

# Production of Apatite from Snail Shells for Biomedical Engineering Applications

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## Abstract

Bioceramics is very important application for dental and orthopedic procedures. Beside all these normal procedures traffic accidents are requiring increasing number of graft, prostheses and [orthosis](#) applications. Bioceramics can be produced from local and natural sources with various methods. Those can be produced from various bone structures through calcination (at high temperatures) or with diluted hydrochloric acid (HCl) application & freeze drying. Beside these methods calcite and aragonite structures like from sea shells and egg shells bioceramic production can be realized through mechanochemical processing via a simple hot-plate or ultrasonic equipment. A fresh water snail shell (Zebra Nerite Snail - *Neritina natalensis*) was prepared as bioceramic production source. The resulting hydroxyapatite (HA) powders were obtained without any impurities. At two varying temperature of 865 and 885 °C the snail shells was transformed to HA bioceramics. Scanning electron microscopy (SEM), X-ray diffraction (XRD) and differential thermal analysis (TG/DTA) were evaluated.

## Introduction

Calcium phosphate-based bioceramics such as HA have received much attention owing to their high biocompatibility, ability to be utilized as synthetic osteoconductive scaffolds in bone and with numerous clinical applications [1]. Beside orthopedic applications those bioceramics are also in use for various dental surgery applications (i.e. graft material for periodontal pockets, coating material and dental implants) and aesthetic surgery applications such as facial, breast and body contouring applications. Also with increasing in traffic accidents biomaterials are gaining a big role for human health reconstruction in the field of orthopedic application.

Aging of the world population and increasing living standards have also boosted the increasing development of the orthopedic industry. Emerging materials, high technologies, applications and procedures will challenge the future market as reported in Ace Business Report [2].

HA based on biomaterials can be produced with various methods from synthetic and natural sources. One of the basic technics is convention precipitation method from reagent grade chemicals. This method is a time consuming method. Natural species of sea origin, such as sea shells, coral and snail shells always receive much attention in biomedical engineering applications. Freeze drying and with diluted HCl treatment are also convention ways for HA graft production however prion diseases such as mad cow and related diseases can be very dangerous, risky and deadly [3-4]. Calcination of natural materials (which posses calcium phosphates) at high temperatures (850° C) [3-4] is another interesting method. From human [3], bovine, [4] sheep [5], chicken bones [6] HA, human-bovine-sheep tooth structures were used as obtaining of biomaterials such as dentine [7-8] and enamel [8-9] Natural HA structures can be produced very economically. HA biomaterial can also obtain from fish scales [10]. Lately natural HA is produced from calcitic aragonitic sources with different methods mechanochemically using ultrasonic equipment [11-12] or a hot-plate stirrer [12].

Calcite and aragonite process material were converted to HA and other related phases the shells of Zebra Nerite Snail (*Neritina natalensis*), which was used (Fig.1). Zebra Nerite Snail shells are approximately 8 and 16 mm in diameter [13].

The aim of this presented study is to obtain HA powders from snail shells using mechanochemical method via hot-plate. Characterization of HA transformed from snail shells sources follow to the evaluation of the final product, which can be used in biomedical engineering.

## Materials and Methods

The shells of Zebra Nerite Snail were used (Fig. 1). Empty zebra nerite snail shells (ZNSS) were obtained from a local gift store in Istanbul – Turkey. ZNSS were washed under tap water from possible dirt remains. Cleaned ZNSS were soaked into distilled water, washed again and dried. After drying shells were ball grinded and sieved through 100 µm sieve.



Fig. 1. Photograph of Zebra Nerite snail shells (*Neritina natalensis*).

The production method, differential thermal and gravimetric analyses (DTA/TGA) of ZNSS were conducted at heating rate of 20 °C/min between 50 and 1000 °C under stream flow of nitrogen gas (Perkin Elmer Diamond TG -TDA). According to a previous study [15]

After completing the production procedure X-ray diffraction (XRD) and scanning electron microscopy (SEM) studies were conducted.

## Results and Discussion

The DTA and TG curves for the ZNSS samples after drying are revealed in Fig. 2. Fig. 2 reveals the weight loss of 43.04% owing to thermal decomposition of calcium carbonate to calcium oxide and carbon dioxide at 861.6 °C, which is the main composition of ZNSS. The weight loss owing to organic matter was 1.55%. The weight loss along with endothermic peak at 700-870 °C indicates the decomposition of calcite reaction. It is confirmed that the thermal analysis ZNSS mainly contains CaCO<sub>3</sub> along with small amount of magnetite and other organic matters.

