Highly adherent biological Hydroxyapatite thin films synthesized by pulsed laser deposition techniques for medical applications

(*HABioMed*)

Project Code: PN-II-RU-TE-2014-4-1570

Project No.: TE 108

Implementation period: 01.10.2015 – 30.11.2017

Project manager: **Dr. Duta Liviu-Marian**

Abstract

The surface functionalization of the implants represents an advanced development in implantology. Hydroxyapatite (HA) coatings for implants are the subject of recent researches pointing to the improvement of biointegration.

This project aims to improve the efficiency of the public health system by studying and developing of Pulsed Laser Deposition protocols for the synthesis of biocompatible nanostructured layers of innovative materials (biological HA, simple or reinforced with various ions) onto metallic substrates (Ti and Ti alloys) for implantology.

The synthesized nanostructures with controlled morphology, structure, thickness and adherence will be thoroughly studied by complex physical-chemical and mechanical investigations. During in-vitro specific bioactivity assays (Simulated Body Fluids), the physical-chemical modifications of the implant's surface will be monitored as an effect of the interaction with the biological fluids. In-vitro cell cultures and antimicrobial tests will consist in the study of adhesion, proliferation and differentiation of cells and bacterial and fungal strains adhesion onto the studied materials, in order to quantify their biological response.

The expected results refer to obtaining competitive biomimetic implants with increased functionality and biological efficiency as compared to the ones synthesized by plasma spraying, the single technique used nowadays for coating of medical implants.

Objectives

The main objective of the project is to improve the efficiency of the public health system, with respect to the state-of-the art, by developing PLD protocols for the synthesis of biocompatible nanostructured layers of innovative materials (biological HA, simple or reinforced with various ions) onto metallic substrates (Ti or Ti alloys) for implantology.

Other objectives:

O1. Nanopowders of biological HA and Ti implants (medical grade) for PLD experiments.
O2. The synthesis of biological HA materials (simple, reinforced, multistructures) by PLD.
O3. Complex characterization methods for the nanostructures synthetized by PLD.
O4. In-vitro evaluation of the best thin films from the morphological, structural and mechanical points of view.

Part of the expected results of the project:
***** protocol for obtaining compact targets, from renewable HA powders (simple or reinforced) as viable alternatives to those commercially available;
***** protocol for the synthesis of uniform, rough and crystalline structures with adherence values superior to those imposed by international standards (>15 MPa) and among the highest reported in literature for PLD technique;
* bioactivity/biocompatibility behaviours appropriate to medical implants after physical-chemical modifications of implant surfaces immersed in biological fluids;
***** in-vitro tests performed on the best structures from the morphological, compositional, structural and mechanical points of view;
* dissemination of the obtained results in well-known ISI journals, oral and poster presentations at National and International Conferences, Workshops, Schools, work stages.

(viii) lista activităților de diseminare,

<u>Dissemination of Project results</u>:

<u>ISI articles</u>

1. „*Thickness influence on in vitro biocompatibility of Titanium Nitride thin films synthesized by Pulsed Laser Deposition*", **Liviu Duta**, George E. Stan, Adrian C. Popa, Marius A. Husanu, Sorin Moga, Marcela Socol, Irina Zgura, Florin Miculescu, Iuliana Urzica, **Andrei C. Popescu**, **Ion N. Mihailescu**, Materials **9**, 38, PP. 1–19, DOI: 10.3390/ma9010038, 2016;

2. „*Fabrication of antimicrobial silver-doped carbon structures by combinatorial pulsed laser deposition*", **Ion N. Mihailescu**, Dorota Bociaga, Gabriel Socol, George E. Stan, Mariana-Carmen Chifiriuc, Coralia Bleotu, Marius A. Husanu, **Gianina Popescu-Pelin**, **Liviu Duta**, Catalin R. Luculescu, Irina Negut, Claudiu Hapenciuc, Cristina Besleaga, Irina Zgura, Florin Miculescu, International Journal of Pharmaceutics **515**, PP. 592–606, 2016;

3. „*Influence of laser pulse frequency on the microstructure of aluminum nitride thin films synthesized by pulsed laser deposition*", K. Antonova, **L. Duta**, A. Szekeres, G.E. Stan, **I.N.**

Mihailescu, M. Anastasescu, H. Stroescu, M. Gartner, Applied Surface Science **394**, PP. 197–204, 2017;

4. „*Optimized silicon reinforcement of carbon coatings by pulsed laser technique for superior functional biomedical surfaces fabrication*", **I.N. Mihailescu**, D. Bociaga, **G. Popescu-Pelin**, G.E. Stan, **L. Duta**, G. Socol, M.C. Chifiriuc, C. Bleotu, V. Lazar, M.A. Husanu, I. Zgura, F. Miculescu, I. Negut, C. Hapenciuc, Biofabrication **9**, 025029, 2017;

5. „*Comparative physical, chemical and biological assessment of simple and titanium-doped ovine dentine-derived hydroxyapatite coatings fabricated by pulsed laser deposition*", **L. Duta**, N. Mihailescu, **A.C. Popescu**, C.R. Luculescu, **I.N. Mihailescu**, G. Çetin, O. Gunduz, F.N. Oktar, A.C. Popa, A. Kuncser, C. Besleaga, G.E. Stan, Applied Surface Science 413, PP. 129–139, 2017;

