จุฬาลงกรณ์มหาวิทยาลัย
ทุนวิจัย
กองทุนรัชดาภิเษกสมโภช

รายงานวิจัย

การเตรียมชิ้นงานไบโอเซรามิกเพื่อใช้ทางการแพทย์

โดย

สุพัตรา จินาวัฒน์
ดุจฤทัย พงษ์เก่า คะซิมา
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ธันวาคม 2549

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กองทุนรัชดาภิเษกสมโภช

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เดือน ธันวาคม พ.ศ. 2549

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Report on

Preparation of Bioceramic Specimens for Medical Application

By

Associate Professor Dr. Supatra jinawath
Dr. Dujreutai Pongkao Kachima
Dr. Pornapa Sujaridworakun

December 2006

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ชื่อโครงการวิจัย **การเตรียมชิ้นงานไบโอเซรามิกเพื่อใช้ทางการแพทย์**

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เดือนและปีที่ทำวิจัยสำเร็จ ธันวาคม พ.ศ. 2549

บทคัดย่อ

ไฮดรอกซีแอพาไทต์(HA) และ วัสดุเชิงประกอบไตรแคลเซียมฟอสเฟต/ไฮดรอกซีแอพาไทต์ (TCP/HA) ถูกสังเคราะห์ขึ้นในห้องปฏิบัติการจากเถ้ากระดูกวัว/ควาย หรือวัสดุเหลือทิ้งจากอุตสาหกรรมที่เกี่ยวข้อง ทำการขึ้นรูปเป็นชิ้นทดลองชนิดพรุน มีรูปร่างเป็นแท่งสี่เหลี่ยม และเป็นเม็ดแบน (ความพรุน30-34 % ความแข็งแรงดัด17 MPa ขนาดรูพรุน <5-30μ) เพื่อนำมาทำการทดสอบทางการแพทย์ โดยปลูกฝังในสุนัขทดลอง และต่อมาในผู้ป่วยอาสาสมัคร ผลจากการปลูกฝังในสุนัขเป็นเวลา 3 ถึง 6 เดือน ประสบความสำเร็จ และผลในผู้ป่วยก็ประสบความสำเร็จเป็นที่พอใจของผู้ที่เกี่ยวข้อง ผลดีที่ได้นี้เป็นการพิสูจน์ว่าชิ้นทดลองไฮดรอกซีแอพาไทต์ชนิดพรุนนี้ มีสมบัติที่เหมาะสมในการที่จะเหนี่ยวนำให้เส้นเลือดและเนื้อเยื่อกระดูกเติบโตและเชื่อมต่อผ่านเข้าไปในรูพรุนได้ดี นอกจากนี้ยังได้ทำการทดลองเพื่อหาสภาวะที่เหมาะสม ในการเคลือบชั้นของไฮดรอกซีแอพาไทต์บนผิวโลหะไทเทเนียม เพื่อการใช้งานที่กว้างมากยิ่งขึ้น

Project Title **Preparation of Bioceramic Specimens for Medical Application**

Name of the investigators Associate Professor Dr. Supatra Jinawath
Dr. Dujreutai Pongkao Kachima
Dr. Pornapa Sujaridworakun

Year December 2006

ABSTRACT

HA (hydroxyapatite, $Ca_{10}(PO_4)_6(OH)_2$, Ca/P = 1.67) and composite TCP/HA (Tricalcium phosphate, $Ca_3(PO_4)_2$, Ca/P = 1.5) were synthesized in our laboratory from bovine bone ash or the waste from the manufacture of products derived from it and fabricated into porous bar and disc shaped specimens (30-34 v% porosity, flexural strength of 17 MPa, pore diameters <5-30μ) for *in vivo* histological study and clinical trial, repectively. The histological results of the 3- and 6-month *in vivo* tests in canines were positive and successful. The clinical trials using titanium D-cages with HA inserts after 3-6 months were also proved positive, and with a satisfactory result. This also proved our belief that the practical limit of the smallest pore size for blood vessel or bone tissue ingrowth could be smaller than 100μ (macropore) in the case of resorbable HA ceramic with interconnected micro-to-mesopores (estimated as <1, 5-30 μ). To explore into a broader application, a coating of biological HA layer on Ti alloy implant was also studied and the results were discussed.

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Preparation of Bioceramic Specimens for Medical Application

1. Background of the research proposal

Bioceramics have long been regarded as a potential biomaterial and become increasingly employed in various medical applications. Among the well known bioceramics, calcium phosphates, i.e. hydroxyapatite (HA) and tricalcium phosphates (TCP) have been highly recommended for bone tissue induction, and hence widely applied in orthopedic surgery. There are quite a number of hydroxyapatite products, commercially available in various forms, i.e. granules, dense or porous pieces, injectable suspension, and as a coating layer on shaped metal inserts. In this project, it is required by the surgeons, our collaborators[1], that the hydroxyapatite should be in (1) the form of porous disc about 1.0 cm in diameter and (2) as a coating layer on Ti alloy.

Previously our research group had successfully synthesized a range of calcium phosphates (Ca/P molar ratios 0.5-2) from chemicals, bovine bone and by-product of bone-gelatin manufacture, and tried several fabrication processes using our synthesized materials[2-10]. The HA produced in our laboratory was in the forms of powder, granule and porous bars. They came out with satisfaction in terms of morphology, chemical purity and mechanical performance. However, in clinical application the shape and size of the HA ceramic to be implanted have to conform with the task of the surgeon, therefore this project is regarded as the extension of the previous one. We highly expect that this collaboration will reveal us more information on the histology of our products and lead to the possibility of independence on import, as well as to further development in the field of biomaterials. Nowadays, we all realized that without the collaboration from researchers in the related fields, most of the research outcomes can never be further developed. The collaboration can open the closed end of the channel.

2. Introduction

Calcium phosphate powders with Ca/P molar ratios of 0.5 (MCPM, $Ca(H_2PO_4)_2.H_2O$) to 2.0 (TTCP, $Ca_4P_2O_9$) were successfully synthesized in our laboratory from bovine bone ash or the waste from the manufacture of products derived from it. The most popular calcium phosphates are HA (hydroxyapatite, $Ca_{10}(PO_4)_6(OH)_2$, Ca/P = 1.67) and β-TCP (β-tricalcium phosphate, $Ca_3(PO_4)_2$, Ca/P = 1.5). Typically, the applications of porous calcium phosphates are as bone graft, augmentation, infilling and preforms to be shaped during surgery, and practically, the powders have to be fabricated into a compact of designed shape and porosity i.e. a granule, a porous bar of appreciable strength or as a composite with polymer matrix. During an appropriate retention time, an ideal porous calcium phosphate implant should be resorbable, hence conduct bone tissues at its expense. Consequently, the practical limit of the smallest pore size for blood vessel or bone tissue ingrowth can be smaller than 100 μ (macropore). This point is vital for the application of porous calcium phosphates with micro-to mesoporosity since an appreciable strength can be achieved with this range of porosity. Accordingly, their pre-forms can be shaped and retained in place long enough to conduct bone tissues effectively. To verify this belief, porous HA bars (30-34 v% porosity, flexural strength of 17 MPa, pore diameters <5-30μ) were implanted in canines' tibias for the periods of 3-6 months for histological study, and later clinical trials were performed using a D-cage

filled with a porous HA disc on voluntary patients whose CT scan and X-ray results would be followed up after a period of 6 months.

