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Abstract of the doctoral thesis
„Bioactive nanocomposites for tissue engineering applications"

Unhealthy lifestyles, population ageing and the rapid development of civilization may lead to various types of diseases and injuries, which increase the need for transplantation and filling of existing tissue defects. In particular, limited physical activity, obesity and ageing processes cause a continuous growth in the incidences of skeletal injuries. Excessive damage of the bone tissue resulting from injuries or chronic diseases may prevent the body from initiating the regeneration process. That is why various approaches are being developed to facilitate tissue repair. In recent years, there has been a growing interest in tissue engineering as an alternative to conventional treatments. It is expected that this approach will eliminate common problems with organ transplantation (transplant rejection or lack of organ donors) as well as exclude the use of materials with relatively low biocompatibility, which is the cause of frequent postoperative complications. Tissue engineering is a field of regenerative medicine aiming at replacement of missing or damaged tissue with properly designed biomaterials, stimulating the reconstruction of body structures. In a case of serious tissue damage not only cells, but also the extracellular matrix (ECM) can be destroyed. The ECM is a natural scaffold for the cells and forms the tissues with the cells. Therefore, a new tissue can be created with cells that are seeded on appropriate ECM substitutes. Well-designed scaffolds should be structures favourable for cell adhesion, proliferation and differentation, being able to fulfill many important functions at the same time. In this regard, materials used for scaffold fabrication play an essential role. The highly biocompatible metals, bioceramics, polymers or composites are tested as such materials. Among them, the hydrogels are the most commonly used class of polymeric materials for scaffold preparation. They can create environment suitable for fast cell growth due to their biocompatibility, high content of water and possibility of diffusion for various substances. Nowadays the design of next-generation materials such as functional nanocomposites is a promising way - interesting structure and properties make them attractive candidates as scaffolds for bone tissue engineering. Until now, none of the materials developed was able to completely mimic the properties of living tissue. Keeping this in mind, **the aim of this thesis was to design, synthesise and characterize bioactive nanocomposites, potentially useful as scaffolds for tissue engineering applications, particularly for bone tissue regeneration.** The developed nanocomposites consisted of hydrogel matrix supplemented with selected inorganic components, forming structurally stable materials with the desired properties dependent on the composition of the nanocomposites.

This thesis provides an overview of the literature (**chapters 1-6**) on important aspects and achievements of tissue engineering, bone tissue engineering, design of scaffolds, polymeric hydrogels and functional nanocomposites as materials suitable for the fabrication of scaffolds as well as 3D printing, the procedure gaining more and more interest in the field of regenerative

medicine. The chapters relating to hydrogels and nanocomposites focus on the description of the components that were used to prepare bioactive nanocomposites. They were biopolymers – collagen, chitosan, hyaluronic acid and inorganic components – silica particles and magnetic nanoparticles, presented within the context of creating multifunctional bioactive nanocomposites useful in bone tissue regeneration assisted by sodium alendronate (anti-osteoporosis drug) or effected by the magnetic properties of the nanoparticles studied. The experimental part of this thesis consists of four main parts (**I-IV**), each of them contains an introduction, description of the materials and methods used, results, discussion and summary. Part I (**chapters 7 and 8**) concerns the preparation and characterization of the physicochemical and biological properties of nanocomposites based on a hydrogel matrix and silica particles, potentially useful as injectable bioactive scaffolds for bone tissue regeneration. The hydrogel matrix composed of collagen, chitosan and hyaluronic acid was optimized in terms of the content of biopolymers and the concentration of the crosslinking agent (genipin). Then amino-functionalized silica particles were dispersed in the selected matrix to form a stable bioactive hybrid material. Functional groups present on the surface of the silica particles ensured their incorporation into hydrogel structure (amino groups) and deposition of the mineral phase in the environment of simulated body fluid (silanol groups). Part II (**chapters 9 and 10**) presents the preparation and characterization of the physicochemical and biological properties of nanocomposites based on a hydrogel matrix and sodium alendronate carrier in the form of silica-apatite-sodium alendronate system, potentially useful as injectable scaffolds for the reconstruction of bone tissue, in particular small bone defects caused by osteoporosis. The hydrogel matrix was improved by the use of amino-functionalized hyaluronic acid, so that not only collagen and chitosan, but also modified hyaluronic acid could be chemically crosslinked with genipin. Then sodium alendronate carriers were incorporated into the selected matrix, producing a system that can support bone tissue regeneration and has therapeutic potential in the treatment of osteoporosis. The material was tested using both *in vitro* and *in vivo* assays. Part III (**chapter 11**) concerns the preparation and characterization of the physicochemical and biological properties of nanocomposites based on a hydrogel matrix and magnetic nanoparticles (magnetic hydrogels), potentially useful for bone tissue engineering applications with the possibility of supporting regeneration enhanced with external magnetic field. The magnetic phase consisted of superparamagnetic iron oxide nanoparticles (SPION) that were coated with a cationic derivative of chitosan. Then they were immobilized into the hydrogel matrix composed of collagen and chitosan crosslinked with genipin, obtaining structurally stable hydrogels with superparamagnetic properties. Polymeric coatings of nanoparticles allowed them to covalently bind to the hydrogel network preventing phase separation and aggregation. The last part – IV (**chapter 12**) concerns the development and initial characterization of the physicochemical and biological properties of 3D scaffolds fabricated by 3D printing using the extrusion method. The composition and crosslinking method of biopolymeric inks were optimized using biopolymers applied in earlier parts of the thesis. The selected biopolymeric inks were enriched with silica particles and magnetic nanoparticles (also developed at the earlier stages of the studies). Effect of inorganic components on the efficiency of the printing process and stability of the 3D scaffolds obtained was also investigated.

The thesis is closed with summary and conclusions. Additionally, the list of scientific achievements of the author is added.