6. „*TEM and AFM studies of Aluminum Nitride films synthesized by Pulsed Laser Deposition*", Zs. Fogarassy, P. Petrik*****, **L. Duta***, **I.N. Mihailescu**, M. Anastasescu, M. Gartner, K. Antonova, A. Szekeres, Applied Physics A: Materials Science & Processing 123:756, doi: 10.1007/s00339-017-1296-4, 2017;

7. „*Sub 100 nm thin carbon films as protection for drilling tools against wear*", **Camelia Popescu**, Daniel Cristea, Bogdan Bita, Rodica Cristescu, Doina Craciun, Georgiana Diana Chioibasu, Catalin Luculescu, Irina Paun, **Liviu Duta**, **Andrei C. Popescu**, submitted for publication in Coatings (Manuscript ID: coatings-235694), Oct 2017;

8. „*New bio-active, antimicrobial and adherent coatings of nanostructured Carbon double-reinforced with Silver and Silicon by Matrix-Assisted Pulsed Laser Evaporation for medical applications*", **L. Duta**, **C. Ristoscu**, G.E. Stan, M.A. Husanu, C. Besleaga, M.C. Chifiriuc, V. Lazar, C. Bleotu, F. Miculescu, N. Mihailescu, E. Axente, D. Bociaga, M. Badiceanu, **Ion N. Mihailescu**, submitted for publication in Applied Surface Science (Ref: APSUSC-D-17-10875), Oct 2017;

9. „*Physical-chemical characterization and biological assessment of simple and Lithium-doped biological-derived hydroxyapatite thin films for a new generation of metallic implants*", **A.C. Popescu**, **P.E. Florian**, G.E. Stan, **G. Popescu-Pelin**, I. Zgura, M. Enculescu, F.N Oktar, R. Trusca, L.E. Sima, A. Roseanu, **L. Duta**, submitted for publication in Applied Surface Science (Ref: APSUSC-D-17-11040), Oct 2017.

<u>Book Chapter</u>

1. „*Laser thin films deposition and characterization for biomedical applications*", F. Sima, **C. Ristoscu**, **L. Duta**, O. Gallet, K. Anselme, **I.N. Mihailescu**, Chapter 3 in: „Laser Surface Modification of Biomaterials. Techniques and Applications", Rui Vilar (Ed.), Woodhead Publishing Series in Biomaterials: Number 111, PP. 77–125, 10.1016/B978-0-08-100883-6.00003-4, 2016.

Proceedings

1. „*Orientation of the nanocrystallites in AlN thin film determined by FTIR spectroscopy*", K. Antonova, A. Szekeres, **L. Duta**, G.E. Stan, N. Mihailescu, **I.N. Mihailescu**, Journal of Physics: Conference Series 682, 012024, DOI:10.1088/1742-6596/682/1/012024, 2016.

Oral presentations

1. "*Cell proliferation and differentiation on biocompatible substrates for implantology applications*", **Paula Florian**, oral presentation at Scientific Seminary, Biochemistry Institute of Romanian Academy, October 14-th, 2016;

2. "*Structure and biocompatibility properties of hydroxyapatite coatings deposited by pulsed laser technique*", Sorin Ciuca, **Liviu Duta**, Andrei Berbecaru, Mihai Branzei, Ruxandra Paunescu-Dumitrescu, Olivia Dobrica, Ioana Gherghescu, oral presentation at „6-th International Conference on Materials Science and Technology (ROMAT2016)", Bucharest, Romania, 9-th – 12-th November, 2016.

Poster presentations

1. „*Highly adherent biological hydroxyapatite thin films reinforced with various oxides and metals for implantology applications*", **L. Duta**, G.E. Stan, **G. Popescu-Pelin**, F.N. Oktar, N. Mihailescu, **I.N. Mihailescu**, **A.C. Popescu**, poster presentation (C.PII.39) at EMRS Spring Meeting Symposium C – Laser-material interactions for tailoring future applications, Lille, France, May 2-nd – 6-th, 2016;

2. „*The influence of thickness upon the biocompatibility of Titanium Nitride thin films synthesized by Pulsed Laser Deposition*", **L. Duta**, G.E. Stan, A.C. Popa, **A.C. Popescu**, poster presentation (C.PI.34) at EMRS Spring Meeting Symposium C – Laser-material interactions for tailoring future applications, Lille, France, May 2-nd – 6-th, 2016;

3. „*Effect of deposition temperature upon multi-stage Pulsed Laser Deposition of Aluminum Nitride structures*", **L. Duta**, G.E. Stan, M. Anastasescu, H. Stroescu, M. Gartner, Zs. Fogarassy, S. Bakalova, N. Mihailescu, A. Szekeres, **I.N. Mihailescu**, **A.C. Popescu**, poster presentation (C.PI.35) at EMRS Spring Meeting Symposium C – Laser-material interactions for tailoring future applications, Lille, France, May 2-nd – 6-th, 2016;