Objective

The objective is to prepare HA ceramics in the forms of porous bar and disc, and as a coating on metal implant to support a team of Chulalongkorn Hospital orthopedic surgeons with the expectation that the information on histology systematically studied by our medical collaborators, will clinically justify the properties of our products, and lead to further development.

Duration

1 year, from May 2005- March 2006.

Scope

For producing porous HA bar and disc, the procedure reported in reference 2, 3 and 8 will be followed. The process for producing the coating will be by electroless plating. A histological study using porous HA bars will be performed on canines for the periods of 3 and 6 m. Later a clinical trial on a number of voluntary patients using porous discs will be performed and followed up by X-ray radiography after 3 and 6 m implantations.

For simplicity, the report will be split into 2 parts:

Part A: ***Experiment on the application of porous HA and TCP/HA bars and disc specimens.***

Part B: ***Experiment on the application of the coating of calcium phosphates on Ti alloy.***

Part A: Experiment on the application of porous HA and TCP/HA bars and disc specimens.

1. Materials and method

Materials

Dicalcium phosphate, dicalcium phosphate anhydrous(DCPD/DCPA) synthesized from the by-product of bone-gelatin industry, calcium phosphate glass, $Ca(PO_3)_2$, and chemical grade $CaCO_3$ powder were used as the starting materials for the fabrication of porous HA and TCP/HA specimens. The experimental procedure is presented as a flow chart in Fig. 1.

Methods

1. ***Sample preparation:*** The experimental procedure is presented as a flow chart in Fig. 1A.

2. ***In vivo experiment and clinical trial:*** The porous HA bars were implanted in canines' tibias for the periods of 3 and 6 months for histological study, and later clinical trials were performed using D-cages (Ti alloy) filled with porous HA discs on a number of voluntary patients whose CT scan and X-ray results were to be followed up after the periods of 3 and 6 months.

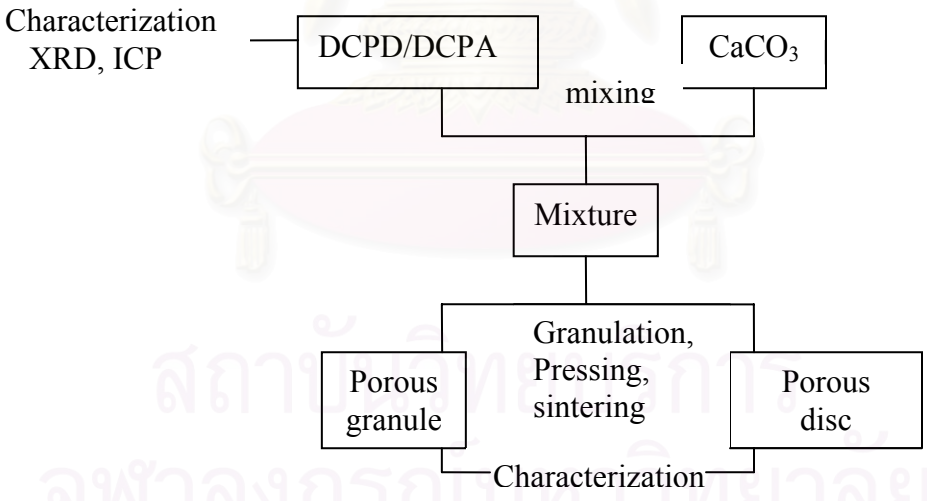


Fig.1A Flow chart of the porous HA, TCP/HA specimens forming process

2. Results and discussion

1. Starting materials

Chemical compositions (ICP) and particle sizes (Shimadzu SA-CP2) of the synthesized DCPD and DCPA are shown in Table 1.

Table 1 Properties of the starting materials

	DCPD	DCPA	$CaCO_3$
MgO (wt%)	0.02	0.04	Purity >99% Fluka
MnO (wt%)	<0.01	<0.01	
Fe2O3(wt%)	0.14	0.14	
Zn (ppm)	73	52	
Cu (ppm)	20	24	
Ca/P molar ratio	1.13	1.21	
*Median particle size (μm)	13	7	24

The chemical compositions confirm the high purity and nonstoichiometry of the starting materials. In addition, the concentrations of heavy metals (ppm) are as follows: Cd = 0.5, Pb<5, Hg<1, As<0.5, Ni = 2. The contents of all these heavy metals well pass the specification of American Standard Test Methods for Composition of Ceramic Hydroxyapatite for Surgical Implants (ASTM: F118-88).

2. Porous HA and TCP/HA specimens

The phase analysis by XRD (Philips: PW1730/10) and by FT-IR (Perkin Elmer 1760X) of the sintered specimens (bars) is illustrated in Fig. 2A and 3A, respectively.

