Micro-Finite Element Model to Investigate the Mechanical Stimuli in Scaffolds Fabricated via Space Holder Technique for Cancellous Bone

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Received: 13 May 2019, Revised: 8 September 2019, Accepted: 17 October 2019

Abstract: In Osteoporosis, bone mechanical strength decreases and as a result, the risk of bone fracture increases. Osteoporosis is also referred as a "silent illness" since it usually develops asymptomatic until it breaks a long bone, like the femur. In recent years, porous scaffolds have been utilized to repair damaged bone tissue. For bone tissue engineering, synthetic scaffolds should have acceptable mechanical properties, in addition to the required biological properties. In this regard, the finite element simulation is used to predict the mechanical properties of porous bone scaffolds as one of the most common methods for reducing the experimental tests, because the acquisition of mechanical properties of such scaffolds is very time-consuming and expensive. Due to the widespread use of hydroxyapatite (HA) in the manufacture of bone scaffold composites, the mechanical properties of HA-wollastonite scaffold composites are obtained by laboratory tests and finite element methods. Comparison of the simulation of finite element analysis (FEA) and the experimental results indicate the success of the FEA simulation. In conclusion, new finding satisfied expectations as being suitable for mechanical and biomaterial aspect of a porous scaffold which is proven by laboratory tests and FEA simulations. Due to that fact, the result of this study can be employed to obtain scaffolds well-suited for bone implementations.

Keywords: Bone Scaffold, Bio-Nanocomposites, Computational and Laboratory Analysis, Finite Element Analysis, Hydroxyapatite

Reference: Seyed Alireza Hashemi, Esmaeili, Saeid Esmaeili, Mazyar Ghadiri Nejad, Saeed Saber-Samandari, Sheikhbahaei, Erfan Sheikhbahaei, Alireza Kordjamshidi, and Amirsalar Khandan, "Micro-Finite Element Model to Investigate the Mechanical Stimuli in Scaffolds Fabricated via Space Holder Technique for Cancellous Bone", Int J of Advanced Design and Manufacturing Technology, Vol. 13/No. 1, 2020, pp. 51–58.

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1 INTRODUCTION

The human body is a complex set of tissues and organs that have a particular function. As the age rises, there are changes in the body that cause damage to the tissues or impairment in their function. One of these hard tissues is bone. The high potential of bone for reproduction has made it an excellent example of tissue engineering. The human body can repair small defects and bone fractures, but its ability to repair large fractures and injuries does not need to be relied upon for repair. Some materials like MgO, Nano clay and magnetite nanoparticle can enhance the bone regeneration [1-6]. Hence, the increased need for treatment of bone problems caused by illness or injury has led to the growing spread of various therapies. One of these therapies is the placement of bone scaffolds at the site of defect, trauma or injuries. Bone scaffolds exhibit very similar mechanical and biological behaviors that are very similar to those of the actual bone, which has led to the much attention to these therapeutic approach [3-5]. The great challenge of bone scaffolds is to be designed in such a way as to have properties similar to those of human bone properties. Biocompatibility, favorable degradability rates, suitable physical and chemical properties, high porosity, together with associated cavities, desired mechanical properties, antibacterial (ZnO), conductive (copper nanoparticle) and stimulating bone growth are examples of these proper properties [6-7], [13]. Bone scaffolds provide a platform for the growth and proliferation of cells. The microstructure of the scaffold is a porous matrix, which helps porosity to attach to the cells properly. The main part of this work is the design of the porous scaffold to determine the suitable size of the cavity, porosity, and degree of degradability of a scaffold. Therefore, the destruction time is resistant to regional stresses and these pressures/forces throughout the implantation region are as homogeneous and equal [9-16]. There are different techniques to overcome these problems such as adding different materials to the base material, creating proper porosity and homogenize dispersion to arrange different components to be replaced with defect bone under various load bearing conduction [16-18]. Applying various techniques like 3D printing, freeze drying and space holder can produce proper porous scaffold with sufficient mechanical and chemical stability under biological condition [13-19]. In this work, we conduct an ABAQUS simulation on cylindrical orthopedic porous scaffold under various load bearings to simulate it mechanical performance.

1.1. Definition and Importance

Tissue Engineering is the science which can be used for repairing and recovering of damaged tissues. In tissue engineering, the scaffold is one of the main components which is responsible for creating a suitable environment for cell growth. For this reason, the construction of proper scaffolds is a very important challenge for tissue engineers. The potential impact of this field in the future is much more extensive.