4. „*Pulsed laser deposition of simple and reinforced biological hydroxyapatites for medical applications*", **Liviu Duta**, Jeanina Lungu, George Stan, **Andrei Popescu**, **Gianina Popescu-Pelin**, **Carmen Ristoscu**, Faik Oktar, **Ion Mihailescu**, poster presentation (S2 P20) at 16-

th International Balkan Workshop on Applied Physics (IBWAP 16), Constanta, Romania, July 7-th – 9-th, 2016;

5. „*In vitro assessment of reinforced biological hydroxyapatite thin films for implantology applications*", **L. Duta**, **A.C. Popescu**, G.E. Stan, **G. Popescu-Pelin**, **I.N. Mihailescu**, **P.E. Florian**, L.E. Sima, A. Roseanu, F.N. Oktar, poster presentation (P84) at 10-th International Conference on Photoexcited Processes and Applications (ICPEPA-10), Brasov, Romania, August 29-th – September 2-nd, 2016;

6. „*The effect of a "seed" layer upon multi-stage Pulsed Laser Deposition of Aluminum Nitride structures at different substrate temperatures*", **Liviu-Marian Duță**, Hermine Maria Stroescu, George Stan, Mihai Anastasescu, Măriuca Gartner, Zsolt Fogarassy, Anna Szekeres and **Ion Mihăilescu**, poster presentation (S05-P10) at International Conference of Physical Chemistry (ROMPHYSCHEM 16), „Dunarea de Jos" University of Galati, Romania, September 21-st – 23-rd, 2016;

7. „*In vitro investigations of highly adherent biological hydroxyapatite thin films for a new generation of implants*", **G. Popescu-Pelin**, G.E. Stan, **A.C. Popescu**, **V. Grumezescu**, M. Enculescu, C. Besleaga, I. Zgura, **P.E. Florian**, L.E. Sima, A. Roseanu, F.N Oktar, **I.N. Mihailescu**, **L. Duta**, poster presentation (P–22) at International symposium „Priorities of Chemistry for a Sustainable Development" (PRIOCHEM, 12-th edition), Bucharest, Romania, October 27-th – 28-th, 2016;

8. „*TEM and AFM Studies of AlN films synthesized by Pulsed Laser Deposition*", Zs. Fogarassy, P. Petrik, **L. Duta**, G. Stan, **I.N. Mihailescu**, M. Anastasescu, M. Gartner, K. Antonova, A. Szekeres, poster presentation (X P_1.20) at EMRS Spring Meeting, Symposium X „New frontiers in laser interaction: from hard coatings to smart materials", Strasbourg Convention Center, France, May 22-nd – 26-th, 2017;

9. „*Physical, chemical and in vitro assessment of biological-derived HA thin films for a new generation of metallic implants*", **L. Duta**, G.E. Stan, **A.C. Popescu**, **G. Popescu-Pelin**, A. Achim, M. Enculescu, I. Zgura, **P.E. Florian**, A. Roseanu, F.N Oktar, poster presentation (X P_1.27) at EMRS Spring Meeting, Symposium X „New frontiers in laser interaction: from hard coatings to smart materials", Strasbourg Convention Center, France, May 22-nd – 26-th, 2017;

10. „*Physical-chemical characterization and biological assessment of bovine derived hydroxyapatite thin films synthesized by pulsed laser deposition for a new generation of metallic implants*", **Paula E. Florian**, **Andrei C. Popescu**, George E. Stan, **Gianina Popescu-Pelin**, M. Enculescu, Irina Zgura, Faik N. Oktar, Livia E. Sima, Anca Roseanu, **Liviu M. Duta**, poster presentation at Annual International Conference of the Romanian Society of Biochemistry and Molecular Biology, Timisoara, Romania, June 08-th – 09-th, 2017;

11. „*Simple and reinforced biological-derived hydroxyapatite coatings for metallic implants*", **L. Duta**, G.E. Stan, **A.C. Popescu**, **P.E. Florian**, **G. Popescu-Pelin**, **V. Grumezescu**, A. Achim, M.

Enculescu, I. Zgura, F.N. Oktar, A. Roseanu, poster presentation at „15th Conference & Exhibition of the European Ceramic Society (ECerS2017)", Budapest, Hungary, July 09^{-th} – 13^{-th}, 2017;

12. „*Optical and mechanical properties of aluminum nitride nanostructured thin films synthesized by pulsed laser deposition at different temperatures*", K. Antonova, A. Szekeres, L. Kolaklieva, V. Chitanov, H. Stroescu, M. Gartner, **L. Duta**, **I.N. Mihailescu**, poster presentation at „Twentieth International Summer School on Vacuum, Electron and Ion Technologies (VEIT)", Sozopol, Bulgaria, September 25^{-th} – 29^{-th}, 2017.

In vivo testing of novel doped biological-derived hydroxyapatite thin films synthesized by pulsed laser deposition techniques for a new generation of metallic implants

(*VivoBHA*)

Project code: PN-III-P1-1.1-PD-2016-1568

Project No.: PD 6

Implementation period: 02.05.2018 – 30.04.2020

Project manager: **Dr. Duta Liviu-Marian**

Abstract

An implant represents a medical device or tissue designed to permanently or temporarily replace or support any damaged part of the human body aiming to enhance its functionality. There are still reports of a great number of metallic implants that suffer integration failure in the host body because of scarce biocompatibility and/or poor osseointegration.

