November 1999

ELSEVIER

Materials Letters 41 (1999) 128–134

MATERIALS LETTERS

www.elsevier.com/locate/matlet

Synthesis of hydroxyapatite from eggshells

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Received 27 November 1998; received in revised form 20 May 1999; accepted 21 May 1999

Abstract

A novel procedure to produce porous hydroxyapatite (HAp) from eggshells is reported. The process is carried out at an elevated temperature. HAp is the only apatite present in the reaction products, apart from minute fractions of certain other calcium compounds. The final product is characterized by X-ray diffraction and scanning electron microscopy (SEM).

Keywords: Hydroxyapatite; Eggshells; X-ray diffraction

1. Introduction

An eggshell is constituted, structurally speaking, by a three-layered structure, namely the cuticle, the spongeous layer and the lamellar layer. The cuticle layer represents the outermost surface and it consist of a number of proteins. Spongeous and lamellar layers form a matrix constituted by protein fibers bonded to calcite (calcium carbonate) crystals in a the proportion of 1:50.

Scanning electron microscopy (SEM) studies have demonstrated that the matrix fibers do not only surround the calcite crystals but pass through the crystals. Therefore, the matrix has a strong influence on the mechanical strength of the entire eggshell. The organic components of the matrix are complexes of mucopolysaccharide proteins, mainly constituted by chondroitin–sulphates A and B, glucosamine, galactosamine, galactose, mannose, fucose and sialic acid. The eggshell represents the 11% of the total weight of the egg and is composed by calcium carbonate (94%), calcium phosphate (1%), organic matter (4%) and magnesium carbonate (1%).

In Mexico, for example, the annual production is estimated as 7.0×10^{10} of units of eggs. Since each dozen has the average weight of 746 g, taking 11% of that weight, we arrive at approximately 480,000 tons at year (1300 tons daily). This material is basically useless after the production of eggs and egg derivatives.

Normally, manufacturers and factories store the material as an industrial residue that can contribute to pollution since it favors microbial action in the

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00167-577X/99/$ - see front matter
PII: S0167-577X(99)00118-4

environment. Occasionally, only some eggshells are used as a fertilizer due to their high content of calcium and nitrogen. Moreover, there have been studies aiming at the conversion of the eggshells into foodstuff for man and animal (chicken) use. This procedure consists in desiccating the eggshells, as soon as they become the final debris of egg production, in a heat chamber at 80°C to minimize pollution and eliminate most organic components. Flour is then produced by crushing and milling the eggshells to obtain fine particles.

In addition to providing a calcium source, the flour produced by this method has an added nutritional value due to proteins remaining from the albumin, membrane and matrix of the shell. However, some other interesting uses of this agriculture waste material remain to be explored. For instance, the possibility of producing hydroxyapatite (HAp) with the formula $Ca_{10}(PO_4)_6(OH)_2$, a biomineral which forms part of the isomorphic family of apatites. In biological systems, HAp represents the main inorganic component of the structure of bones and teeth [1,2]. Therefore, several studies have been reported on attempts to produce or synthesize materials with chemical characteristics similar to HAp, for implant or prosthesis purposes; there is a clear need for large, inexpensive amounts of clinical grade HAp [3–13]. Accordingly, the aim of the present work is to propose a simple method for producing HAp from eggshells.

2. Experimental procedure

Eggshells were collected and their surfaces mechanically cleaned. They were then placed in an oven for a two-stage thermal treatment. The first stage consisted of heating the eggshells to 450°C for 2 h at the heating rate of 5°C/min; at this temperature, any organic residue is expected to be destroyed. The second stage consisted of heating the samples to 900°C also for 2 h but with the heating rate of 0.5°C/min. At this temperature, the eggshells trans-