Engineered tissues can reduce the need for tissue replacement and generally address the need for organ bundles. According to statistics, bone disease in Europe afflicted more than 39-40 million people, and in the United States, it is more than 20 million people and prognosticated to be doubled by 2020. For example, in Germany, the costs of directly or indirectly treating bone loss are reported to be over € 8 billion a year. Composite scaffolds are made for the replacement of bone tissue, mostly containing a mineral phase such as Hydroxyapatite (HA) or tricalcium phosphate (TCP) with a synthetic polymer such as poly glycolic acid (PGA), poly lactic acid (PLA) or poly lactic-*co*-glycolic acid (PLGA) or natural, such as collagen, alginate, chitosan and gelatin. These materials can have synergic properties with composition of polymer and ceramic together [20-25]. With the advent of nanotechnology, composites are replaced by nanocomposites. In addition to maintaining the properties of conventional composites, nanocomposites have more mechanical involvement with the background because of their free surface and, if formed by chemical bonds, their number and hence the added strength resulting from them will be higher. Biologically, experiments performed on nanocomposites have shown their superiority [14], [24-27]. On the other hand, looking at the final product with laboratory methods and performing animal experiments is a time-consuming process. Numerical simulations are used to reduce costs and save the time without any experimental work like simulations with the finite element analysis (FEA) [15].

1.2. Previous Researches

Lee et al. [16] produced two different structures in the form of conventional grid-type and microstructure by three-dimensional printing. Working in craniomaxillofacial (CMF) of rabbit over 16 weeks to measure mechanical properties, numerical analysis was used with complete elastic assumption and finite element method. Finally, the structure of the scaffold of the microstructure was better evaluated. Doyle et al. [17] investigated the selective laser sintering (SLS) method using polycaprolactone (PCL)/β-tricalcium phosphate (β-TCP) scaffolds by FEA. By comparing experimental results and simulation, it was concluded that numerical simulation with FEA has little error than reality, and this method can be used to predict the micro-mechanical behavior of scaffolds. Eshraghi et al. [18] used different percentages of hydroxyapatite and PCL to produce bone scaffolds with appropriate mechanical properties, and the SLS methodology that was modelled with FEA. The results of the FEA and the mechanical tests were in good agreement with each other. Son et al. [19], in a study measured the process of repairing long bones such as the tibia, with regard to blood vessel growth. In this study, epoxy/carbon and polypropylene/glass composites were used which were analyzed by Abacus software, and the results showed that a more flexible composite (polypropylene/glass), which has a Young modulus more than cortical bone, provides a better repair process. Researchers performed the pressure to obtain the elastic modulus on the scaffolds produced by the process of making the additive extrusion of the polymeric layer. With the acquired parameters, the FEA model was developed to predict the mechanical behavior of their production scaffolds. Finally, in reviewed researches a variety of materials were used, and the result of FEA was validated comparatively to laboratory tests. Hence, FEA was implemented for the new composition consist of different materials in comparison to previous studies.

1.3. Current Research

In the provided researches, a variety of methods and materials were hired to approach a scaffold mimicking a genuine bone property. Considering the above in this study, we simulate the HA-wollastonite model by finite element method to predict mechanical behaviors and examine the accuracy of the model by comparing the results of simulation and mechanical tests. In addition, the mentioned approach and materials were utilized to validate whether it can be used in the human body. In sum, using HA-wollastonite and means of determination of ultimate scaffold's properties stand out.

2 MATERIALS AND METHODS

2.1. Manufacture of HA powder

The hydroxyapatite obtained from the bovine bone is very similar to that of the HA in the other mammalian bones, although rarely the presence of rare elements has been reported. In this research, HA was produced according to the proposed method of Bahrololoom et al. [22]. After preparing the adult calf's hip, first, let it boil for two hours in order to separate the meat and fats attached to it. Then, the bones were placed in a temperature of 60°C for 24 hours to dry up the moisture. To prevent the formation of soot in the material during heating, the bones were cut into small pieces (10 mm×10 mm×10 mm) and then burned with a blowtorch in the air for up to 400°C for 3 hours leading in its organic compounds decomposition. The material produced by this process is black bone ash, which is used to produce hydroxyapatite (HA). After the process for the production of hydroxyapatite powder, and also analyzing the temperature of its production process, this black ash was exposed to heat treatment at temperatures of 600°C, 700°C, 900°C and 1100°C for 2 hours. The material obtained from this step is the white powder of the nanocrystal HA used to make ceramic scaffold composites.