Hydroxyapatite (HA) is a biocompatible material used as a bone replacement due to its proven osseoconductive properties. However, bulk HA presents weak mechanical properties. In contrast to their excellent mechanical properties, the Ti implants elicit low osseointegration rates. Therefore, their surface functionalization with HA coatings is considered to significantly improve osseoconductivity.

The reliability, fast processing and low production costs have advanced Pulsed Laser Deposition as a versatile method used in the field of thin film synthesis.

A great challenge of the nowadays intensive research in the field of bioactive materials is to reach biomimetism. Therefore, these materials have to behave similar to the human bone in order to be easily integrated into the living body, without inducing adverse reactions. The targeted material should contain oligoelements, which are normally present in the human healthy bone and are important for its functionality, and eventually dopants able to boost its bioactivity. The simplest way to obtain a HA with a bone-like composition, is to manufacture the powders from sustainable materials, such as those of biological origins. Compared to synthetic HA, these novel materials have a great ability to create perfect connections with the host tissue at the implantation site.

The limited reports available in the literature and the recent progresses made by our research team on the synthesis and exhaustive analyses of these renewable materials, validated their use for in vivo tests and the envisaged results should offer guidance toward future medical applications.

The general goal of the Project is to obtain novel, competitive biomimetic metallic implants functionalized with biological-derived HA materials, characterized by excellent mechanical properties, in vitro biological efficiency and high in vivo functionality, aiming to enable the

production of cheaper medical devices, with faster osseointegration rates and long-time stability inside the living body.

Other objectives of the Project:

O1. Explore the limits of the approach on small deposition areas, which are suited to PLD.

O2. Optimization of the deposition parameters to deliver protocols for the synthesis of high-quality biological-derived HA coatings for in vivo tests' validation.

O3. Using complex physical-chemical techniques for the characterization of the synthesized coatings.

O4. Defining a pull-off model to study the kinetics and strength of bone bonding, with negligible influence of mechanical inter-locking or shear forces.

O5. Evaluation of biochemical bonding strength at the interface of bone and simple Ti and doped biological-derived HA films as guiding parameter holding a strong potential to influence the direction toward future biomedical applications.

O6. Dissemination of results.

Part of the expected results of the project:

* sustainable doped biological-derived HA powders as viable substitutes to synthetic ones;

* optimized protocols for compact targets fabrication and for accurate elimination of any risk of disease transmission in powders and synthesized coatings;

* Ti implants as proper substrates for PLD experiments and further in vivo tests;

* optimized protocol for the fabrication of high-quality coatings to be validated for in vivo testing;

* post-deposition thermal treatments for the preservation of stoichiometry and/or improvement of coatings' crystallinity.

* protocol for the evaluation of biochemical bonding strength at the interface between bone and simple and biological-derived HA-functionalized Ti implant-disks fixed on rabbit tibia.

* papers published in ISI journals, oral and poster presentations defended at National/International Conferences, Workshops, Seminaries, Schools and/or other manifestations in the field;

* web page, edited and up-dated regularly, containing the most representative results of the Project.

Project results dissemination:

ISI articles

1. „*Pulsed laser deposited biocompatible lithium-doped hydroxyapatite coatings with antimicrobial activity*", **L. Duta***, M.C. Chifiriuc, G. Popescu-Pelin, C. Bleotu, G.P. Gradisteanu, M. Anastasescu, A. Achim, A.C. Popescu*, Coatings **9** (54), doi:10.3390/coatings9010054, 17 January 2019;

2. „*Prototype orthopedic bone plates 3D printed by laser melting deposition*", D. Chioibasu, A. Achim, C. Popescu, G.E. Stan, I. Pasuk, M. Enculescu, S. Iosub, **L. Duta**, A.C. Popescu, Materials **12**, 906, doi:10.3390/ma12060906, 19 March 2019;

3. „*Pulsed Laser Deposition of Aluminum nitride films: correlation between mechanical, optical, and structural properties*", L. Kolaklieva, V. Chitanov, A. Szekeres, K. Antonova, P. Terziyska, Zs. Fogarassy, P. Petrik, I. N. Mihailescu, **L. Duta***, Coatings **9**, 195, doi:10.3390/coatings9030195, 17 March 2019;

4. „*Current Status on Pulsed Laser Deposition of Coatings from Animal-Origin Calcium Phosphate Sources*", **Liviu Duta**, Andrei C. Popescu, Coatings 9 (5), 335, 2019, doi:10.3390/coatings9050335, 24 May 2019;

5. „*Corrosion behaviour in simulated body fluid of chemical-mechanical polishing titanium surfaces*", Laura Floroian, Zeynep Ozdemir, Doina Craciun, Mihaela Badea, Julia Claudia Mirza Rosca, G. Bahar Basim, **Valentin Craciun**, manuscript submitted for publication to Applied Surface Science, September 2019;