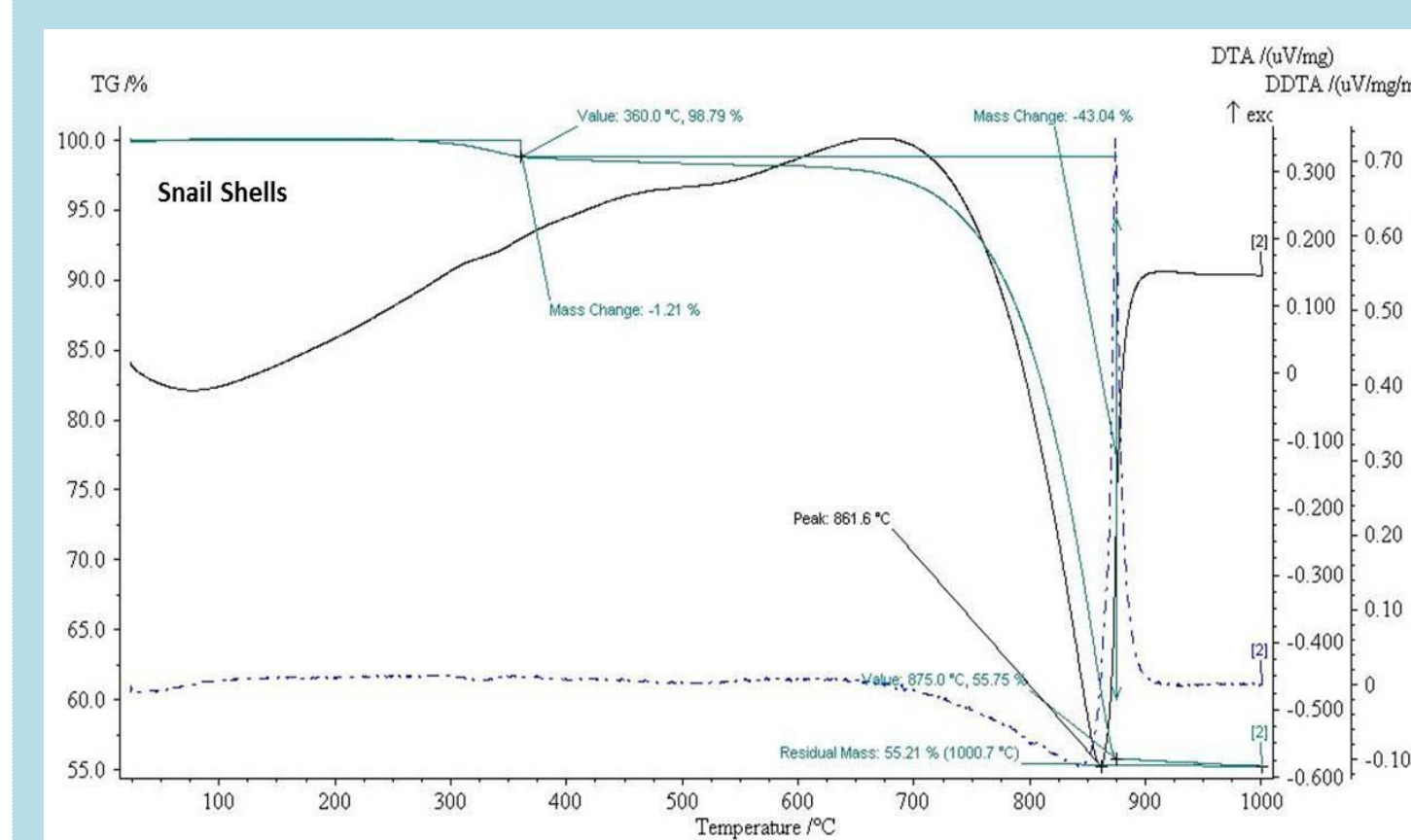


Fig. 2. TGA/DTA analysis of as raw powders of ZNSS after ball milling and sieving.