The material is HA, which is obtained after grinding and sizing [23-24]. Considering the dimensions of samples needed for research, die was designed with CAD software. After that, the CK45 steel was used to provide a suitable toughening die. With grinding, the punch was sized to 6×10 mm (diameter height). For the matrix to be precisely 6×10 mm, it was cut to the right size with the wire-cut.

2.2. Method for Fabricating the Hydroxyapatite-Wollastonite Bio-Nanocomposite with Space Holder Technique

Nano-hydroxyapatite was considered as the base material, and the wollastonite was added to the weight ratios (35, 40, 45 and 50 wt%). Each of the samples with different measured weight percentages was placed in a separate container and tagged. Then each sample was placed into Chinese mortar and mixed for 5 minutes. After that, sodium chloride was added to each dish with a weight percentage of 80 and stirred for 5 minutes. In order to remove the samples easily, die was impregnated with oil (here edible oil) and the material of each tagged container was placed separately and stepwise into the die. The device was pressurized to 150 MPa; the press data was stored when pressed. In order to bring the specimens out of the die, two bundles of wood were placed around the die, and the samples were gently removed from the die. Then die was cleaned again, and the subsequent samples were placed into it. Afterwards, at the thermodynamic laboratory, the furnace was heated to a temperature of 800°C and the samples were put in a crucible and placed in the oven for 45 minutes until the salt evaporated and the desired scaffold, which is shown in "Fig. 1" was synthesized.

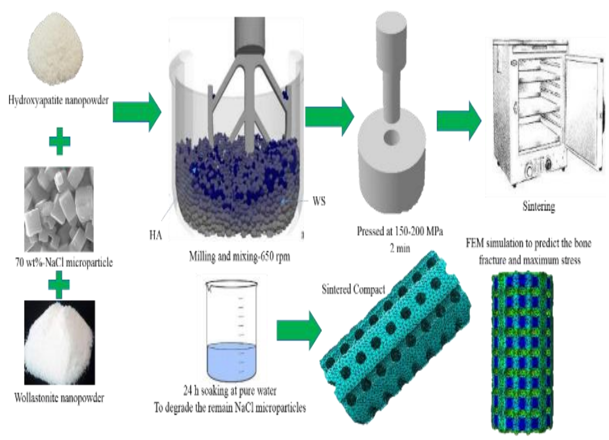

Fig. 1 Schematic preparation of nHA/$CaSiO_3$ in this study using space holder technique.

2.3. Preparation of Scaffold Composite

In order to build the structure of nanocrystalline HA particles using space holder, a certain proportion of powder with 80 wt% was mixed with sodium chloride, with a purity of 98%. The reason for this choice is exclusively due to the high strength of this space holder against the pressure, nontoxicity of this substance and the lack of chemical reaction of this material with the base [23]. An overview of the process for the production of hydroxyapatite scaffolds by using space holder is presented in “Fig. 1”. The powder and space holder were homogenized by amalgamation machine. In order to compress the homogeneous mixture, along with edible liquid oil (one percent by weight of homogenized powder), a cylindrical die of 6 mm diameter under pressure of 65 MPa was used. In order to completely remove the salt, the specimen was subjected to sintering at various temperatures of 1000-1250°C as shown in Schematic “Fig. 1”.

2.4. Structure Analysis by X-Ray Diffraction Infraction (XRD)

The X-ray diffraction (XRD) and Equinox 300 apparatus were used to determine the phases in the synthesized powder and the composite scaffold at a range of 2θ from 10 to 60 degrees under 40 kV and 30 mA which is shown in “Fig. 2”.

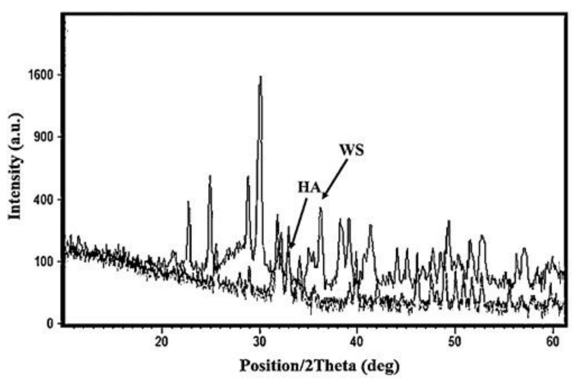

Fig. 2 The XRD comparison pattern of HA, WS, and HA-WS nanocrystalline powder used to fabricate cylindrical scaffold for femur fracture.