6. „*Lithium-Doped Biological-Derived Hydroxyapatite Coatings Sustain In Vitro Differentiation of Human Primary Mesenchymal Stem Cells to Osteoblasts*", Paula E. Florian, **Liviu Duta**, Valentina Grumezescu, Gianina Popescu-Pelin, Andrei C. Popescu, Faik N. Oktar, Robert W. Evans, Anca Roseanu Constantinescu, Coatings **9**, 781; doi:10.3390/coatings9120781, 21 November 2019;

7. „*Animal Origin Bioactive Hydroxyapatite Thin Films Synthesized by RF-Magnetron Sputtering on 3D Printed Cranial Implants*", Diana Chioibasu, **Liviu Duta**, Gianina Pelin-Popescu, Nicoleta Popa, Nichita Milodin, Stefana Orobeti (Iosub), Liliana Marinela Balescu (Trinca), Aurelian Catalin Galca, Adrian Claudiu Popa, Faik N. Oktar, George E. Stan, Andrei C. Popescu, Metals **9**, 1332, doi:10.3390/met9121332, December 2019;

8. „*Antimicrobial and cytocompatible bovine hydroxyapatite-alumina-zeolite composite coatings synthesized by pulsed laser deposition from low-cost sustainable natural resources*", G. Popescu-Pelin, C. Ristoscu, **L. Duta**, G.E. Stan, I. Pasuk, T. Tite, M.S. Stan, C. Bleotu, M. Popa, M.C. Chifiriuc, F.N. Oktar, A. Nicarel, I.N. Mihailescu, ACS Sustainable Chemistry & Engineering **8** (10), 4026–4036, doi:10.1021/acssuschemeng.9b05031, February 2020.

9. „*In vivo* assessment of bone enhancement in the case of 3D printed implants functionalized with lithium-doped biological-derived hydroxyapatite coatings: a preliminary study on rabbits", **L. Duta**, J. Neamtu, R. Melinte, O.A. Zureigat, G. Popescu-Pelin, D. Chioibasu, F.N. Oktar, A.C. Popescu, Coatings **10**, 992; doi:10.3390/coatings10100992, September 2020.

Patent

1. *Additive manufacturing of fixing devices for metallic implants by "Laser Melting Deposition" method / Fabricatie aditiva a unor dispozitive de fixare pentru implanturi metalice prin metoda "Depunere Laser prin Topire",* Chioibasu Diana, Mihai Sabin-Andrei, **Duta Liviu-Marian**, Popescu C. Andrei, patent aplication submitted to OSIM, **A00214 / 22.04.2020**.

Oral presentations

1. "*Implant-type bioceramic coatings synthesized from various low-cost sustainable biological-derived resources: physical-chemical properties and in vitro biofunctional performance*", **L. Duta***, oral presentation at Workshop CETAL, National Institute for Lasers, Plasma and Radiation Physics, Magurele, Romania, July 17^{-th} - 18^{-th}, 2018;

2. "*Biocompatible biological-derived hydroxyapatite coatings with antimicrobial activity*", **L. Duta***, M.C. Chifiriuc, G.E Stan, F.N. Oktar, V. Craciun, oral presentation (V 5.2) at E-MRS Spring Meeting, Nice, France (Congress & Exhibition Centre Acropolis), May 27^{-th} - 31^{-st}, 2019;

3. "*AlN thin films for piezoelectric devices*", H. Stroescu, M. Anastasescu, **L. Duta**, I.N. Mihailescu, G.E. Stan, A. Szekeres, C. Moldovan, M. Zaharescu, M. Gartner, oral presentation (F 1.6) at E-MRS Spring Meeting, Nice, France (Congress & Exhibition Centre Acropolis), May 27^{-th} - 31^{-st}, 2019;

4. "*Study of different metal oxides films for integration in piezoelectric microsystems*", H. Stroescu, M. Anastasescu, M. Chelu, E. Tenea, D. Mitrea, P. Terziyska, A. Szekeres, C. Moldovan, **L. Duta**, I.N. Mihailescu, R. Birjega, I. Boierasu, M. Dinescu, G.E. Stan, M. Zaharescu, M. Gartner, oral presentation (PR–13) at Twenty-First International Summer School On Vacuum, Electron And Ion Technologies (VEIT 2019), Sozopol, Bulgaria, May 23^{-rd} - 27^{-th}, 2019.

Poster presentations

1. „*Lithium-doped biological hydroxyapatite coatings for biomedical applications*", **L. Duta**, A.C Popescu, G.E. Stan, G. Popescu-Pelin, P.E. Florian, A. Roseanu, I. Zgura, F.N. Oktar, poster presentation (F.P1.27) at EMRS Spring Meeting Symposium F – Biomaterials and nanomedicine - Advanced biomaterials: elaboration, nanostructure, interfaces with tissues, Convention Center of Strasbourg, France, June 18^{-th} – 22^{-nd}, 2018;

2. „*Physico-chemical and biological evaluation of synthetic and lithium-doped biological hydroxyapatite coatings obtained by PLD*", G. Popescu-Pelin, G. Stan, S. Ciuca, F.N. Oktar, A. Achim, A. Berbecaru, **L. Duta**, poster presentation (F.P2.27) at EMRS Spring Meeting Symposium F – Biomaterials and nanomedicine - Advanced biomaterials: elaboration, nanostructure, interfaces with tissues, Convention Center of Strasbourg, France, June 18^{-th} – 22^{-nd}, 2018;

