# Tensile Strength of 3D Printing Scaffold Design Truncated Hexahedron for Tuberculosis Drug Delivery

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*Engineering Innovations*
*ISSN: 2813-1002, Vol. 4, pp 43-48*

*Submitted: 2022-07-24*
*Revised: 2022-09-10*
*Accepted: 2022-09-30*
*Online: 2023-02-15*

# Tensile Strength of 3D Printing Scaffold Design Truncated Hexahedron for Tuberculosis Drug Delivery

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**Keywords:** 3D printing scaffold, tensile strength, Injectable Bone Substitute, tuberculosis

**Abstract.** Mechanical properties are important characteristics of scaffolds as biomaterials implant in tissue engineering. This study focused on the analysis of the tensile strength of the 3D printing scaffold with a geometric design of the truncated hexahedron unit with pore size variation and combined with Injectable Bone Substitute (IBS) paste for treatment of spinal tubercolusis. Five variations of pore size of the scaffold (600, 800, 1,000, 1,200, and 1,400 μm) were fabricated from Polylactide acid (PLA) filament using the Fused Deposition Modelling (FDM) method through an ordinary commercial 3D printer. The IBS paste was synthesized from hydroxyapatite (HA), gelatin, hydroxypropyl methylcellulose (HPMC), and streptomycin. The characterization performed in this study were the pore size test with a digital microscope, tensile strength, elongation test, porosity, and contact angle. The 3D printed scaffold formed micropores after injected with IBS paste from a range of 130-230 μm. The tensile test results showed that the tensile strength of the 3D printing scaffold increased after being injected with IBS paste. In addition, the elongation test also shows a positive trend with increasing values of elongation after injection of IBS paste. The contact angle test results indicated that the scaffold was hydrophilic. From those characterizations, it could be concluded that 3D printing scaffold meet the criteria of scaffold for bone tissue engineering and drug carrier for tuberculosis.

## 1. Introduction

Tuberculosis (TB) is a communicable disease that is a major cause of ill health and one of the leading causes of death worldwide. TB is caused by the bacillus Mycobacterium tuberculosis, which is spread when people who are sick with TB expel bacteria into the air. World Health Organization reported that an estimated 9.9 million people suffered TB in 2020, and Indonesia is one of the largest TB sufferers in the world [1]. Generally, MTB infected the lungs, but it can also infect organs outside the lungs and is called extra-pulmonary tuberculosis. The most common type of extra-pulmonary tuberculosis involves the spine or spinal tuberculosis and is known as Pott's disease.

The bacteria of tuberculosis will cause bone destruction and risk of spinal cord compression [2]. The effective treatment of spinal tuberculosis is a surgical treatment to remove the spinal deformities and replaced them with the scaffold. The development of industrial revolution 4.0 offers an efficient way to make scaffold using a 3D printer. Scaffold is a medium that provides an environment for support stem cells to conduct processes of adhesion, proliferation, and differentiation that will produce the desired new tissue [3]. Scaffold combined with Injectable Bone Substitute (IBS) paste can be used as a drug carrier consisting of Hydroxyapatite (HA), gelatin, Hydroxy Propyl Methyl Cellulose (HPMC), and Streptomycin, and was proven to be non-toxic and antibacterial [4].

The scaffold was fabricated using the 3D printing Fused Deposition Modeling (FDM) method, based on the principle of layer by layer display to construct a complete 3D material [5]. The 3D geometry model was designed from a Computer-Aided Design (CAD) application, using SolidWorks software. The scaffold requirements were biocompatible, and the physical structure of the scaffold must also qualify for the biological function of the Extracellular Matrix (ECM), which was influenced by the geometric design and pore size of the scaffold [6].

One of the requirements for scaffold as a bone implant is to meet the requirements of human mechanical properties. In tissue engineering scaffolds, porosity is a crucial parameter that affects cell proliferation, attachment, migration and differentiation [7]. The truncated hexahedron design means an intersecting hexagon shape. The intersection creates a new support, and the design appears to be octagonal. Through the addition of struts, it causes an increase in the support load and a reduction in the volume of the scaffold cavity. Based on this description, the strength of the scaffold is influenced by the pore size and geometric design. This study was aimed to synthesize a 3D printing scaffold, then combine it with IBS paste to improve its effectiveness in the treatment of spinal tuberculosis, tensile test was carried out to determine the tensile strength of the scaffold so that it can withstand loads that meet the characteristics of human bones, and contact angle test was also conducted to determine its biocompatibility when interact with human body solution.