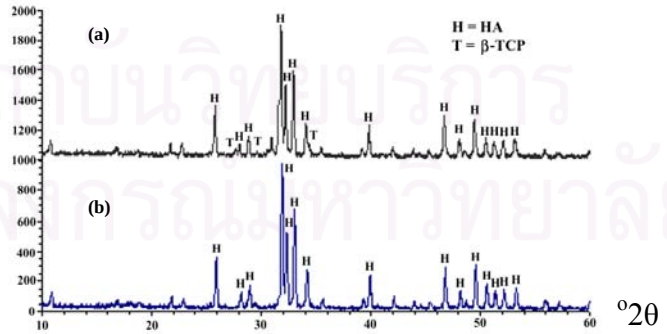


Fig. 2A XRD of the sintered specimens (bars), at 1200°C, 2h

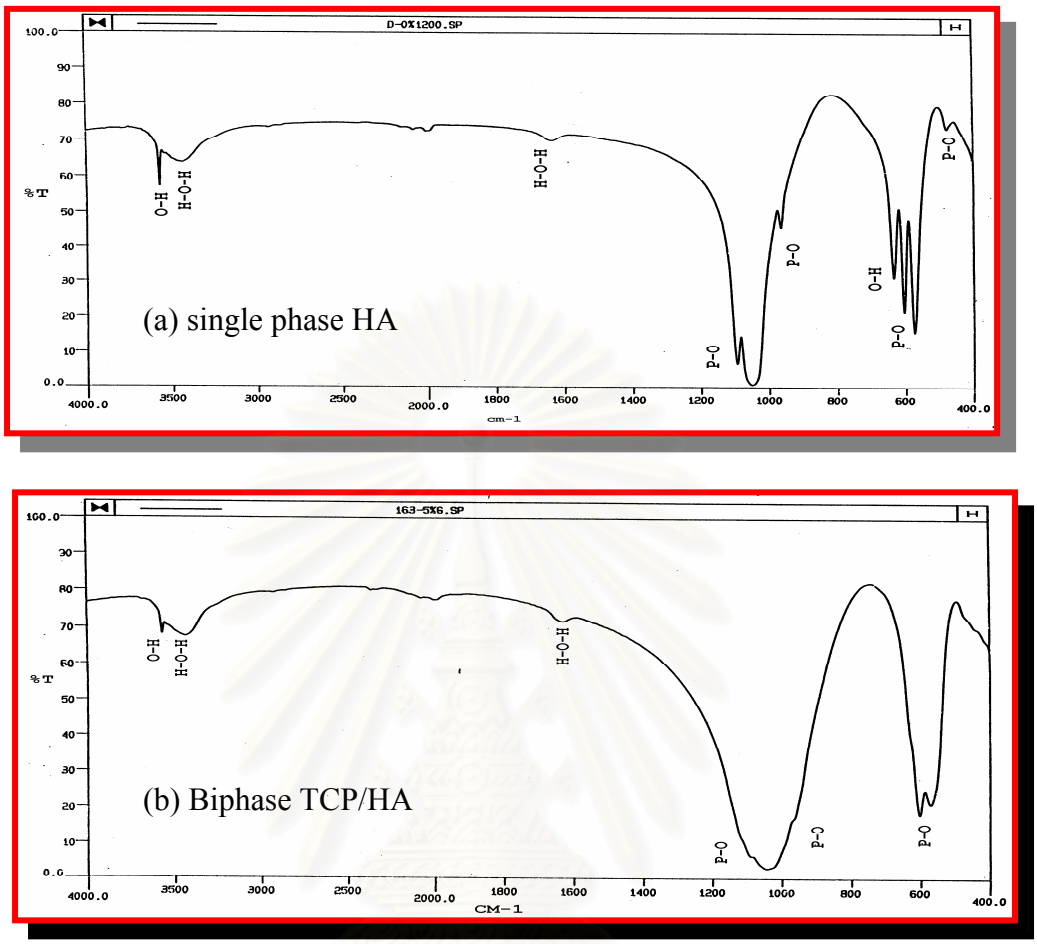


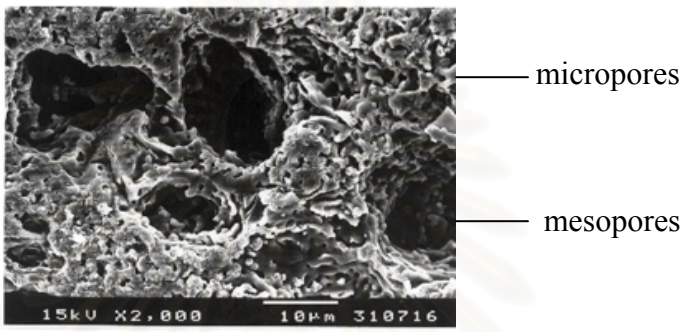
Fig. 3A FT-IR spectrograms of the sintered specimens (bars), at 1200 °C, 2h

Table 2 Properties of the sintered specimens (bars)

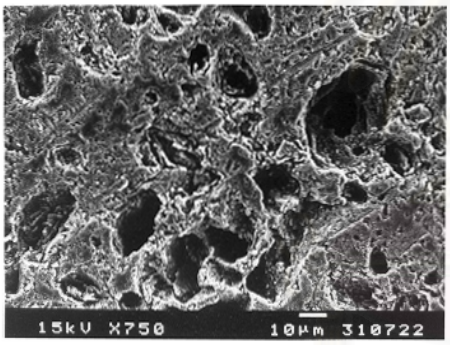
Temp 1200 °C, 2h 3 °C/min	DCPD starting material		DCPA starting material	
	0.5+wt% glass 50MPa	84MPa	0.5+wt% glass 50MPa	8 4MPa
Bulk density, g/cm^3	1.72	1.67	1.87	1.86
Apparent porosity, vol%	46.6	45.15	36.17	37.35
Flexural strength, MPa	8.05±0.51	9.78±0.20	15.84±0.82	17.51±1.77
Phase	HA+β-TCP	HA	HA+β-TCP	HA

The summary of all the analytical data of the sintered specimens is tabulated in Table 2. It is found that small amount of calcium phosphate glass enhances the formation of β-TCP, and single phase HA could be obtained at the sintering temperature of 1200°C.

The SEM micrographs showing the morphology of the porous specimens (bars) are presented in Fig. 4A. A range of mesopores (~5-30 μ) and that of micropores (<1 μ) are visible and all types of pores are clearly interconnected. This type of morphology is highly resorbable and should be ideal for bone tissue ingrowth.



(a) Mesopores and micropores



(a) Mesopores and micropores (low magnification)

(b) Solid part enlarged, showing micropores

Fig. 4A SEM micrographs (JEOL: JDX-8030, CuKα, λ =1.5405A) of the fractured surface of the HA sample (bars)

3. Results of in vivo test and clinical trial

Fig. 5A is the SEM micrograph of the cross section of the HA bar implanted in a canine's tibia for 6 months. The micrograph reveals a very high compatibility between the implant and the surrounding bone tissue confirmed by the absence of noticeable gaps at the interface. The visible white spots on the surface indicate resorption of the implant over time.

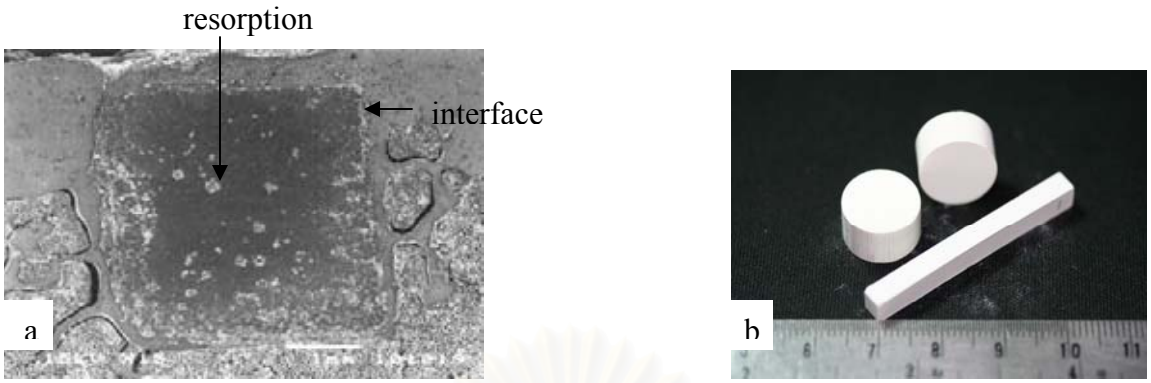
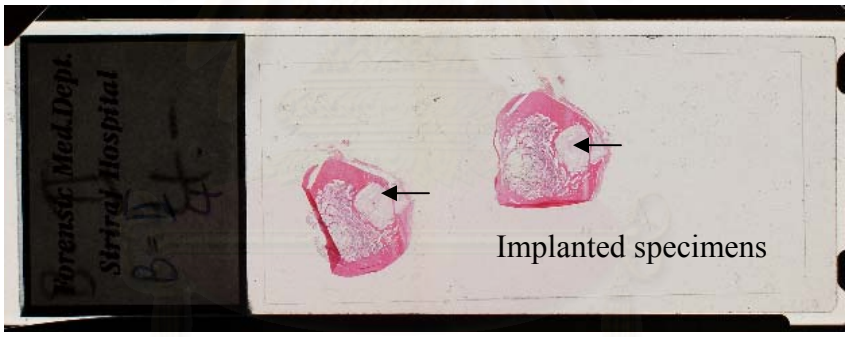


Fig. 5A SEM micrograph of the cross section of the HA bar implanted in canine's tibia for 6 m (a), and digital photograph of sintered porous HA bar and discs (b)

Fig. 6A is the optical micrographs of the thin sections (dyed in toluidine) of the above implant under histological study, in collaboration with the team of orthopedic surgeons of Dr. Piboon Ithiraviwong, Chulalongkorn Hospital and the team of pathologists of Dr. Somboon Thamatakerngkij, Siriraj Hospital, showing old bone tissue in deep red color and new bone tissue (pink) growing into the implant, with an increasing amount over time.