3. „*Mechanical, optical and structural properties of aluminum nitride synthesized by Pulsed Laser Deposition*", L. Kolaklieva, V. Chitanov, K. Antonova, P. Terziyska, A. Szekeres, P. Petrik, Zs. Fogarassy, A. Romanenko, **L. Duta**, I. N. Mihailescu, poster presentation (X.PI.16) at EMRS Spring Meeting Symposium X – Material processing and characterization - Photon-assisted synthesis and processing of materials in nano-microscale, Convention Center of Strasbourg, France, June 18^{-th} – 22^{-nd}, 2018;

4. „*Lithium-doped biological-derived hydroxyapatite coatings synthesized by pulsed laser deposition for biomedical applications*", Paula E. Florian, A.C. Popescu, G.E. Stan, G. Popescu-Pelin, I. Zgura, F.N. Oktar, Livia Elena Sima, Anca Roseanu, **Liviu Duta**, poster presentation (P 22) – **selected for oral presentation** – at "The Annual International Conference of the Romanian Society of Biochemistry and Molecular Biology", Romanian Academy Library, Heliade Radulescu Amphitheatre, Bucharest, September 5^{-th} – 7^{-th}, 2018;

5. „*Novel biocompatible lithium-doped hydroxyapatite coatings with antimicrobial activity synthesized by Pulsed Laser Deposition*", **L. Duta**, M.C. Chifiriuc, G. Popescu-Pelin, C. Bleotu, G.P. Gradisteanu, A.C. Popescu**, V. Craciun**, poster presentation (D.P.18) at EMRS Fall Meeting Symposium D – Layered, hybrid and biomaterials - Multifunctional advanced composite materials: from idea to market, Warsaw University of Technology, Poland, September 17^{-th} – 20^{-th}, 2018;

6. „*Pulsed laser deposition of biocompatible lithium-doped biological-derived hydroxyapatite coatings with antimicrobial properties*", **L. Duta,** M.C. Chifiriuc, G. Popescu-Pelin, C. Bleotu, G.P. Gradisteanu, A.C. Popescu, **V. Craciun**, poster presentation (T9-P2) at "12^{-th} International Conference on Physics of Advanced Materials (ICPAM-12)", Heraklion, Greece, September 22^{-nd} – 28^{-th}, 2018;

7. „*Biological-derived hydroxyapatite coatings synthesized by PLD from renewable resources*", **L. Duta**, M.C. Chifiriuc, G.E Stan, F.N. Oktar, V. Craciun, poster presentation (V.P1.29 – invited for Young Scientists Forum, V.YSF.23) at E-MRS Fall Meeting, Warsaw University of Technology - Plac Politechniki 1, Warsaw, Poland, September 16^{-th} – 19^{-th}, 2019;

8. „*Influence of the pulsed laser deposition conditions on the microstructure and mechanical properties of thin AlN films*", A. Szekeres, A. Cziraki, Zs. Fogarassy, L. Kolaklieva, V. Chitanov, P. Terziyska, G.E. Stan, I.N. Mihailescu, **L. Duta**, poster presentation (PB–4) at Twenty-First International Summer School On Vacuum, Electron and Ion Technologies (VEIT 2019), Sozopol, Bulgaria, September 23^{-rd} – 27^{-th}, 2019.

Other results

The Project leader together with (***i***) a close collaborator from "Center for Advanced Laser Technologies" (CETAL) and (***ii***) Prof. Faik N. Oktar, from Marmara University, Istanbul, Turkey, are co-Quest Editors of two Special Issues of the International Journal "Coatings" (MDPI), with the running titles "***Current Research in Pulsed Laser Deposition***" and "***Synthetic and Biological-Derived Hydroxyapatite Implant Coatings***". The thematics of these Special Issues are dedicated to PLD synthesis of thin films and nanoparticles, with applications in various scientific domains, and to obtaining and characterisation techniques of synthetic and natural-origin hydroxyapatites, with applications in the medical field, respectively. More details related to these Special Issues can be obtained by accessing the following links (http://www.mdpi.com/journal/coatings/special_issues/pulse_laser_depos and https://www.mdpi.com/journal/coatings/special_issues/hydroxyapatite).

Marine-derived hydroxyapatite coatings as sustainable biomaterials for implantable applications

(*MarineHA*)

Project Code: PN-III-P1-1.1-TE-2019-1449

Project No.: TE 189

Implementation period: 13.01.2021 – 30.12.2022

Project manager: **Dr. Duta Liviu-Marian**

Abstract

Over the last decades, the bone tissue engineering field has expanded to be able to address a plethora of bone-related traumas and to deliver a viable and efficient substitute to allografts or autografts, by combining bioactive materials and cells for bone tissue ingrowth.