## 2. Materials and Method

### 2.1 Materials

The materials used in this study included Poly Lactic Acid (PLA) filaments, the ingredient of IBS paste including hydroxyapatite from bovine bones from PT Inobi, Surabaya, Indonesia, Gelatin (150 bloom Rousselot, Guangdong, China) from cowhide, Hydroxy Propyl Methyl Cellulose (HPMC) (Sigma Aldrich H7509, Singapore), and streptomycin sulfate in 1 gram vial purchased from PT. Meiji Indonesia.

### 2.2 Manufacturing of 3D printing Scaffold

The manufacturing of scaffold was designed using the Solidworks 2014 application. The pore size was determined by the distance between the fiber (strut). The design was carried out by making a unit cell of a truncated hexahedron with a 2 mm width. The process of the design begins by making octagon shapes on the front, top, and right. Then flipping planes at an angle of $90^0$, so the design framework is obtained. All of the sides are swept using Swept Boss/Base, so a truncated hexahedron unit cell is obtained (Fig. 1).

The process of merging or assembly is carried out on the unit cell to obtain a geometric design. The tensile test was designed to follow ASTM D638 (Fig. 2) using a ratio of 1:0.5 [8]. The extension of the Solidworks 2014 application in the form of *.stl* form, via the Cura from Ultimaker application by looking at the slicing parameters of the design. The designed scaffold will be printed using a 3D printing machine with the FDM method with the nozzle used having a hole diameter of 0.4 mm. The filament used is Polylactic Acid (PLA) with a diameter of 1.75 mm. Strut size and distance between struts of designed scaffold can be seen in Table 1.

Table 1. Strut size and distance between struts of designed scaffold

| Pore size of Scaffold, [µm] | Distance between strut, [mm] | Width of scaffold, [mm] | Radius of strut,[mm] |
|---|---|---|---|
| 600 | 0.6 | 2 | 0.7 |
| 800 | 0.8 | 2 | 0.6 |
| 1000 | 1 | 2 | 0.5 |
| 1200 | 1.2 | 2 | 0.4 |
| 1400 | 1.4 | 2 | 0.3 |

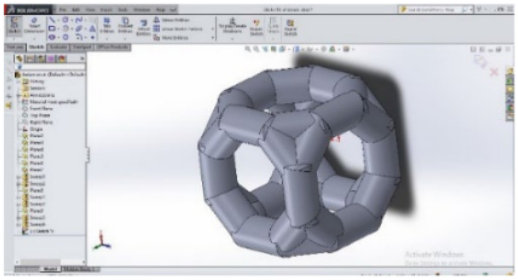

Figure 1. Unit design of bone scaffold

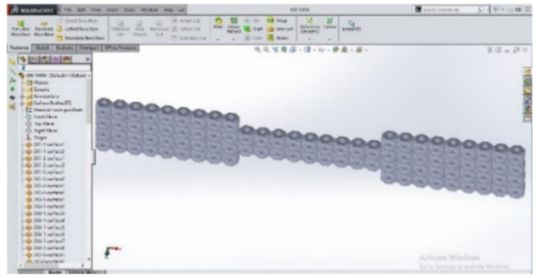

Figure 2. Design of tensile test sample

### 2.3 IBS Paste Synthesis

IBS paste synthesis was conducted according to previous research [4]. At first, 20% w/v gelatin was dissolved in distilled water at 40°C for 1 hour. At the same time, HPMC 4% w/v was dissolved in distilled water at 90°C for 1 hour, then the solution was cooled until the temperature became 40°C. In the dissolved gelatin solution, hydroxyapatite was added with a composition ratio of HA: gelatin 65:35 w/w and stirred for 1 hour. After the solution became homogeneous, the next step was added streptomycin as much as 10% of the total mass of HA, gelatin, and HPMC, and dissolved it again until the solution became homogeneous. In the last step, the HPMC solution was added to the HA-gelatin-streptomycin at a mixing temperature of 40°C for 6 hours.

### 2.4 Sample Characterization

Scaffold pore size test was conducted using the digital microscope to determine the pore size formed on the scaffold both before and after the injection of IBS paste. The porosity test was conducted to determine the percentage of pores formed in the scaffold. The density of PLA is 1.24 g/cm$^3$ [9]. The mechanical strength of the scaffold was obtained by performing tensile tests both on the scaffold without and after injection of IBS paste. The tensile strength and elongation can be calculated from stress-strain graph. The porosity value of the scaffolds was measured by liquid displacement. In this method ethanol was used as the displacement liquid because it penetrated easily into the pores. The contact angle test was conducted to determine the hydrophilicity and biocompatibility of the scaffold.