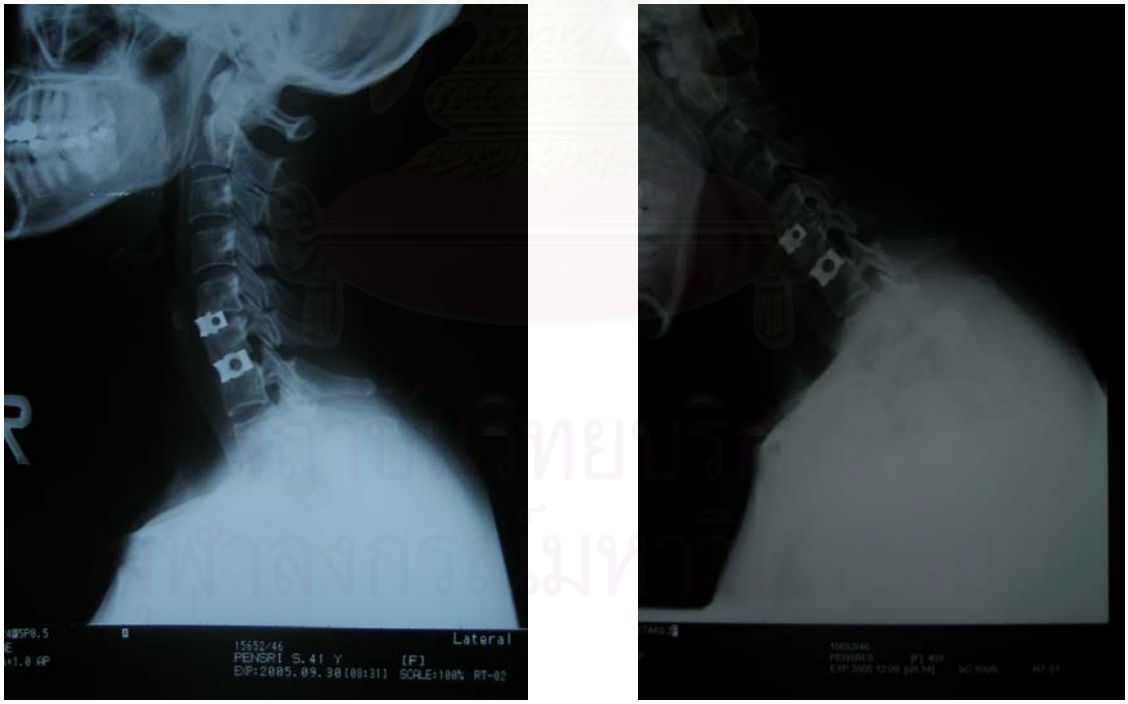
Optical micrographs of implants in canine's tibia, 3 and 6 m

Fig. 6A Optical micrographs of the thin sections

Fig. 7A is the digital photograph of the D-cage devices filled with porous HA discs to be inserted between the cervical bones in patient's neck. The surgery is known as '***anterior cervical disectomy and fusion*** (***ACDF***)'.

Fig. 7A Photograph of the D-cage devices

A clinical trial in collaboration with the team of orthopedic surgeons of Dr. Prakit Tienboon, Chulalongkorn Hospital, had been performed on about 50 cases of voluntary patients so far, and the results after 3 and 6-month implantation are shown in Fig 8A and 9A, respectively.



(a) (b) bending action

Fig. 8A X-ray radiograms (a) after 3-m and (b) after 6-m implantations

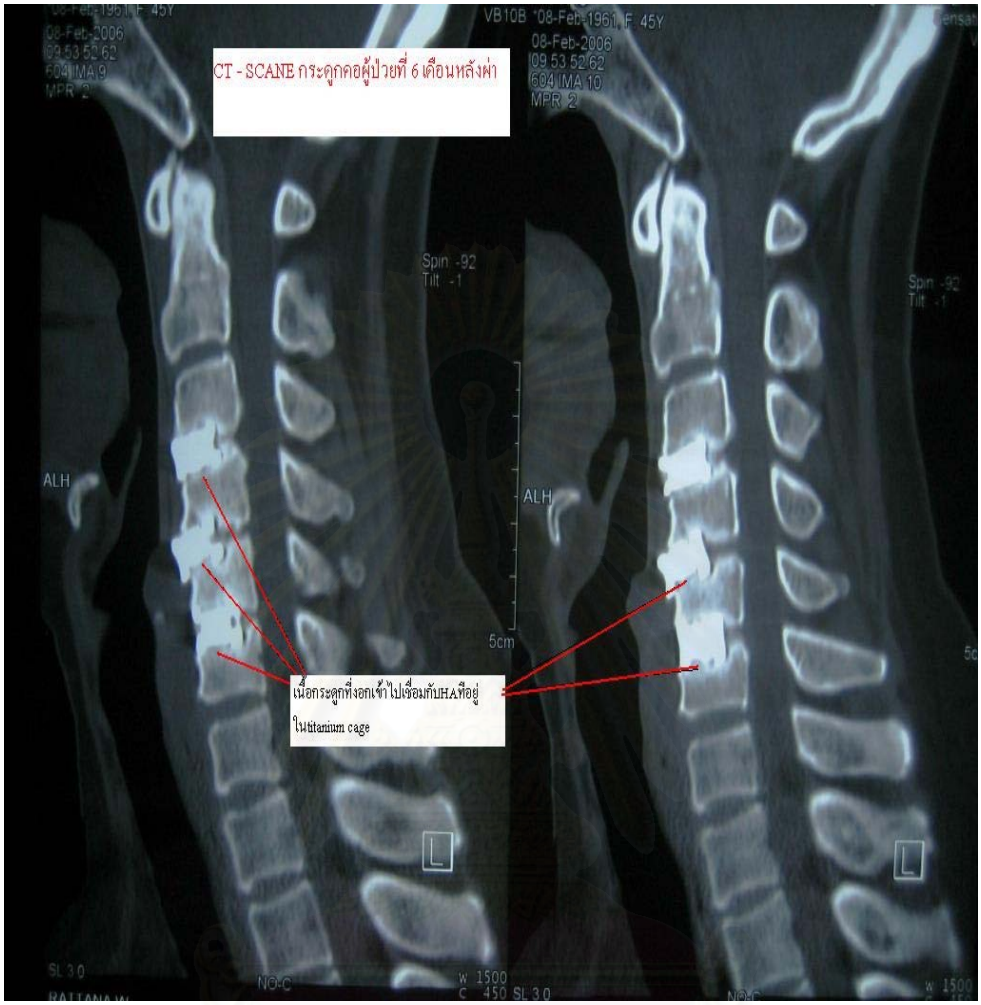


Fig. 9A CT scan after 6-m implantation

The results from X-ray radiography, X-ray radiograms and CT scan (CAT scan) after 3- and 6-m implantation (Fig. 8A and 9A) reveal that the titanium devices have been tightly fixed in place by the aid of the new bone tissues conducted by the porous ceramics. There is no shadow area visible at the interface between the device and the adjacent cortical bone and no any inflammation detected. All the patients heal rapidly and can turn their necks to an angle of almost 180^{o}, moreover the operation can be done in 1 day.