Synthetic hydroxyapatite (HA) is one of the most widely used calcium phosphates in implantology. Because obtaining synthetic HA implies polluting, time-consuming and expensive approaches, researchers found a simple, economic and highly productive alternative to produce HA, i.e. to extract it from sustainable biological resources (bones and biogenic materials), considered nowadays only wastes of the food industry. Despite its excellent bone regeneration properties, HA is very brittle in bulk and characterized by poor mechanical properties. In contrast to their excellent mechanical properties, the Ti implants elicit low osseointegration rates. To overcome these shortcomings, HA can be applied as a coating onto the surface of Ti implants, to significantly improve the overall performances of the structures, by successfully combining the excellent bioactivity of the ceramic with the mechanical advantages of the metallic substrates. Therefore, in the field of thin film growth, the PLD technique stands as a simple, versatile, fast-processing, and cost-effective method, which allows for a precise control over the growth rate and morphology to obtain high-quality structures.

To the best of our knowledge, this is the first report in literature on the synthesis by PLD of marine-derived HA coatings and their complex physical-chemical, mechanical and biological investigation. The aim is to fabricate biocompatible implant coatings, from low-cost resources of important application potential in the biomedical field.

Project Objectives and Expected results

The main objective of this Project is to fabricate, in a single-step PLD process, innovative biomimetic metallic implant coatings from sustainable, low-cost marine-derived hydroxyapatite (MdHA) resources. These structures will present increased mechanical properties and excellent

biocompatibility. In addition, the prevention of bacterial adhesion and further biofilm development are also envisaged. In a context of sustainable development and competitiveness, the accomplishment of this goal will enable the fabrication of considerably lower-cost medical devices, with faster osseointegration rates and long-time stability inside the living body.

Other specific objectives:

– to deliver protocols containing optimized deposition parameters for the synthesis of novel materials (MdHA coatings, simple or doped with various concentrations of reactive agents);

– to tailor the adherence at the film-substrate interface;

– to synthesise both biocompatible and antimicrobial MdHA coatings;

– to use small-area deposition metallic substrates;

– to identify the roughness values for optimum cell viability, proliferation and adhesion, by tailoring the deposition parameters according to the biological feedback;

– to disseminate the obtained results.

Expected results:

– optimized protocols to (i) extract HA from MdHA materials and (ii) obtain compact targets for PLD experiments;

– protocols to prepare the surface of medical-grade implants by laser irradiation and chemical etching;

– optimized protocols to synthesize high-quality MdHA coatings, with bonding strength adherence values superior to those imposed by international standard;

– morphological, compositional, and structural complex investigation techniques as key-factors for choosing the best fabricated coatings for an optimized in vitro response;

– tests in simulated physiological solutions as preliminary evaluation of the biomineralization capability of the MdHA coatings;

– fabrication of both biocompatible and antimicrobial MdHA coatings, as viable substitutes to the synthetic HA ones;

– dissemination of the obtained results in prestigious ISI journals and delivering of oral/poster communications at National/International Conferences, Workshops and Schools in the field;

– web page, edited and up-dated regularly, containing the most representative results of the Project.

Dissemination:

ISI articles

1. "*A Review on Biphasic Calcium Phosphate Materials Derived from Fish Discards*", **L. Duta**, G. Dorcioman, **V. Grumezescu**, *Nanomaterials* 11 (11), 2856, https://doi.org/10.3390/nano11112856, 2021;

2. "*Marine-Derived Bioceramics for Orthopaedic, Reconstructive and Dental Surgery Applications*", Faik Nuzhet Oktar, Semra Unal, Oguzhan Gunduz, Besim Ben Nissan, Innocent J Macha, Sibel Akyol, **Liviu Duta**, Nazmi Ekren, Eray Altan, Mehmet Yetmez, *Journal of the Australian Ceramic Society*, https://doi.org/10.1007/s41779-022-00813-3, 2022;

3. "*Bioactive Hydroxyapatite-Magnesium Phosphate Coatings Deposited by MAPLE for Preventing Infection and Promoting Orthopedic Implants Osteointegration*", Denisa Alexandra Florea, **Valentina Grumezescu**, Alexandra Cătălina Bîrcă, Bogdan Ștefan Vasile, Andrei Iosif, Cristina Chircov, Miruna S. Stan, Alexandru Mihai Grumezescu, Ecaterina Andronescu, Mariana Carmen Chifiriuc, *Materials* 15, 7337, https://doi.org/10.3390/ma15207337, 2022;

4. "*Influence of post-deposition thermal treatments on the morpho-structural, and bonding strength characteristics of lithium-doped biological-derived hydroxyapatite coatings*", **L. Duta**, George E. Stan, Gianina Popescu-Pelin, Irina Zgura, M. Anastasescu, F.N. Oktar, *Coatings*, 12 (12), 1883, https://doi.org/10.3390/coatings12121883, 2022;

5. "*Hydroxyapatite thin films of marine origin as sustainable candidates for dental implants*", G. Dorcioman, **V. Grumezescu**, G.E. Stan, M.C. Chifiriuc, G. (Gradisteanu) Pircalabioru, F. Miculescu, E. Matei, G. Popescu-Pelin, Irina Zgura, V. Craciun, F.N. Oktar, **L. Duta**, *Pharmaceutics*, 15 (4), 1294, https://doi.org/10.3390/pharmaceutics15041294, 2023.