## 3. Result and Discussion

### 3.1 3D Printing Scaffold Injected with IBS Paste

The results of the 3D Printing scaffold macroscopically showed that it has good interconnectivity between pores. While the pore size design is 600; 800; 1,000; 1,200 and 1,400 µm, after printed, measured and produced 610; 820; 1,020; 1,210 and 1,400 µm on average respectively. Then scaffold was injected with IBS paste for drug carrier application in the treatment of spinal tuberculosis. In the scaffold that has been injected with IBS paste, the average micropores are 130-230 µm. The porosity test obtained the data of porosity respectively 38.18%, 44.47%, 49.28%, 52.99% and 56.31%.

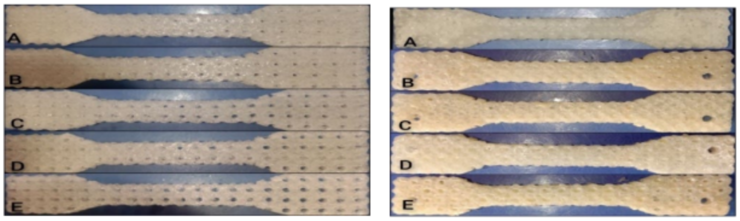


Fig. 3. Samples of 3D printing scaffold for tensile test (a) before and (b) after IBS Paste Injected A) 600 , B) 800 µm, C) 1,000 µm, D) 1,200 µm, E) 1,400 µm

### 3.2 Tensile Test

The tensile test was conducted to determine the maximum stress that could be received from the scaffold before fractured with applied axial force. The tensile test was conducted 2 times for each variation of the scaffold without IBS and after IBS paste injection. The tensile strength test resulting a graph of stress and strain as shown in Fig. 4 for the PLA scaffold, before and after injected with IBS paste. And Fig. 5 and 6 shown both the tensile strength and elongation.