In this application, the porous HA was used as a resorbable bone graft material. It has been well known that there are 4 techniques for bone graft.

1. *Autograft:* using the patient's own bone. This is limited by the size of the wound and long healing time.

2. *Allograft:* using human bone from bone bank. There is a drawback from genetic diseases.

3. *Xenograft:* using bones from other species. The problems encountered are genetic diseases and compatibility.

4. *Synthetic materials:* Biomaterials such as bioceramics, biopolymers and composites of the two, and novel metals come into this one. In the exception of the inert metals, the drawbacks from this group of materials are low compatibility and resorption when compared to the above choices. Nevertheless, this discrepancy has been improved tremendously over the past decade by numerous researchers. Many new materials (monolith and composite) having directed microstructure and function have been introduced. Old concepts of biomaterials have been refined and many new ones proposed.

3. Conclusion

The histological results of the 3- and 6-month *in vivo* tests in canines are positive and successful. The clinical trials using titanium D-cages with about 100 pieces of the porous HA specimens on a number of voluntary patients have also been performed and the results of 3- and 6-month implantations in 13 cases have been followed up as scheduled. All the D-cages are firmly fixed so there is no dislodging under neck movement. Hence all the 13 cases prove positive, and with satisfactory results. This also proves our belief that the practical limit of the smallest pore size for blood vessel or bone tissue ingrowth can be smaller than 100 μ (macropore) in the case of resorbable HA ceramic with interconnected micro-to-mesopores (estimated as <1, 5-30 μ).

Part B: Experiment on the application of the coating of calcium phosphates on Ti alloy.

1. Materials and method

Materials

Chemical grade dicalcium phosphate, DCPD, Fluka.
Titanium screws (courtesy of Dr. Prakit Tienboon)

Methods (electroless plating)

1. *Surface treatment and incubation:*

The experimental procedures for a surface treatment of Ti substrates were conducted in either H_2O_2 or NaOH solutions prior to calcium phosphate coating by ***biomimetic*** process, i.e. **soaking in R-SBF solution**[11] which is regarded as an ***electroless plating*** process. The procedures are presented as flow charts in Fig. 1B-3B.

1.1 Treatment by soaking in H_2O_2 solution and followed by incubating in R-SBF solution

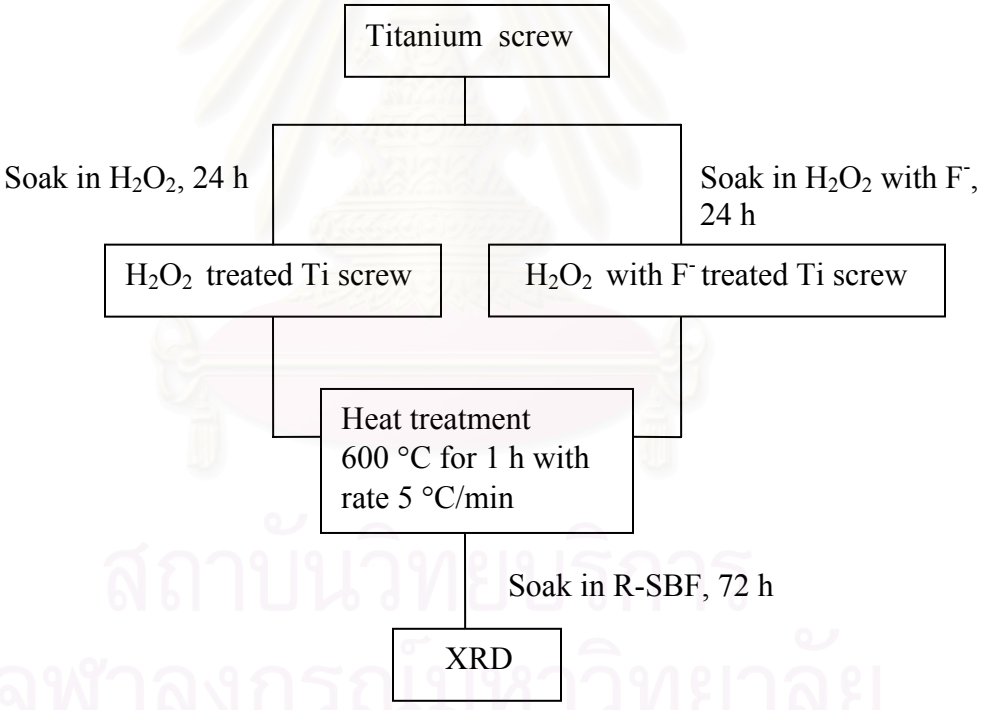


Fig.1B Flow chart for the surface treatment of titanium screw, soaking in H_2O_2 solution and incubating in R-SBF solution

1.2 Treatment by autoclaving in NaOH solution and followed by incubating in R-SBF solution

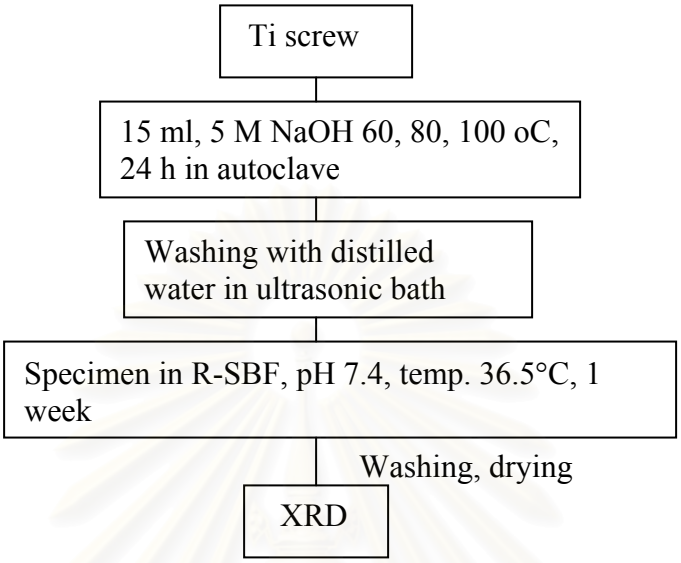


Fig. 2B Hydrothermal treatment of Ti screw in NaOH and incubating in R-SBF solution