2.5. Evaluation of Mechanical Properties of Porous Scaffolds

To evaluate the mechanical characteristics, a compressive strength test and a SANTAM-STM50 machine made in Iran were used. One of the significant problems for the mechanical characterization of porous ceramic scaffolds is the type of device sample holder, tests such as stretching, and two-stage impact are not usually used for porous scaffolds. Instead, compression test for porous bone and hydroxyapatite samples is acceptable. Compressive strength test and compressive modulus for each sample (SANTAM-Eng. Design Co. LTD) with a load cell 10 KN were performed according to ASTM-D5024-95a standard. Dimensions of each specimen for the compression test were considered to be 10×14.3×10 mm^3. Due to the brittleness of the ceramic scaffold structure, the jaw speed for imposing pressure is considered to be 0.5 mm/min, so stress centres would not damage the ceramic structure. The applied load on the sample was about 30% of the initial length of the scaffold. The elastic modulus was calculated from the slope of the stress-strain curve, which is considered as the amount of compressive strength.

2.6. Determination of Porosity of Bio-Nanocomposites of HA Reinforced by WS

One of the physical properties of porous materials is their porosity. The association between the growth of bones in a scaffold with porosity has been well documented. Also, some researchers have reported that continuous porosity is much more critical than the porosity. The relative porosity and total porosity percentage are investigated and evaluated by calculating apparent density and actual density. Archimedes method was used for this purpose. The process of evaluation was as follows: at first two samples of each composition were selected and the dry weight (Wd) of the samples was measured. Samples were placed in water. They were then placed in a vacuum desiccator for half an hour to allow full penetration of water into the porosity. The wet weight of the samples (Ww) was measured after removing them from the water. Finally, the immersion weights of the samples (Ws) were recorded when they were suspended in water. The permeability of the samples was calculated using “Eqs. (1-2)”:

$$\text{Apparent porosity } (\%) = \frac{W_w - W_d}{W_w - W_s} \times 100 \quad (1)$$

$$\text{Swelling Ratio } (\%) = \frac{W_2 - W_1}{W_1} \times 100 \quad (2)$$

Where W_2 and W_1 are sample weight, respectively, swollen and dry.

3 RESULTS AND DISCUSSION

In order to predict the mechanical behavior of scaffolds, the finite element software of ABAQUS version 2019 was selected. Modeling was done using ABAQUS/CAE and Standard Solver. In order to study the porosity effect, a basic model was developed, and three models with different porosities were derived from it, which are described below. As mentioned, a base cylinder with a diameter of 6 mm and a height of 12 mm was created. Two models with the same porosity geometry, but different percentages were created. The third model was defined with a different porosity structure as the models are demonstrated in “Fig. 3”.

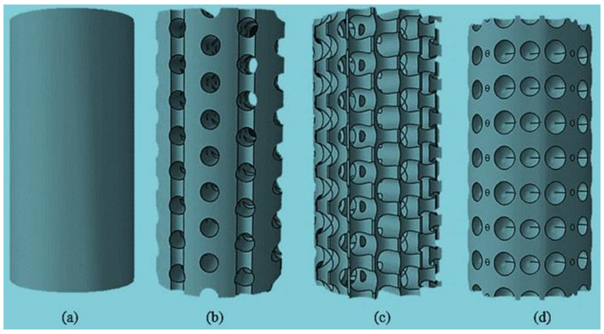

Fig. 3 (a): Base model without porosity, (b): first porous model, (c): second porous model and (d): spherical porosity model.

The properties of materials such as the Poisson coefficient and the elastic modulus are obtained from the compression test. The problem was defined in the software statically. The top surface of the models was under specific and equal pressure. This pressure is equal to the force that a person with a specific mass exerts on the surface of the scaffold. In such a way that the weight of a person is neutralized by two feet, and the force applied to one leg is equal to half the mass of a person multiplied by the acceleration of gravity of the earth.
Figure 2 shows the XRD pattern of the HA, WS, and sample with 10 wt% WS in HA powder at 10-70°. The XRD analysis indicated that the HA and WS both have nanocrystalline phase with spherical shape as the size of the powder were detected in the range of 100-150 nm. The following outcome can prove the increased mechanical strength in the Abaqus software evaluation. As the bioceramics with the spherical shape were added to each other, they can increase their elastic modulus of the final product according to the rule of mixture theory (ROM). All degrees of freedom of the bottom surface of the other cylinders were fixed similar to the compression test. Meshing the base scaffold, sweep and the medial axis method were used. For other models, the second-degree tetragonal element was used. In order to ensure the independence of the results from the number of meshes used, three different mesh size was obtained for each model shown in "Fig. 4".