Editorial

1. "*Current Research in Pulsed Laser Deposition*", **L. Duta**, **A.C. Popescu**, *Coatings* 11, 274. https://doi.org/10.3390/coatings11030274, 2021;

2. "*Editorial: Calcium phosphates of synthetic and natural origin: current status and future challenges*", **Liviu Duta**, **Valentina Grumezescu**, *Frontiers in Bioengineering and Biotechnology* 11:1237272, https://doi.org/10.3389/fbioe.2023.1237272, 2023.

Oral presentations

1. "*Cytocompatible biological-derived hydroxyapatite coatings presenting antimicrobial activity*", **L. Duta**, M.C. Chifiriuc, **V. Grumezescu**, G.E. Stan, **O. Gherasim**, G. Popescu-Pelin, V. Craciun, F.N. Oktar, oral presentation (session J, ref. J.7.3) at the E-MRS 2021 Fall Meeting, VIRTUAL Conference, September 20^{-th} to 23^{-rd}, 2021;

2. "*Thin films deposition of hydroxyapatite derived from various natural resources for implantological applications*", **Liviu Duta**, George Stan, Carmen Chifiriuc, Florin Miculescu, **Valentina Grumezescu**, Gabriela Dorcioman, Elena Matei, Irina Zgura, Gianina Popescu-Pelin, Johny Neamtu, Faik Oktar, oral presentation at "20^{-th} International Balkan Workshop on Applied Physics and Materials Science", Ovidius University, Constanta, Romania, July 12^{-th} to 15^{-th}, 2022;

Poster presentations

1. "*Pulsed laser deposition of cytocompatible biological-derived hydroxyapatite coatings with antimicrobial activity*", **L. Duta**, **V. Grumezescu**, M.C. Chifiriuc, G.E Stan, **O. Gherasim**, **D. Chioibasu**, **A.C. Popescu**, F.N. Oktar, poster presentation (session P, ref. 17) at the E-MRS 2021 Spring Meeting, VIRTUAL Conference, May 31^{-st} to June 3^{-rd}, 2021;

2. "*Influence of thermal treatments on the morpho-structural and mechanical characteristics of biological-derived HA coatings*", **L. Duta, V. Grumezescu,** G.E Stan, **O. Gherasim,** G. Popescu-Pelin, F.N. Oktar, poster presentation (session P, ref. 18) at the E-MRS 2021 Spring Meeting, VIRTUAL Conference, May 31^{-st} to June 3^{-rd}, 2021;

3. "*Bi-phasic calcium phosphate implant coatings fabricated by pulsed laser deposition of natural origin*", Gianina Popescu-Pelin, Carmen Ristoscu, **Liviu Duta**, Iuliana Pasuk, George E. Stan, Miruna Silvia Stan, Marcela Popa, Mariana C. Chifiriuc, Claudiu Hapenciuc, Faik N. Oktar, Anca Nicarel, Ion N. Mihailescu, poster presentation (session I, ref. P.35) at the E-MRS 2021 Spring Meeting, VIRTUAL Conference, May 31^{-st} to June 3^{-rd}, 2021;

4. "*Marine-derived hydroxyapatite coatings synthesized by Pulsed laser deposition*", **L. Duta**, G.E Stan, **V. Grumezescu**, G. Dorcioman, E. Matei, I. Zgura, **O. Gherasim**, G. Popescu-Pelin, F.N. Oktar, poster presentation (O 8.27) at the Symposium O: *Nano-engineered coatings and thin films: from design to applications,* E-MRS 2022 Spring Meeting, VIRTUAL Conference, May 30^{-th} to June 3^{-rd}, 2022;

5. "*Hydroxyapatite of various biological origin: from powders to thin film cytocompatibility assessment*", **L. Duta**, **V. Grumezescu**, M.C. Chifiriuc, G.E Stan, **O. Gherasim**, I. Zgura, F.N. Oktar, poster presentation (O 8.29) at the Symposium O: *Nano-engineered coatings and thin films: from design to applications,* E-MRS 2022 Spring Meeting, VIRTUAL Conference, May 30^{-th} to June 3^{-rd}, 2022;

6. "*Hydroxyapatites of marine origin as sustainable candidates for implantology*", **Liviu Duta**, George E Stan, **Valentina Grumezescu**, Gabriela Dorcioman, Elena Matei, Irina Zgura, **Oana Gherasim**, Gianina Popescu-Pelin, Faik N Oktar, poster presentation (sciforum-057958) at the "3-rd International Online-Conference on Nanomaterials (IOCN 2022)", VIRTUAL format, April 25-th to May 10-th, 2022;

7. "*Pulsed laser deposition of hydroxyapatite derived from various biological resources for suitable use in implantology*", **Liviu Duta**, George Stan, Mariana Chifiriuc, Florin Miculescu, **Valentina Grumezescu**, Gabriela Dorcioman, Elena Matei, Irina Zgura, Gianina Popescu-Pelin, **Oana Gherasim**, Johny Neamtu, Faik Oktar, poster presentation (P4.09) at the "International Conference on Laser, Plasma and Radiation Science and Technology (ICLPR-ST)", Palace of Parliament, Bucharest, Romania, June 07-th to 10-th, 2022.