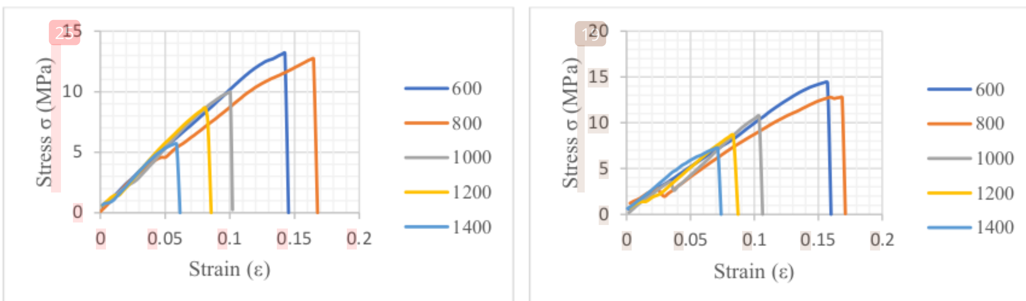


Fig. 4. The tensile strength (a) before and (b) after IBS Paste Injected

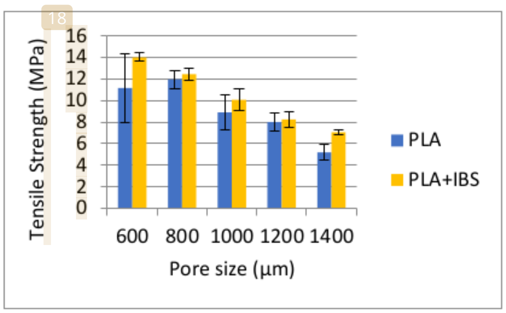


Fig. 5. The tensile strength test result

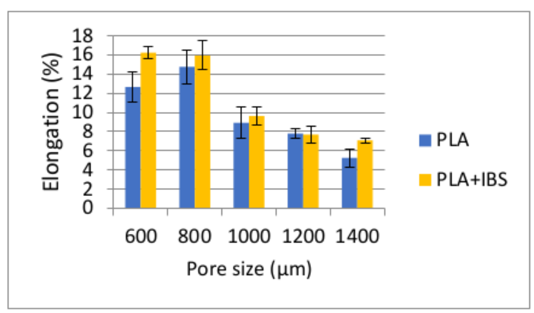


Fig. 6. The elongation test result

The tensile strength and elongation of the scaffold decreased with increasing pore size. The tensile strength value was higher for scaffolds injected with IBS paste, from 5.176-11.954 MPa to 7.047-13.993 MPa and elongation from 5.247-14.743% to 7.071-16.224%. Based on these results, the trend of the relationship between pore size and tensile strength and elongation is obtained. Based on the literature [10], the ultimate tensile strength of vertebral trabecular bone is (2.23 ± 0.76) MPa. So that based on this results, the tensile strength of the scaffold qualifies for the use of tissue engineering. The mechanical properties determine the load of scaffold that can withstand to support the human body. IBS paste can increase the tensile value because IBS paste fills the volume of the cavity in the scaffold so that the tensile support area becomes larger.

Based on research conducted by [11], a study of the effect of pore size on the mechanical properties of the box design scaffold with a pore size range (50-1000 μm) was carried out using PLA filament. The result was obtained that a negative correlation of pore size and tensile strength. Scaffold to qualified bone repair requirements, smaller pore sizes could be beneficial for the mechanical strength of the porous scaffold. This is followed by the results obtained in this study, which a negative trend relationship between pore size and tensile strength.

3.3 Contact Angle Test

The contact angle test was conducted using the digital microscope and supported by using the HiView application to observe the angle formed between the liquid and the scaffold. Based on the results of the contact angle, showed that the scaffold which before and after IBS paste injected was hydrophilic with a contact angle of less than 90°. There are variations of the pore size of the scaffold that obtained the effect of pore size on the mechanical properties of the scaffold. The porosity of the scaffold is required for supporting the growth of new bone cells. The scaffold porosity of 30-50% was qualified to support cell proliferation [12]. Based on the data of this study, the porosity of 38.18-55.86% means that the scaffold suitable for its use in supporting the regeneration of bone cells.

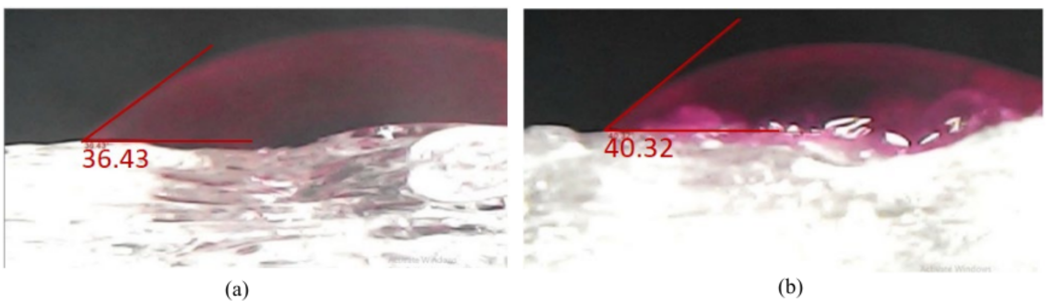


Fig. 7. The contact angle test results (a) before IBS Paste Injected, (b) after IBS paste injected

The contact angle result was obtained that scaffold has hydrophilic behavior with angle of 22.91°-44.55° for scaffolds without IBS paste and 25.48°-54.44° for scaffold injected by IBS paste. The scaffold was hydrophilic and qualified to be applied in tissue engineering. The hydrophilic behavior shows that the scaffold has good biocompatibility properties when being applicated in the body.

## 4. Conclusion

3D printing scaffolds with various pore sizes were synthesized with porosity of 38.18-55.86%, that could support the cell proliferation process. Hydroxyapatite combined with gelatin, HPMC, and streptomycin, then injected into the 3D printing scaffold as a means of drug carrier and a method of treating spinal tuberculosis. The tensile strength value was higher for scaffold which injected with IBS paste, from 5.176-11.954 MPa to 7.047-13.993 MPa and elongation from 5.247-14.743% to 7.071-16.224%, porosity values 38.18-56.31% and contact angle 22.91°-44.55° for scaffold without IBS paste, and 25.48°-54.44° for scaffold injected with IBS paste. This result indicated that the scaffold was hydrophilic. From those characterizations, it could be concluded that 3D printing scaffold qualified the criteria of scaffold for bone tissue engineering and suitable method for drug delivery in spinal tuberculosis treatment.

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**Acknowledgments**

The authors would like to thank Universitas Airlangga for the funding of this research [Decree number 8/E1/KPT/2021 and Contract agreement number 4/E1/KP.PTNBH/2021 and 677/UN3.15/PT/2021].

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