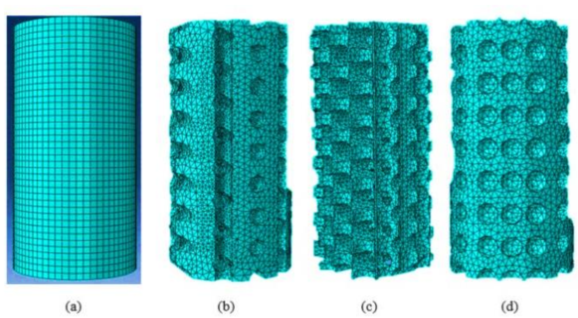

Fig. 4 Meshed models used for analysis of the porous bio-nanocomposite: (a): cubic mech, (b): non homogenised, (c): randomize and (d): spherical porous architecture.

After that the independence of the results is ensured, the larger meshes for efficiency. Then simulation of each model was performed with four processing cores. Bone scaffolds should have suitable mechanical properties according to the application. Using the finite element model presented in this study, and with proper boundary conditions, mechanical properties of the bone scaffold can be evaluated. Therefore, it will save time and money. In this section, the results of simulation of bone scaffolds are presented. Figure 4(a-d) shows the meshed models used for analysis of the porous bio-nanocomposite with the cubic mech, non-homogenized, randomize, and spherical porous microarchitecture. As it is shown in "Fig. 5(a-d)", under identical conditions and porosity variation, the stresses formed in the porous scaffolds are different. The base model is depicted in "Fig. 5-a", which is a perfectly solid cylinder, the maximum Von-Mises stress (VMS) obtained from a constant and specific load is approximately 16 MPa. By increasing porosity, the maximum stress is also increased. It is interesting to note that minimum stress on the base model is greater than the fracture strength obtained from the compression test, but both models are portrayed in "Fig. 5-b and 5-c", the minimum stress is less than the ultimate strength. According to the color contours that show the distribution of stress, most of the model experiences tensions that are less than the ultimate strength of matter. The model d, with spherical porosity, has experienced more uniform stress than two models in "Fig. 5-b and 5-c".

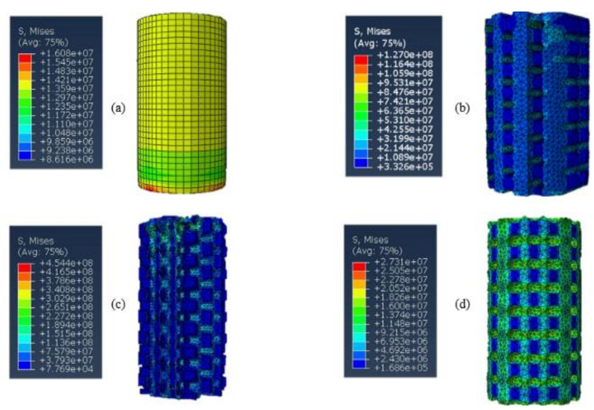

Fig. 5 Von-mises stress in: (a): base model without porosity, (b): first porous model, (c): second porous model and (d): spherical porosity model.

Since the spherical porosity does not create sharp points and there is no stress concentration, the results are reasonable and expected. It can be concluded, between spherical porosity and random, the Random model is more suitable because, in addition to providing empty space for blood exchange, protein and other materials, in most cases it experiences less stress than the ultimate strength. Ultimately, although some parts of b and c models are suffering immense tension that results in their destruction, it can be expected that scaffold can sustain the load as long as bone growth is achieved. In “Fig. 6 (a-d)”, the VMS, the axial stress, displacement, and strain-stress diagram of model b are presented. The slope of the graph representing the elastic modulus which is 250 MPa.

