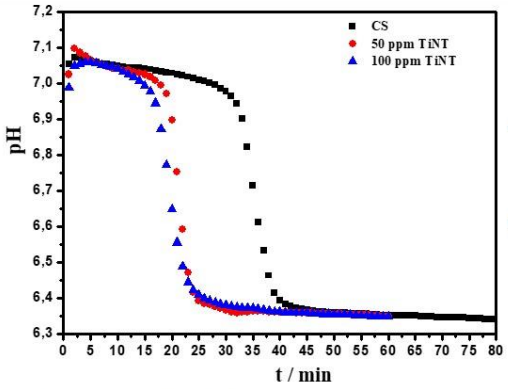
Representative pH vs. time curves

Three stages of curves respond to:
a) Initial amorphous CaP formation during which the changes in pH or calcium concentration are small or absent.
b) Secondary precipitation of crystal phase followed by an abrupt decrease in pH.
c) Crystal growth and transformation associated with slight change in pH value

Induction time of investigated systems

γ / mg L^{-1}	0	50	100	
t_{ind} / min	29.4 ± 3.2			

CaP/TiNT nanocomposites Characterization – Induction time



Representative pH vs. time curves

- The induction time is time passed from initiation of precipitation process until the amorphous phase transformation
- It is determined from intersection of the tangents drawn on first two parts of pH vs. time curve

Induction time of investigated systems

γ / mg L^{-1}	0	50	100	
		17.4 ± 1.0	17.9 ± 2.0	TiNT
t_{ind} / min	29.4 ± 3.2			

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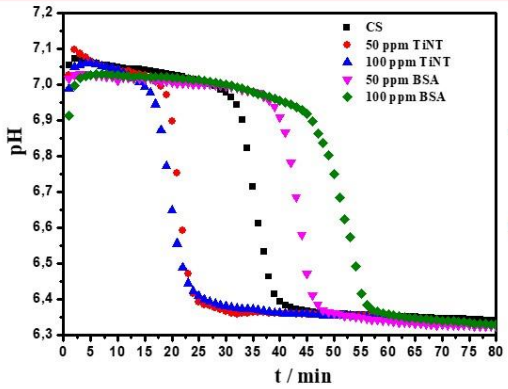

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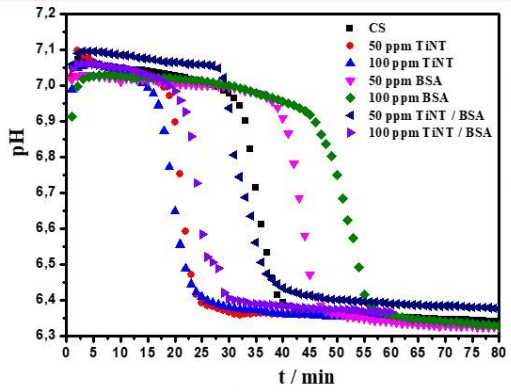
t_{ind} / min	29.4 ± 3.2	39.7 ± 1.0	45.1 ± 0.9	BSA

CaP/TiNT nanocomposites Characterization – Induction time



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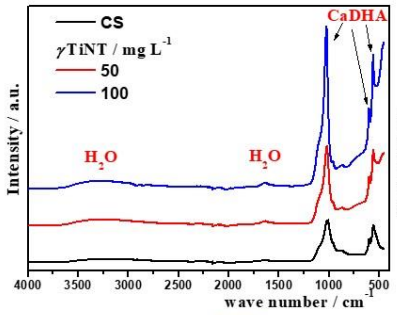
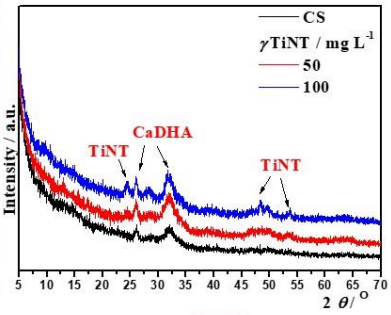
Induction time of investigated systems

γ / mg L^{-1}	0	50	100	
t_{ind} / min	29.4 ± 3.2	17.4 ± 1.0	17.9 ± 2.0	TiNT
		39.7 ± 1.0	45.1 ± 0.9	BSA
		28.2 ± 1.1	20.3 ± 1.5	TiNT/BSA

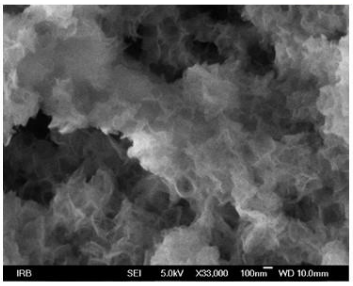
CaP/TiNT nanocomposites characterization