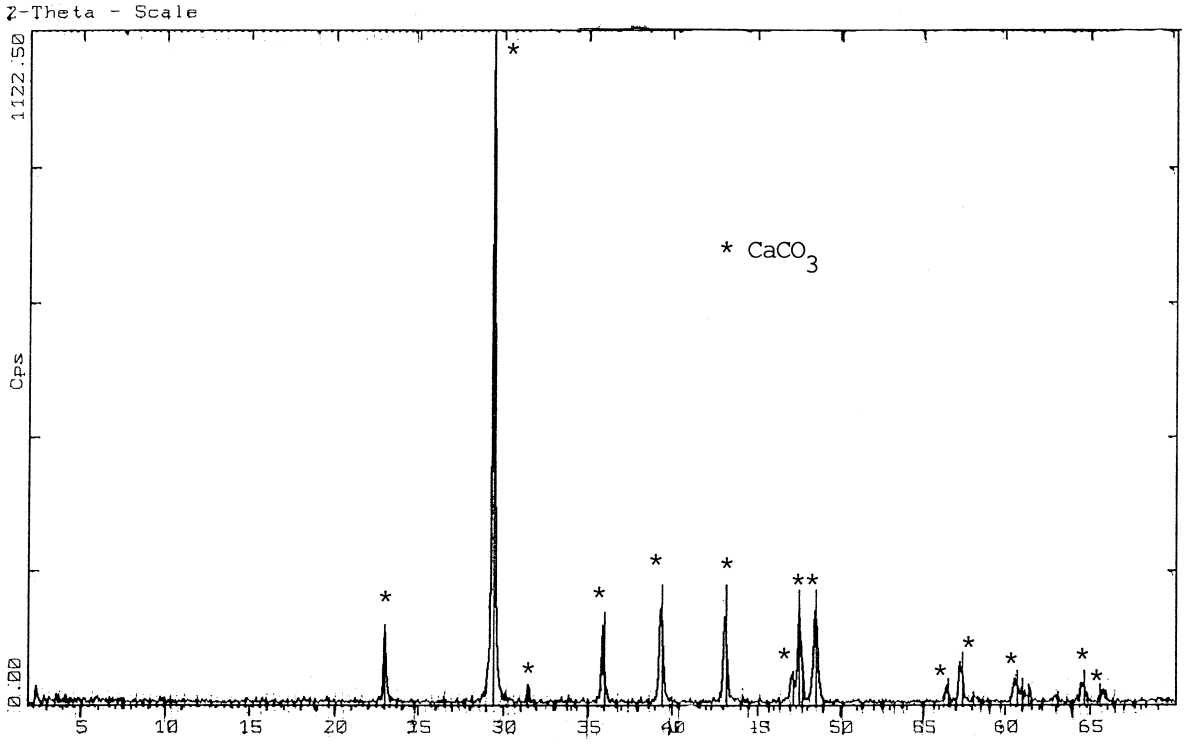

Fig. 1. The X-ray diffraction pattern of the raw eggshell material.

form into calcium oxide by freeing carbon dioxide (CO_2) according to the following equation:

$$CaCO_3 \rightarrow CO_2 + CaO \quad (1)$$

The CaO so obtained from the eggshells was then transformed into HAp in a phosphate solution following a procedure elaborated before by Roy [14] and subsequently used by others [15,16]. The reactant concentrations were simply determined by the fact that 5.54 g of CaO correspond to 1 g of $Ca_3(PO_4)_2$ and also by the Ca/P ratio for HAp equal to 1.67. Then the CaO from eggshells was added to the solution in a container designed to reproduce a moist atmosphere. Subsequently, the container was sealed and heated to 1050°C for 3 h at the heating rate of 10°C/min. The expected reaction is:

$$3Ca_3(PO_4)_2 + CaO + H_2O \rightarrow Ca_{10}(PO_4)_6(OH)_2 \quad (2)$$

Once the reaction was completed, the solution was filtered and the resulting material dried overnight at 80°C in an oven.

3. Characterization of the products

X-ray powder diffractometry was carried out in a Siemens D-5000 diffractometer and SEM observations were performed in a JEOL 5200 apparatus on carbon-coated specimens.

The mineral phase present in the as-collected eggshells was identified as calcite; no other crystalline species was detected. As shown in Fig. 1, the X-ray reflections correspond to the JCPD files for calcite (5-0586). The material obtained after the thermal processing at 900°C has a porous appearance, white color and fragile consistence.

The corresponding X-ray diffraction pattern (Fig. 2) shows reflections corresponding to JCPD 37-1497 files for CaO; again no other species was found. The observed change in the weight corresponds to the loss of gaseous CO_2.