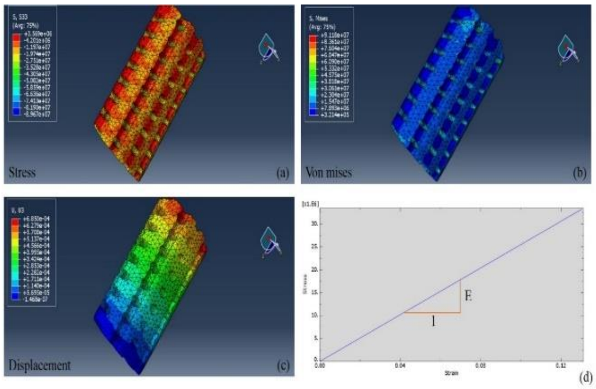

Fig. 6 (a): Von-Mises stress, (b): Axial stress, (c): displacement and (d): stress-strain diagram in the elastic region.

The maximum displacement of the model is less than one millimeter in size when considering the placement of the scaffold in the bone and the proper arrangements to be made to ensure that the scaffolding remains in place. Figure 7 shows the simulation of femur body with Abaqus software with the porous scaffold architecture under various load, the red area shows the maximum stress that the body can tolerate. The overall elastic and non-static properties of cortical and trabecular bone microstructure are derived from bone density, which can be estimated from the Hounsfield units, also it can be used from multiscale modelling of femur part. Figure 7 shows the simulation of femur body with ABAQUS software with the porous scaffold architecture under various load, the red area shows the maximum stress that the body can tolerate.

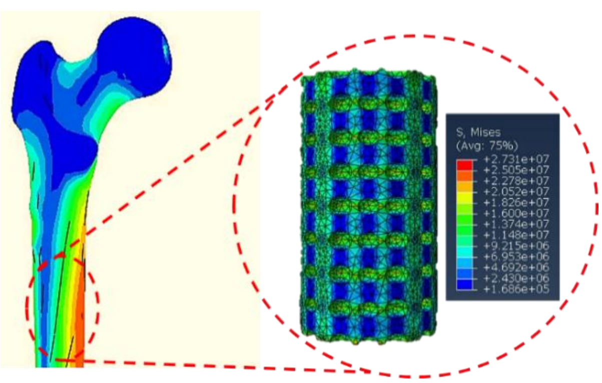

Fig. 7 Simulation of femur body with Abaqus software with the porous scaffold architecture under various load, the red area shows the maximum stress.

The overall elastic and non-static properties of cortical and trabecular bone microstructure are derived from bone density, which can be estimated from the Hounsfield units, also it can be used from multiscale modelling of femur part [25-31].

4 CONCLUSION

In recent years, research on new bone alternatives has focused on non-metallic composite materials, especially polymer/ceramic composites and nanocomposites. These (nanoscale) composites consist of polymer based and biologically active micro/Nano-fillers that provide biologic and mechanical properties suitable for bone substitutes. These materials have sufficient advantages to the polymers (structural stability, strength, biological compatibility, and optimal shape) rather than ceramics that are similar to bone microstructure.

Two primary parameters in the production of nanocomposites with similar properties to the bone can be convenient adhesion in the interface between organic polymers and inorganic HA and the uniform dispersion of a uniform Nano particulate HA in the polymeric matrix. The lack of adhesion between the HA particles and the polymeric substrate is usually leading to early failure at the interface of the two phases. On the other hand, in the absence of uniform dispersion of HA with suitable processing methods on the polymeric field, HA agglomerates and causes poor mechanical properties of the composite material.

In the last decade, many attempts have been made to produce polymer/nanoHA systems similar to bone structure. Another important consideration is the use of scaffold in optimizing, rebuilding and repairing the target tissue so that the first cells that are planted on the scaffold begin to divide and fill the area of implantation. Many tissues of the human body are under stress and mechanical strain; therefore, it is vital that the mechanical properties of the scaffold are close to that of the tissue that is supposed to be replaced. The mechanical properties of a scaffold made using a space holder is influenced by various factors such as the temperature of the sintering, the punch pressure, the size and shape of the space holders, and the percentage of use of them.

In this research, the mechanical behavior of these scaffolds has been evaluated using different micromechanical methods. The results of the simulation indicate that porous tissue can be useful for bone repair. More porosity means the better transfer of materials and blood inside the tissue. On the other hand, according to the simulation results, the increase in the porosity yields tension increase in the scaffold. Therefore, a balance between interchangeability and strength should be established. Although simulation results predict the durability of the scaffold composites, it should be ensured by verifying the results with animal experiments. Other proposed methods for enhancing strength along with increasing porosity can be adding other materials to the HA.

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