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## Synthesis of biogenic silica nanoparticles from rice husks for biomedical applications

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#### Abstract

Silica nanoparticles have numerous potential applications and can be manufactured in several forms, including fumed silica, colloidal silica, silica gel, and silica aerogel. However, biogenic silica is an excellent alternative to synthetic silica because of its variable structure, density, and composition. Among the available agricultural bioresources, rice husk is considered to be a cost-effective and non-metallic bio-precursor for biogenic silica nanoparticle synthesis. In this investigation, we synthesized biogenic silica nanoparticles (bSNPs) using rice husk as a precursor. Under pressurized conditions, the rice husk was acid pretreated to remove inorganic impurities and induce the hydrolysis of organic substances. Residues from the acid pretreatment were calcinated at different temperatures for 1 h. The structure and morphology of the synthesized biogenic nanoparticles were analyzed using transmission electron microscopy (TEM). Fourier transform infrared (FT-IR) spectra of the synthesized samples showed O–Si–O stretching vibrations between 1056 and 1078 cm<sup>-1</sup>. The X-ray diffraction patterns of the bSNPs showed the presence of amorphous biogenic silica. The TEM images suggested that irregular particles with dimensions between 10 and 30 nm had formed. During the calcination process, the primary particles were aggregated; however, upon increasing the calcination temperature, the sizes of the primary particles decreased. The biological properties of bSNPs were studied by MTT assay and by assessing cellular morphological changes in human mesenchymal stem cells (hMSc). The cell viability studies revealed that the bSNPs had excellent hMSc biocompatibility. Our results revealed that these bSNPs can be used in bone tissue engineering.

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Keywords: Rice husk; Human mesenchymal stem cells; Biogenic silica; Biocompatible

### 1. Introduction

Nanotechnology has become an active and vital area of research that is growing rapidly in industrial sectors such as electronics, aerospace, defense, medicine, and dentistry. Nanotechnology involves the design, modeling, fabrication, characterization, and manipulation of materials at the nanometer scale [1,2]. Nanomedicine is an emerging area of research in

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nanotechnology that entails detecting, inhibiting and treating diseases at the molecular level using nanodevices and nanostructures [3]. Nanoparticles (NPs) including noble metals, semiconductors, magnetic materials and their composites have promoted a revolution in biomedical applications, such as drug carrier, gene delivery, labeling and tracking agents, sensors, hyperthermia, tissue engineering, wound healing and bioimaging [4–6]. Interestingly, silica-based NPs provide a versatile toolbox for biomedical applications due to their biocompatibility and bioactivity [7–9]. Trifunctionalized mesoporous  $SiO_2$  NPs have been used to image, target and treat cancer [10]. Manzano et al. reported the use of aminefunctionalized mesoporous silica microspheres for the controlled drug delivery of ibuprofen [11]. According to Csogor

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et al., silica NPs functionalized with different alkoxysilanes and amines can control gene delivery [12]. Organic-modified silica has been widely used to separate pyridylamino sugars from proteins and nucleosides [13]. Silicon plays a crucial role in bone mineralization, collagen synthesis, skin aging and the appearance of hair and nails [14]. Therefore, silicon deficiency causes damaged hair, wrinkled skin, brittle nails and abnormalities in the structure of bone and cartilaginous tissue [15,16]. Silica taken in through diet translocates through the gut and is found in the connective tissue, bone and blood vessels [17]. Several studies have demonstrated that dietary silica supplementation improved bone formation and bone health. Jugdaohsingh et al. suggested that high-concentration silicon intake was associated with increased bone mineral density in men and younger women [18]. Soluble silicon has been shown to enhance collagen type 1 synthesis and osteoblastic differentiation in human osteoblast-like cells [19]. Keeting et al. proved that silica containing zeolite A material induced the proliferation and differentiation of the osteoblast lineage [20]. Wiens et al. reported that biosilica-induced osteoprotegerin expression resulted in an increase in the level of cytokines and prevented osteoclast differentiation through the abolition of RANKL function [21]. Previous studies have suggested that Si (silica) from seawater triggers the differential expression of several genes, including bone morphogenetic protein-2 (BMP-2), runt-related transcription factor 2, collagen type-I, osteoprotegerin and RANKL, which results in stimulated osteoblast development [22].

Recently, natural-resources-based silica has garnered considerable interest in the materials science and biomedicine fields because of the availability, low cost and eco-friendliness of these materials. A number of living organisms, including higher plants, mollusks, sponges and protozoa, have evolved to produce biogenic silica at the astounding rate of gigatons/year [23]. Rice husk is an agro-waste product, and approximately 600 million tons are generated each year around the world [24]. However, the utilization of rice husk (RH) is extremely low due to its adverse properties, such as low nutritive value, resistance to degradation and high ash content [25]. Moreover, RH contains 15-20% inorganic (SiO<sub>2</sub>) and 65–75% organic substances (lignin, cellulose and hemicellulose) [26]. Biogenic silica nanoparticles have been prepared from RH using a number of methods, including sol-gel processes [27], microwave hydrothermal processes [28], flame synthesis [29], and combustion synthesis [30]. However, most silica production methods are time consuming and produce low-purity silica. In addition, the knowledge of bSNPs biological properties remains poor. Hence, in this study, we propose a simple method to prepare high purity biogenic silica nanoparticles (bSNPs) from RH and evaluate their biocompatible properties using in vitro cell-based approaches. The bSNPs were well characterized by X-ray diffraction, Fourier infrared spectroscopy, Energy-dispersive X-ray spectroscopy, Photoluminescence spectroscopy and TEM. Furthermore, we used human mesenchymal stem cells as in vitro models to assess biocompatibility because of the multipotent ability of these stem cells to differentiate into multiple lineages, including osteogenic, adipogenic and chondrogenic lineages [31]. In this preliminary study, bSNPs exhibited excellent biocompatibility with hMSc, and they may induce osteogenic and chondrogenic differentiation.

#### 2. Materials and methods

#### 2.1. Chemicals

Rice husk was procured from rice mills in India. Analytical reagent-grade hydrochloric acid (HCl) was purchased from Merck, Saudi Arabia. Milli-Q water was used throughout all experiments. All chemicals were used as supplied without further purification. DMEM was procured from ATCC (USA). 3-(4, 5-Dimethylthiazol-2-yl)-2, 5-diphenyltetrazolium bromide (MTT), dimethyl sulfoxide (DMSO) and fetal bovine serum (FBS) were purchased from Invitrogen (Carlsbad, CA, USA). Propidium iodide (PI) was purchased from Sigma-Aldrich (St. Louis, MO, USA).

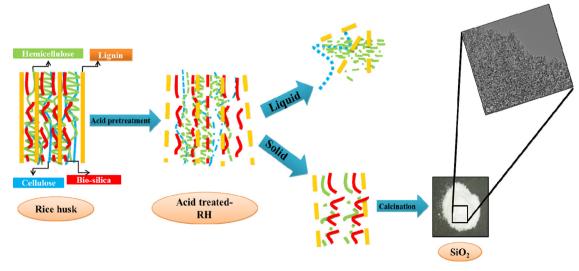


Fig. 1. Schematic of the formation mechanism for biogenic silica nanoparticles.

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