Thermal processing of CaO in the phosphate solution at 1050°C produces a solid material with porous texture, white color, high mechanical strength and pores with irregular diameters. X-ray diffraction of this sample shows several species (Fig. 3). The

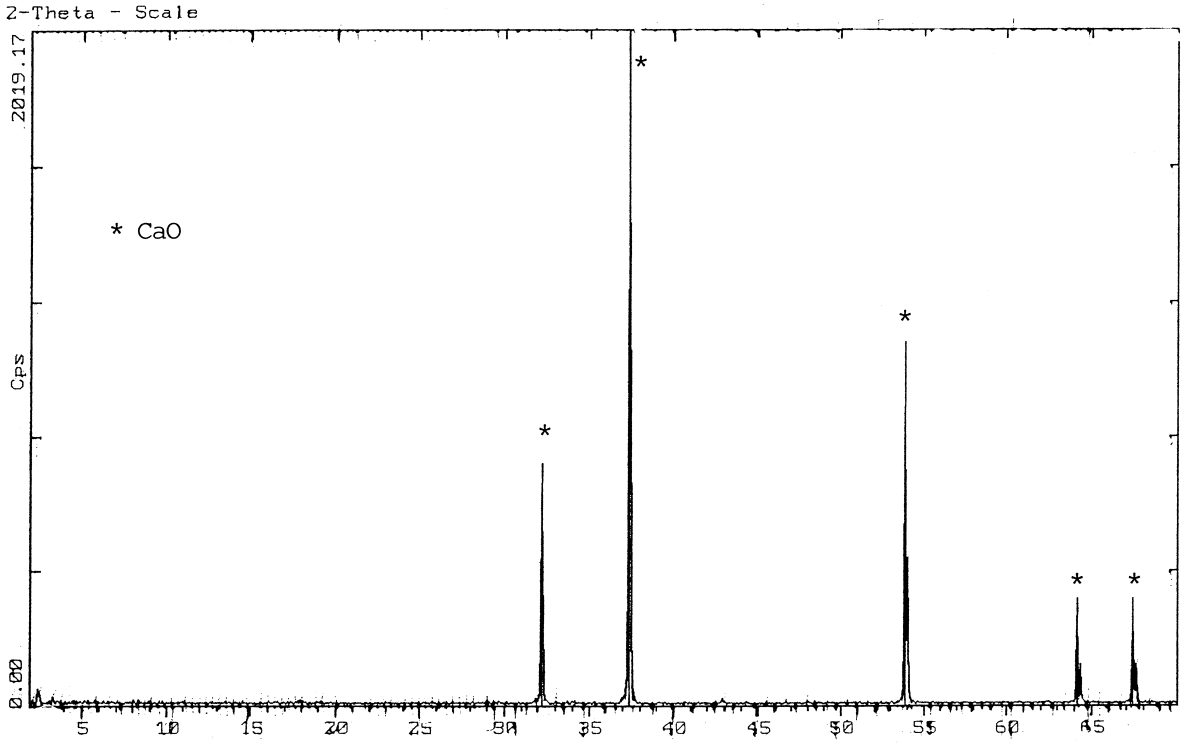

Fig. 2. The X-ray diffraction pattern for the same sample as in Fig. 1 but after the thermal treatment at 900°C.