1.3 Treatment by soaking / autoclaving in H_2O_2 solution and followed by incubating in R-SBF solution

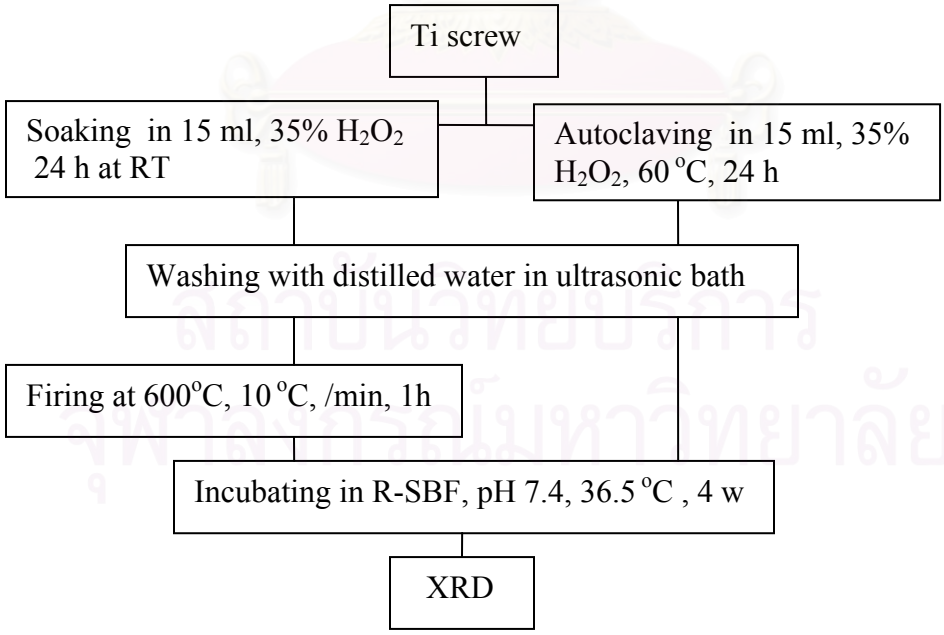


Fig. 3B H_2O_2 treatment of Ti screw surface and incubating in R-SBF solution

2. *Results and discussion*

Improving the bioactivity of titanium screw by electroless plating

Titanium screw used in orthopedic application is so called as “pedicular screw”. It is treated in either H_2O_2 solution or H_2O_2 solution with fluoride ion (from NaF) addition. Pedicular screw is a complex shape screw made from an alloy of titanium metal used in the fixing of human spinal bone as shown in Fig. 3B.

Fig.4B Titanium pedicular screw for orthopedic application

2.1 Treatment by soaking in H_2O_2 solution and followed by incubating in R-SBF solution (**preliminary experiment**)

Generally, titanium is classified as a bio-inert metal which is not able to induce bone formation. Hence an appropriate surface treatment technique is employed to modify its surface property. However, due to its complex shape, the surface treatment in solution should be a right way to bring about the expected outcome, i.e. surface treatment in a strong oxidizing solution of H_2O_2 would allow the development of a homogeneous titanium dioxide (TiO_2) layer on the surface of titanium metal. This surface treatment was reported[12] to be a means to improve the bioactivity of titanium since the surface TiO_2 in body fluid (R-SBF solution) would act as nucleation site for HA.

The result from our preliminary experiment showed that H_2O_2 treated titanium could induce the apatite formation after incubation in R-SBF (Revised-Simulated Body Fluid) for 72 hours as shown in the XRD patterns of the screw surface Fig. 5B.

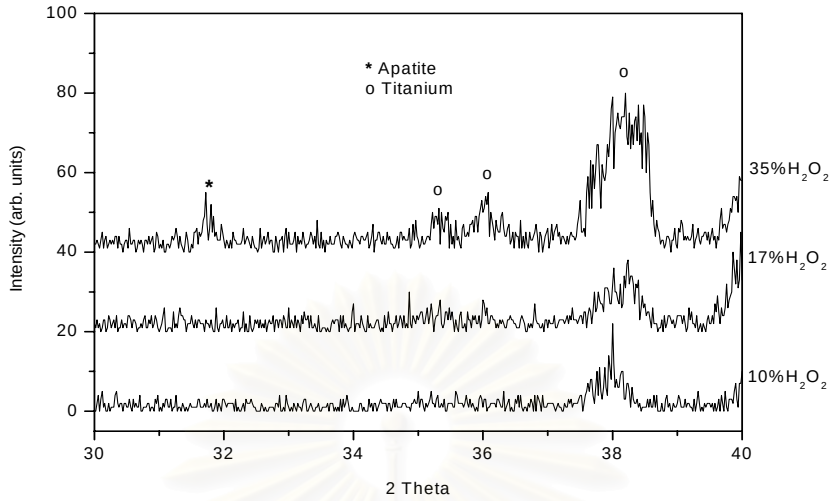


Fig. 5B XRD patterns of the H_2O_2 treated pedicular screws at various concentrations of H_2O_2 (10%, 17% and 35%) after incubation in R-SBF for 72 h

From Fig. 5, it can be observed that apatite (HA) could form on the surface of 35% H_2O_2 treated titanium screw after incubation in R-SBF for 72 hours. However, we can neither observe the peak of apatite on 17% and 10% H_2O_2-treated Ti screws nor on the ones treated with F^- added H_2O_2. Therefore fluoride ion did not play any role to induce the apatite formation on titanium screw.

From the literature[12], hydrothermal treatment has been reported as another means to enhance the surface treatment of substrate so the content of apatite on the Ti surface is expected to increase by hydrothermal treatment in an autoclave.

2.2 Treatment by autoclaving in NaOH solution and followed by incubating in R-SBF solution

This method is aimed to improve the bioactivity of titanium screw by hydrothermal process in homogeneous solution

Results

Physical appearance: As-received screw, shiny.
After soaking in NaOH, dull.
After incubating in R-SBF solution, dull. The photographs of the specimens are shown in Fig. 6B.