FTIR



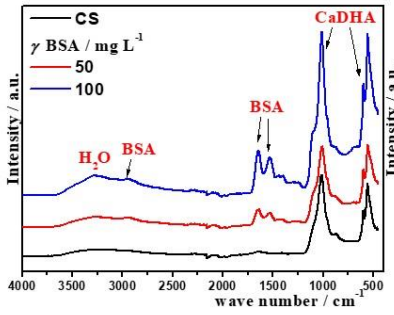
PXRD



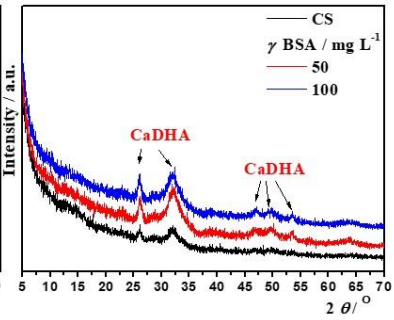
SEM

- TiNT does not influence the composition of precipitated CaP form
- SEM investigation has shown that CaDHA grows on TiNT surface in a linear layout

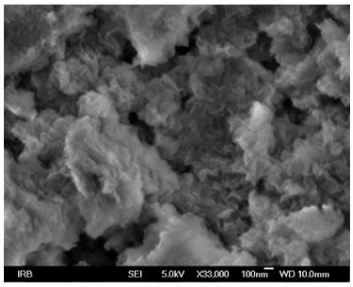
CaP/BSA nanocomposites Characterization



FTIR



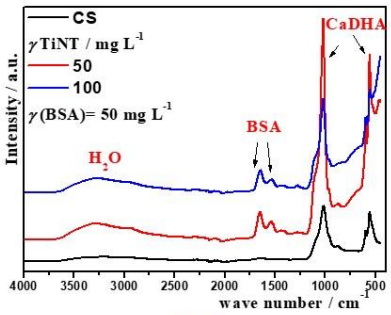
PXRD



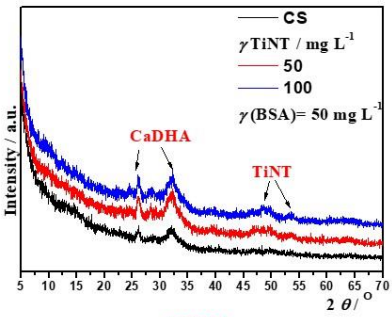
SEM

- BSA does not influence the composition of precipitated CaP form nor the morphology of CaDHA

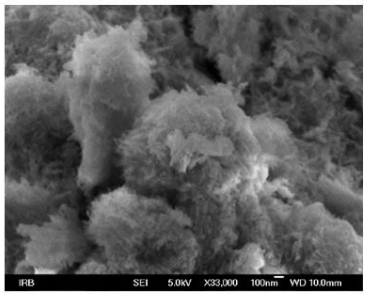
CaP/TiNT nanocomposites in the presence of BSA Characterization



FTIR



PXRD



SEM

- BSA does not influence the composition of precipitated nanocomposites
- In the presence of BSA, the linear layout of CaP on TiNT surface is not observed

To conclude…

- TiNTs act as promotors while BSA acts as inhibitor of CaDHA formation
- Neither of the additives influence the composition of the formed precipitate
- Changes of morphology were observed
- Combined effect of both additives is intermediate between the effects observed in the presence of TiNT or BSA alone
- The obtained results point to a biomimetic preparation route of multifunctional CaP based biomaterials

https://www.everydayhealth.com/arthritis/pain-and-stiffness.aspx

Acknowledgement

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Fabio Faraguna

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Goran Dražić

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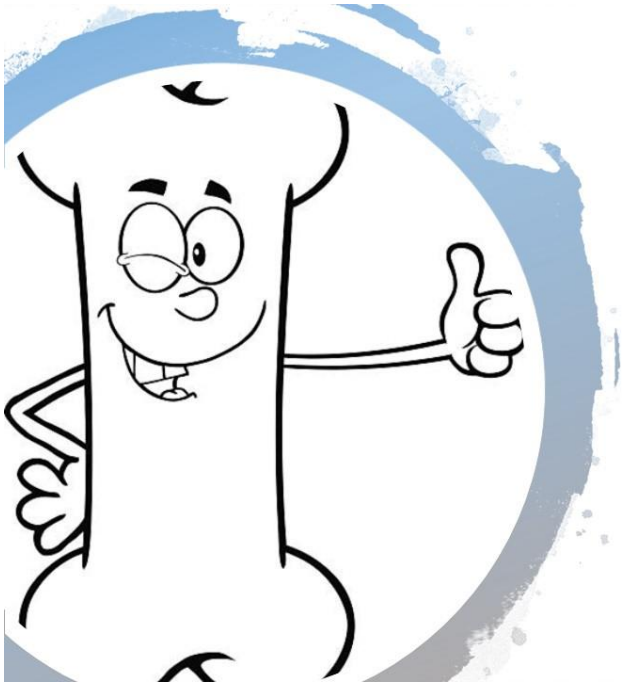

Thank you for your attention!