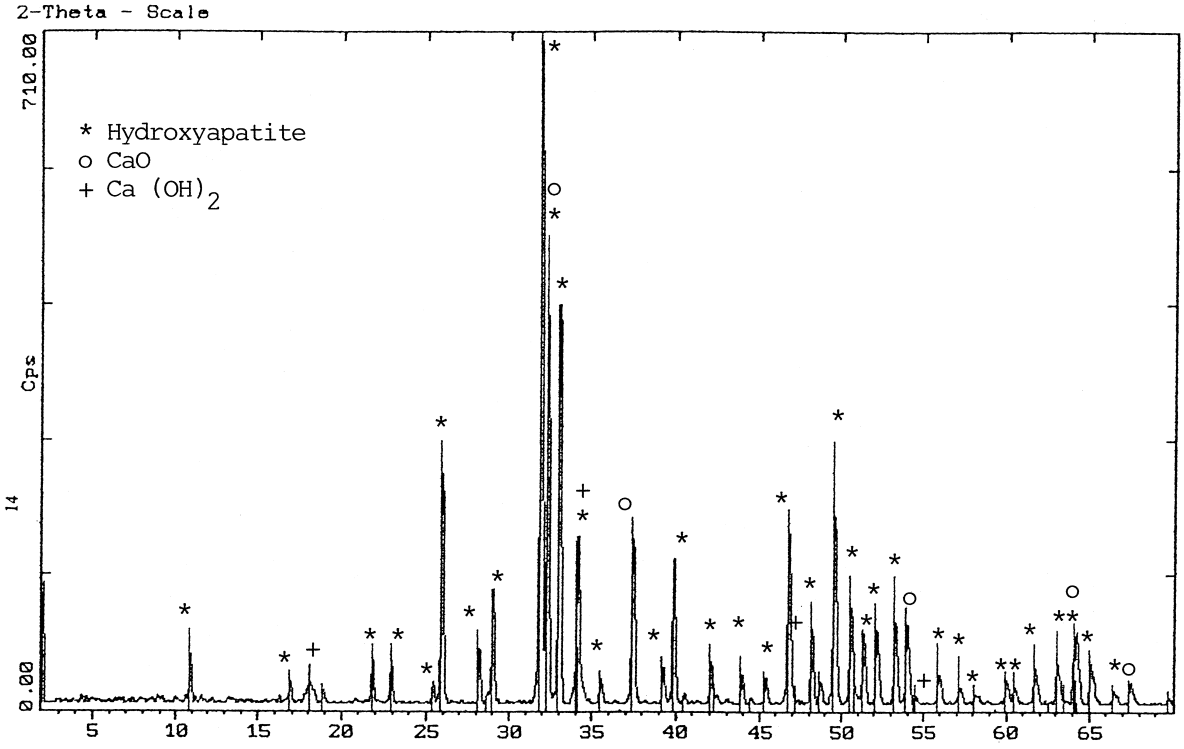

Fig. 3. The X-ray diffractogram for the same sample as in Fig. 2 but after the thermal treatment at 1050°C in the phosphate solution.

crystalline phases are identified are HAp (JCPD, 9-0432), calcium oxide (JCPD, 37-1497) and calcium hydroxide (JCPD, 4-0733). These results can be explained as an incomplete transformation of calcite, due to a temperature not high enough or else to the annealing time which is too short.

Prior to mixing the reactants, the X-ray diffraction analysis of $Ca_3(PO_4)_2$ showed traces of $Ca_{10}(PO_4)_6(OH)_2$ and monetite ($CaHPO_4$). The existence of these species as impurities in the initial reactant seems, however, to have little if any influence on the final material. HAp is by large the main crystalline phase present. Moreover, HAp is the unique apatite phase detected as a product of the process. Fourier transformed infrared (FTIR) spectra of the final material show the presence of bands at 605 cm^{-1} and 1050 cm^{-1} which correspond to $(PO)_4$ functional groups in HAp and tricalcium phosphate; these results will be discussed in a later more detailed publication.

The thermal processing used for eliminating the organic component of eggshell at 900°C produces the conversion of calcite into calcium oxide. The phases detected after heating to 1050°C in moist atmosphere can be explained as follows. The reaction theoretically expected for calcite and the phosphate solution is:

$$3Ca_3(PO_4)_2 + CaCO_3 + H_2O \rightarrow Ca_{10}(PO_4)_6(OH)_2 + CO_2 \quad (3)$$

However, when calcium carbonate is mixed with water, there is a hydration that produces calcium hydroxide: $CaO + H_2O \rightarrow Ca(OH)_2$. Therefore, this phase is present also as an initial reactant, as well as during the reaction. On this basis, we infer that the actual reaction is:

$$3Ca_3(PO_4)_2 + Ca(OH)_2 \rightarrow Ca_{10}(PO_4)_6(OH)_2 \quad (4)$$

The existence of monetite as impurity in the tricalcium phosphate initial reactant can be explained by the occurrence of the following reactions:

$$6CaHPO_4 + 4CaO \rightarrow Ca_{10}(PO_4)_6(OH)_2 + 2H_2O$$

$$6CaHPO_4 + 4Ca(OH)_2 \rightarrow Ca_{10}(PO_4)_6(OH)_2 + 6H_2O \quad (5)$$

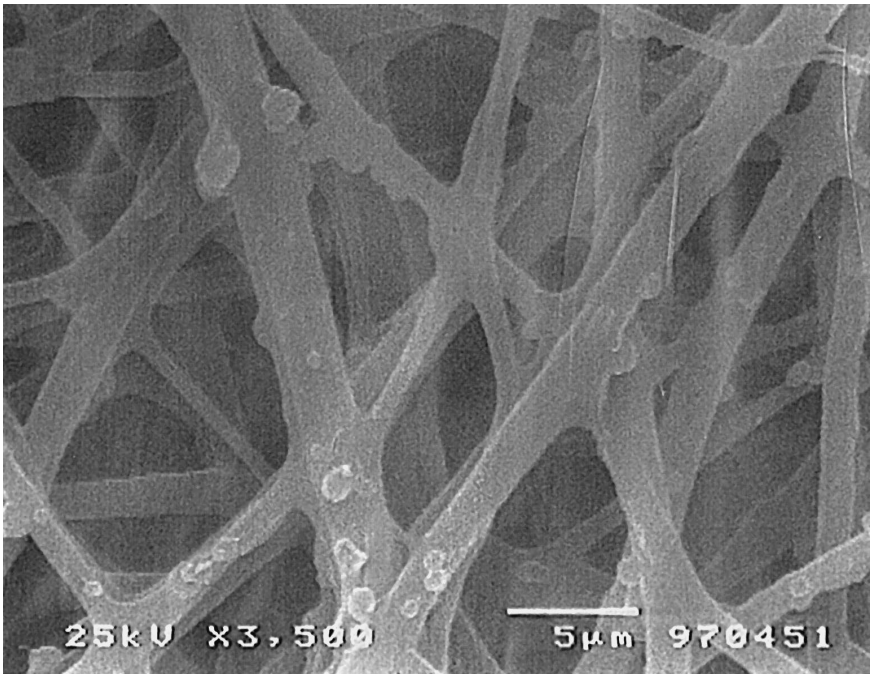

Fig. 4. SEM micrograph of the inner part of a white eggshell.

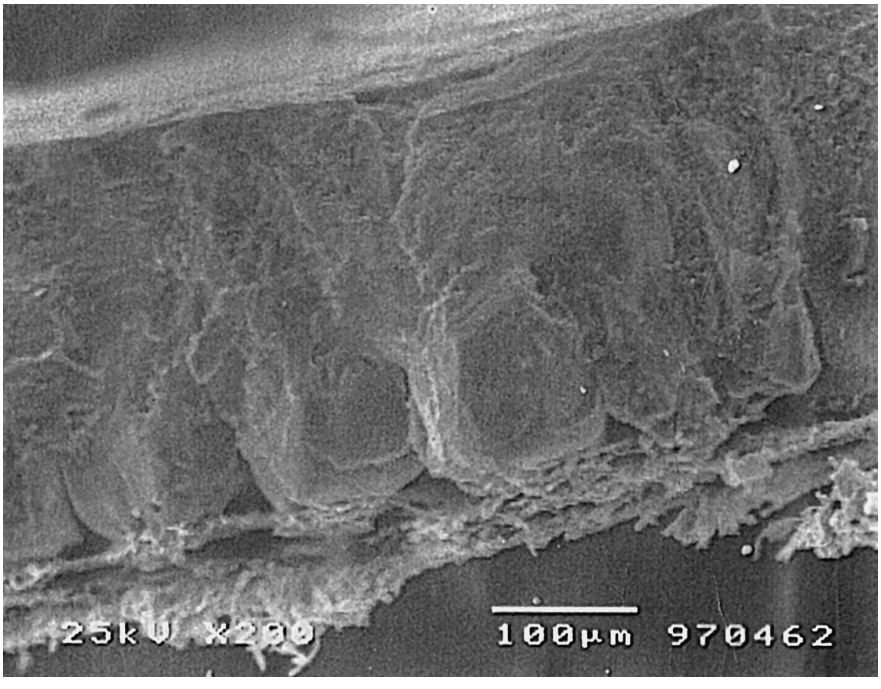

Fig. 5. SEM micrograph of the sample as in Fig. 4 but thermally treated at 450°C.