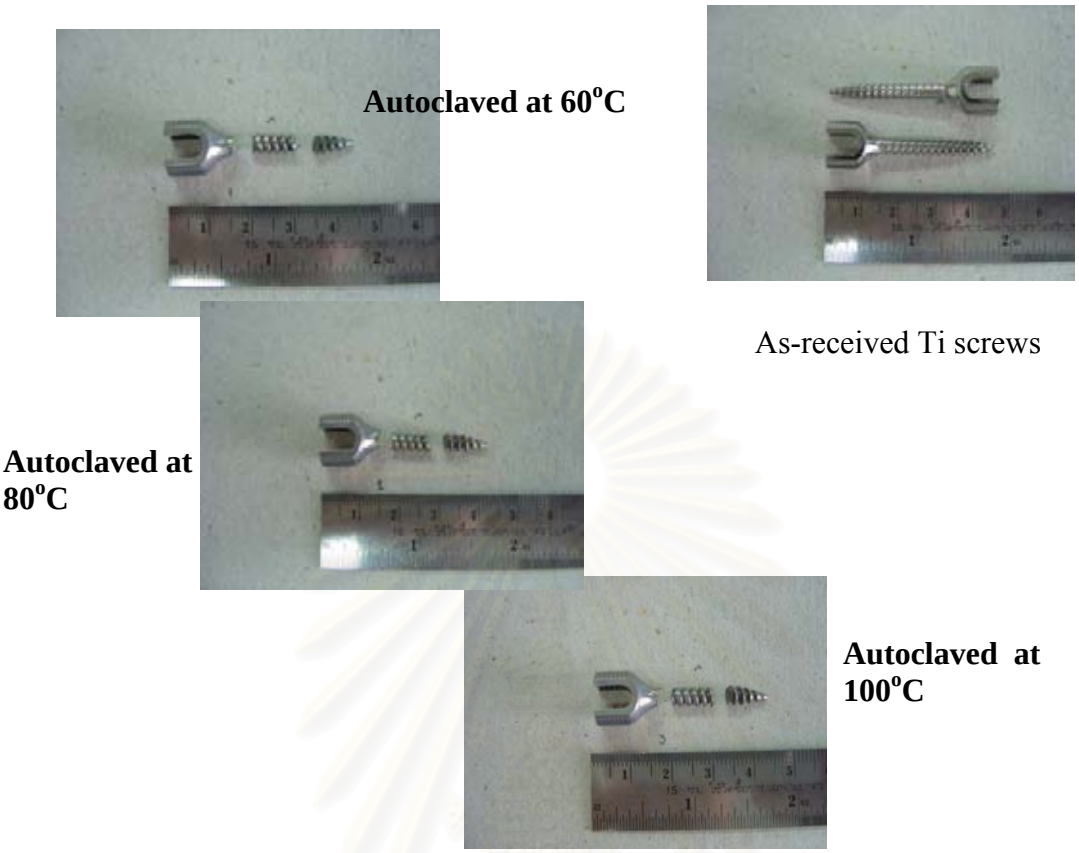


Fig. 6B. Photographs of the specimens after autoclaving in NaOH solution

Phase analysis by XRD of the surface of the specimens(cut into 3 pieces) after autoclaving in NaOH and incubating in R-SBF solution are shown in Fig. 7B.

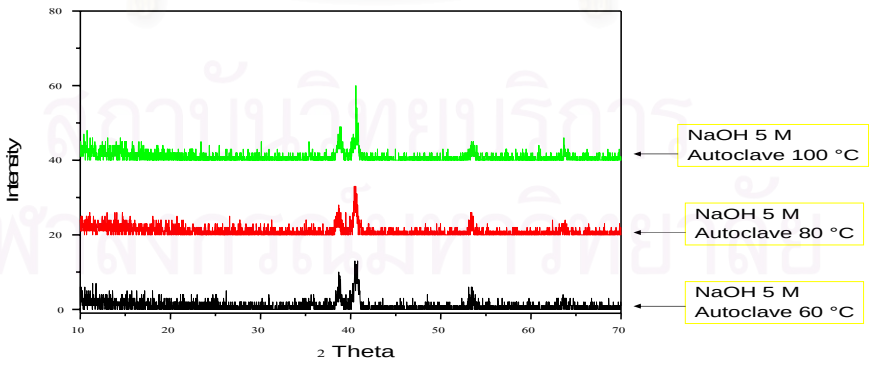


Fig. 7B XRDs of the specimens, autoclaved in NaOH and incubated in R-SBF solution

No HA phase ($2\theta \sim 32^{o}$) was detected in Fig. 5B. Therefore, the next experiment was performed by further treatment of Ti screw surface by either soaking or autoclaving in 35 % H_2O_2 solution, and is schematically presented in Fig. 3B.

2.3 Treatment by soaking / autoclaving in H_2O_2 solution and followed by incubating in R-SBF solution

Results (soaking in H_2O_2)

Physical appearance: As-received screw, shiny.
After soaking in H_2O_2, rusty brown.
After firing , dark blue.
After incubating in R-SBF solution, dark blue.

The photographs of the specimens after soaking in H_2O_2 and incubating in R-SBF solution are shown in Fig. 8B.

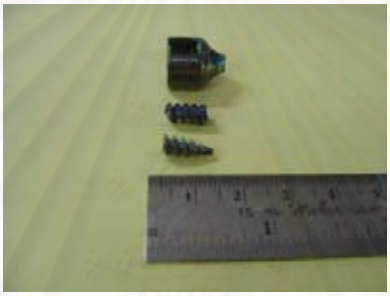


Fig. 8B Photographs of the specimens after soaking in H_2O_2 and incubating in R-SBF solution

Results (autoclaving in H_2O_2)

Physical appearance: As-received screw, shiny.
After autoclaving in H_2O_2, black.
After incubating in R-SBF solution, black.

The photographs of the specimens after autoclaving in H_2O_2 and incubating in R-SBF solution are shown in Fig. 9B.

Fig. 9B Photograph of the specimens after autoclaving in H_2O_2 and incubating in R-SBF solution

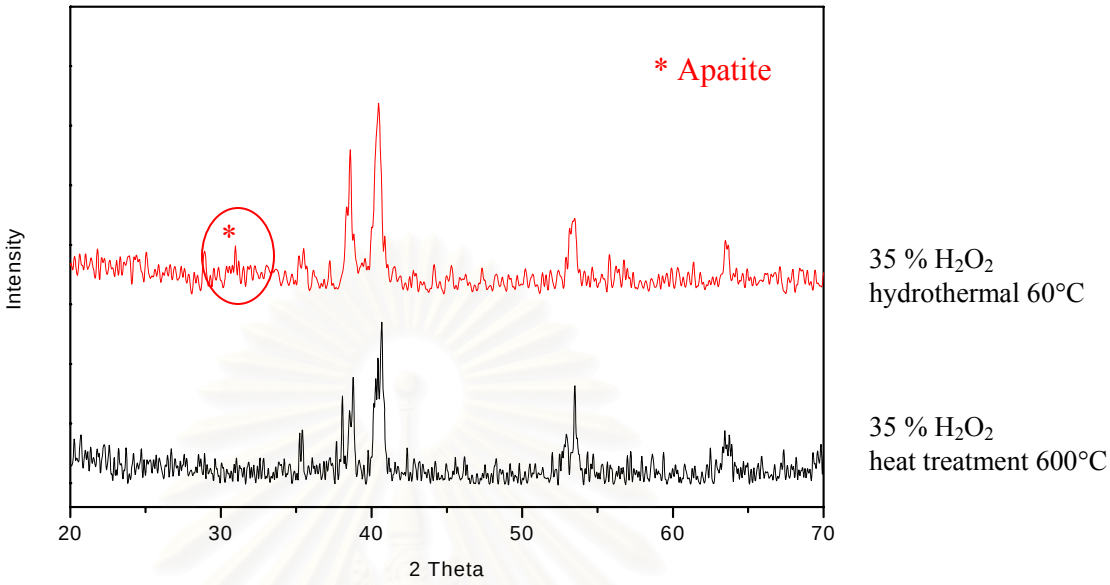


Fig. 10B XRDs of specimens after treating with H_2O_2 and incubating in R-SBF solution

It was found that there was HA (Apatite) layer detected around $2\theta = 31.77\text{-}32.90^{o}$. Therefore, the attempt to get an HA coating on the Ti screw surface by biomimetic coating has been accomplished within 1-year duration of the project. Moreover, there is some information obtained from this experiment that will be of help in pursuing further experiment (on both biomimetic and electroplating) and reproducibility.

3. Conclusion

The results from XRD showed that 35% H_2O_2-treated titanium screws, $60^{o}C$, for 24 hour by hydrothermal process could induce the apatite phase as clearly shown in the XRD pattern (Fig.10 B) at 2 theta = 32^{o}. This result implied that the hydrothermal process could replace heat treatment at $600^{o}C$ which was reported by Kokubo group[13, 14]. However, the hydrothermal process in H_2O_2 was too strong and it could corrode the autoclave equipment. Moreover, the color of Ti screw was also changed to the dull black color which might be from the oxidizing power of H_2O_2 solution.