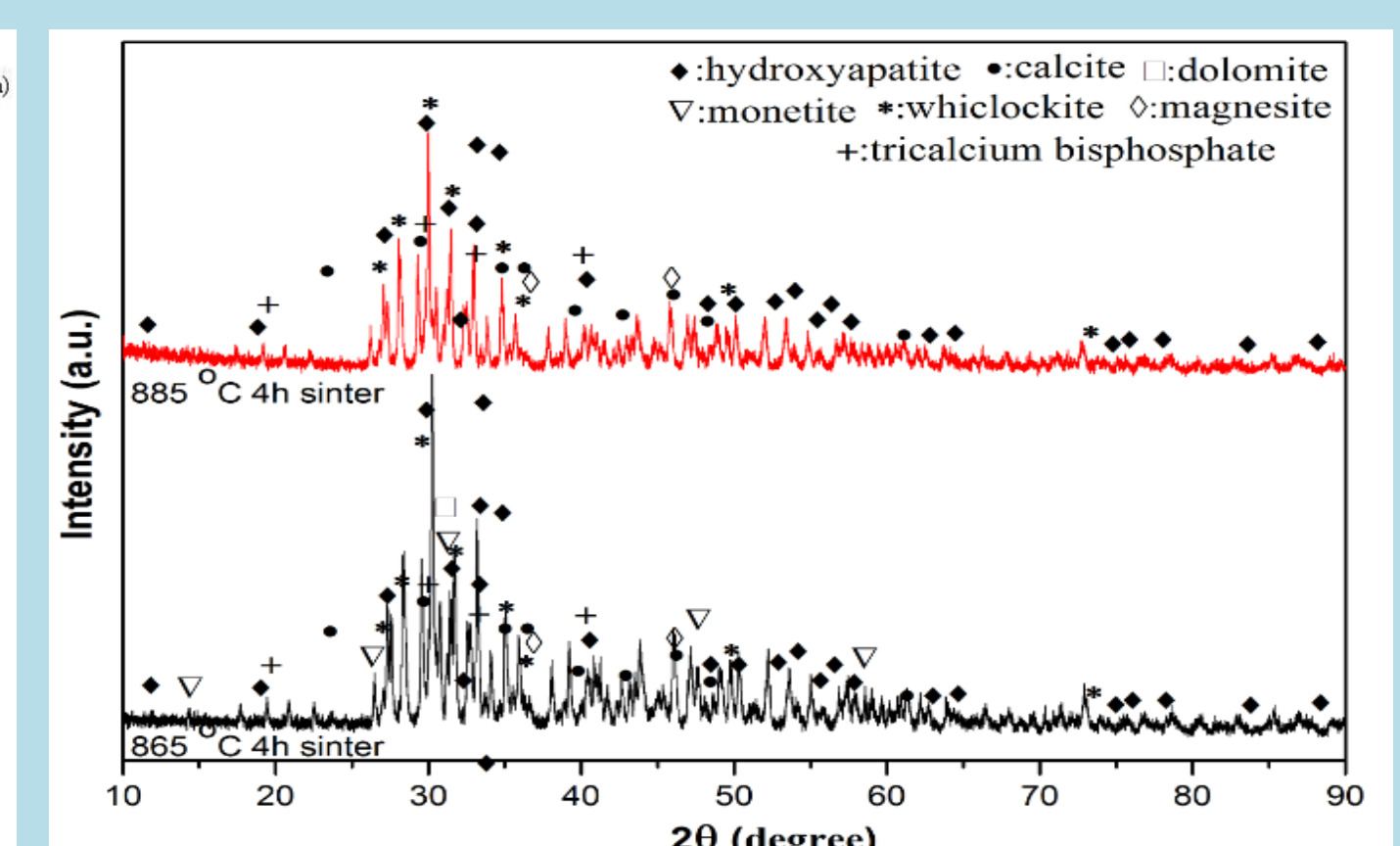


Fig. 3. X-ray diffraction diagrams of at 865 and 885 °C sintered sample in 4h

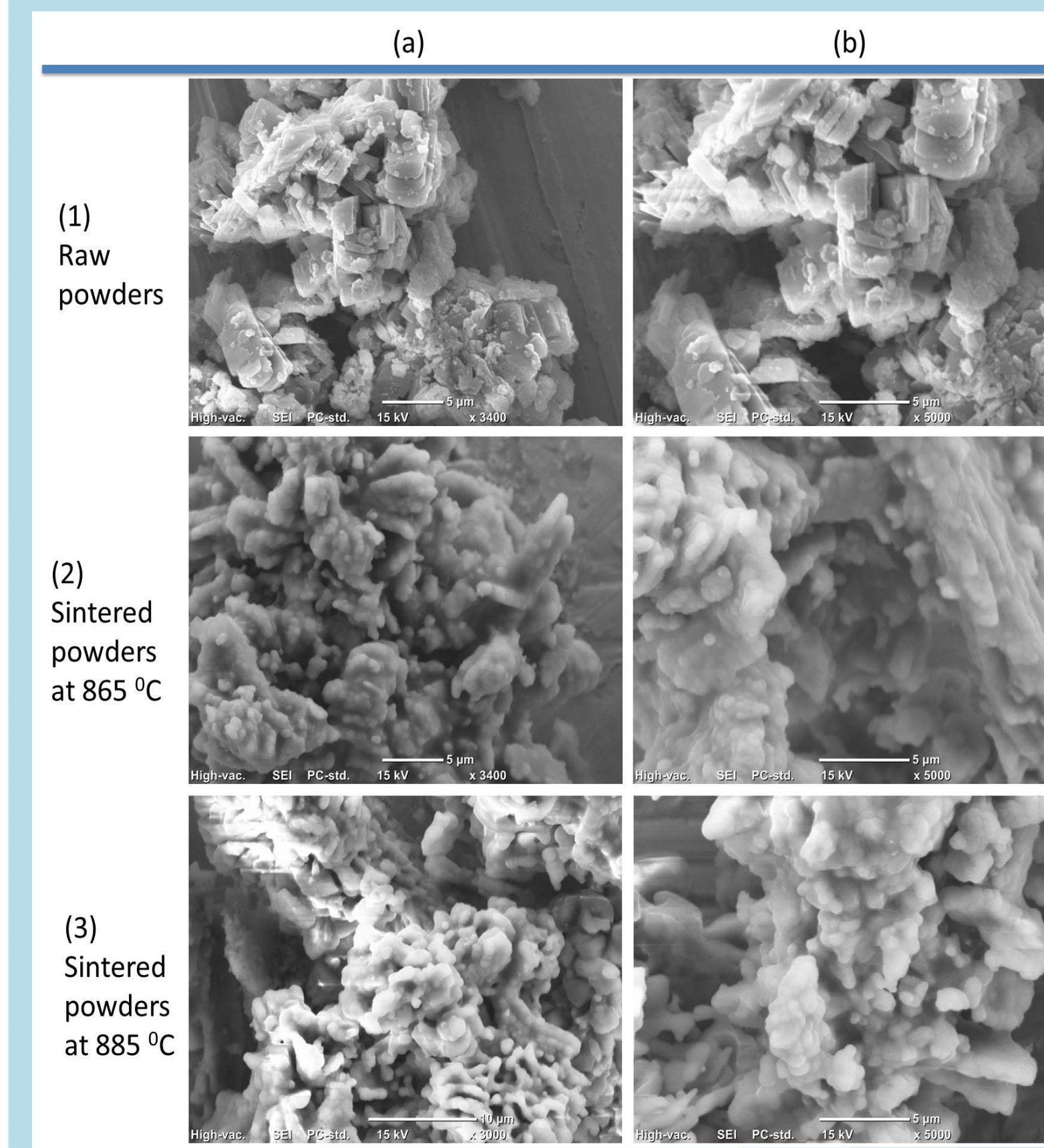


Fig. 4. Scanning Electron microscopy images (a) low and (b) high magnifications of (1) ZNSS raw powders, (2) ZNSS sintered powders at 865 °C and (3) ZNSS sintered powders at 885 °C.

SEM micrographs of the surface of the raw powders are shown in Fig. 4 1a-b. It is clear from Fig.4 1a-b that ZNSS consists mainly of calcite which is the most stable calcium carbonate. Agglomerates were also clearly observed as revealed in Fig. 4. The calcite particles are very large which is about 3µm x 3µm in square form and thin layer prismatic form on each other. After sintering, the morphology was converted to nanoparticles which are about 60-100nm in Fig. 4 2a-b, 3a-b.

## Conclusion

Hydroxyapatite powders were fabricated through mechanochemical process via a simple hot-plate stirrer from the fresh water sea snail shell (Zebra Nerite Snail - *Neritina natalensis*) powders. The produced biomaterials powders were obtained some nanoparticles after sintered at 865 and 885 °C. This process can be simple and economical in producing biomedical scaffold materials.

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