Fig. 6. SEM micrograph of the sample as in Fig. 5 but thermally treated in air at 900°C.

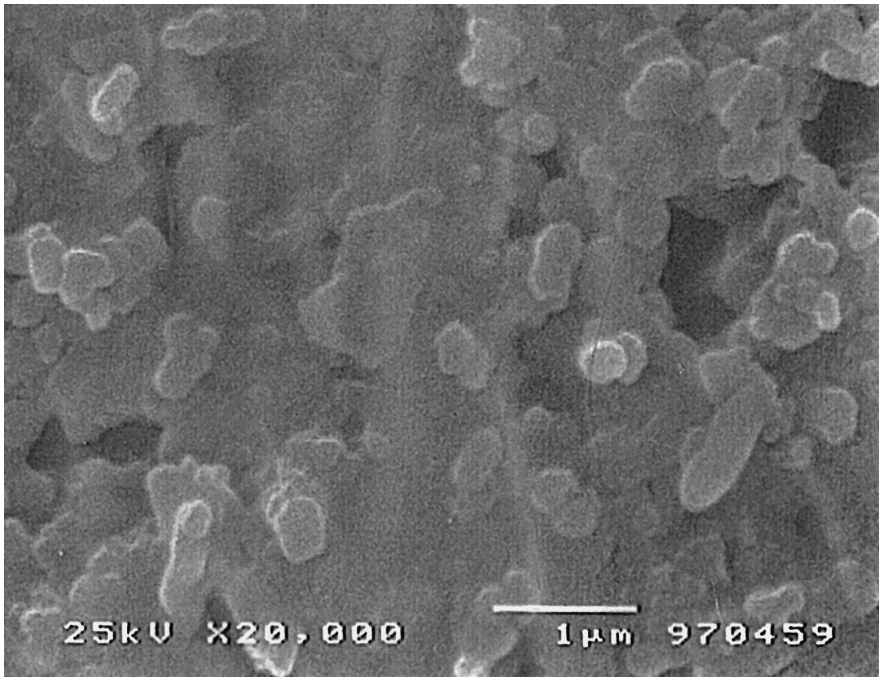

Fig. 7. SEM micrograph of the sample as in Fig. 6 but thermally treated at 1050°C in the phosphate solution.

Morphologies of the materials at different stages of the thermal treatment are shown in Figs. 4–7. In Fig. 4, we observe a fiber-like morphology corresponding to the inner part of a white eggshell; a uniform pore size is visible. In Fig. 5, the SEM of the sample treated at 450°C (which corresponds to calcium oxide) is shown; it is important to note that the fiber-like morphology is maintained; apparently the thermal treatment does *not* affect it in any significant way. The SEM micrograph of the sample treated at 900°C is shown in Fig. 6, while for the sample treated at 1050°C in the phosphate solution in Fig. 7.

4. Concluding remarks

Our procedure of synthesis of HAp starting from eggshells in a phosphate solution at an elevated temperature represents a novel way for producing a useful biomedical material. The concentration of HAp obtained by this method can be improved further by optimizing the composition of the phosphate solution, the time and the temperature of annealing. Needless to say, the amounts of the phosphate solution used at any location and time are not a cause for any environmental concerns. The amounts mentioned in Section 1 pertain to a whole year and to a country with a population of the order of 100 million people. The use analytical grade reactants which minimize the existence of species that alter the expected reactions may also improve the phosphate procedure. It is necessary, however, to perform a mechanical characterization of this material and to relate it to microstructural aspects; this work is currently in progress.

Acknowledgements

One of us (W.B.) acknowledges discussions with Prof. Rustum Roy of the Materials Research Laboratory, the Pennsylvania State University, University Park. Comments on the manuscript of this paper have been kindly provided by several colleagues, in particular by Dr. Anatoly Y. Goldman, Alcoa CSI, Crawfordsville, IN. An equipment donation from Perkin-Elmer, Norwalk, CT, is gratefully acknowledged.

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