Future suggestion

To pursue a success in coating, the right condition for Ti surface treatment has to be found and the following experiments should be tried:

1. Using new chemical additives to enhance the formation of biological HA on Ti surface.
2. Electroplating method on the Ti substrate using calcium phosphate solution.

We also would like to inform that, due to the time limit, the coating by electroplating was not included in this research.

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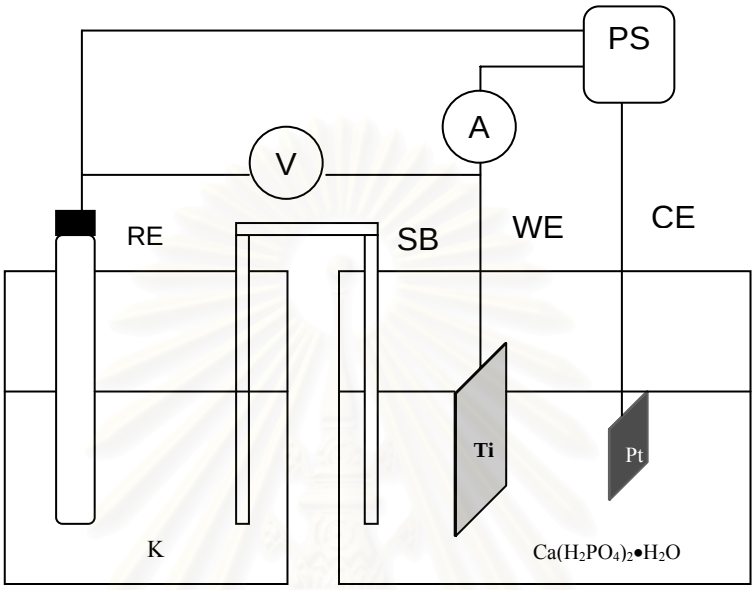
Presentations

1. Jinawath, S., Kashima P.D., Sujaridworakun, P., Jaruwungsunti, N., Tienboon, P., and Thamatakerngkit, S., Porous calcium phosphates: In-vivo and clinical studies (I). A poster presentation at 50th Birthday Anniversary of H.R.H. Princess Maha Chakri Sirindthorn, U.S.-Thai Symposium on Biomedical Engineering in Thailand, Chulalongkorn University, Bangkok, Dec.12th-15th, 2005.

2. Sujaridworakun, P., Kashima P. D., Tienboon, Jaruwungsunti, N., Itiravivong, P., Thamatakerngkit, S., and Jinawath, S., Porous calcium phosphates: In-vivo and clinical studies (II). A poster presentation at 6th ASIAN BIOCERAMIC SYMPOSIUM (ABC 2006), Sofitel Central Plaza, Bangkok, Thailand, Nov. 7-10, 2006.

Appendix 1

The experimental set-up for electroplating cell.



RE : Reference electrode Ag/AgCl
WE : Working electrode Ti
CE : Counter electrode Pt
SB : Salt bridge
PS : Potentiostat Galvanostat

Schematic diagram of experiment set up for electroplating on titanium substrate.

Appendix 2

The chemical amount and their purities for preparation of 1L of R-SBF (Revised-Simulated Body Fluid).

Formula of R-SBF solution

Reagent	Purities	Chemical Amount for preparation of R-SBF
NaCl	99.5 %	5.403 g
$NaHCO_3$	99.7 %	0.736 g
Na_2CO_3	99.9 %	2.036 g
KCl	99.5 %	0.225 g
$K_2HPO_4 \bullet 3H_2O$	99.0 %	0.238 g
$MgCl_2 \bullet H_2O$	98.0 %	0.311 g
HEPES*	99.5 %	11.928 g
$CaCl_2$	94.0 %	0.293 g
Na_2SO_4	99.0 %	0.072 g
1M-NaOH	-	1.5 mL

HEPES : [4-(2-Hydroxyethyl)piperazine-1-ethanesulfonic acid]

** HEPES = 2-(4-(2-hydroxyethyl0-1-piperazinyl)ethane sulfonic acid)

Porous Calcium Phosphates: *In-vivo and Clinical studies*

P. Sujaridworakun [1] D. Keshima Pongkao [1] P. Tienboon[2] , N. Jaruwungsunti[2] , P. Itiravivong[2] , S. Thamatakerngkit[3] and S.Jinawath[1]

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tract : *HA and HA/TCP composite were synthesized from DCPD/DCPA and $CaCO_3$, shaped and sintered at 1200°C to obtain porous mens. Chemical compositions of the materials involved were determined by ICP, phase analysis by XRD, and morphology by SEM. The porous mens having 37 vol% porosity composed of interconnected micro (< 5 μm) to meso (>20 μm) pores and a flexural strength of 17 MPa, were nted in a canine for 3 to 6 months. Histological study of the implant was performed and the result showed that there were new bone tissues rated into the implants which still survived after 6 months. Also some resorption of the implants was observed. The results of clinical trial on nts' anterior cervical neck bones after 6 month implantation, followed up by x-ray and CT scan radiography proved positive.*

terials and Methods

DCPD/DCPA + $CaCO_3$

↓ Mixing

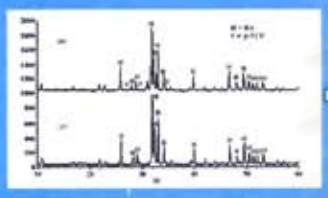
↓ Granulation

↓ Pressing (bars / pellets)

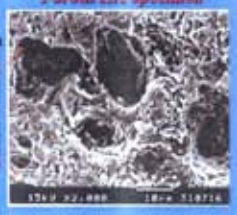
↓ Sintering at 1200°C / 2h

↓

, HA/TCP specimens



Specimens for *In Vivo* and Clinical studies

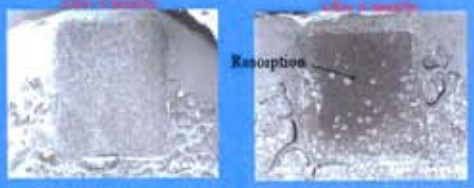


D-cage : substitute for human cervical neck bone

Porous HA : artificial bone used instead of bone graft from patient's hip bone

Results of histological study (*In Vivo*)

The porous HA specimen implanted in the canine tibia



icrograph of canine bone tissues indicated that there was no gap between the calcium phosphate specimen after implantation in the canine tibia for 6 and there was some noticeable resorption shown in the implant.

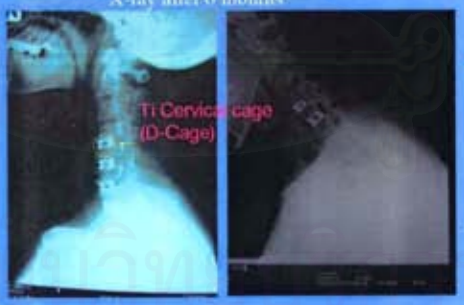
Bone tissue was dyed in toluidine

micrographs showed that the generation of new bone (paler color) tissues was ible and more clearly detected in the 6-month specimen, interpenetrating into the ed specimen and interconnecting with the old tissues (deep red color). The en showed a good bone conduction and biocompatibility.

Results of Clinical trial

Ti Cervical D-cage was used as a replacement of the patient's anterior cervical neck bone

X-ray after 6 months



CT scan after 6 months

The CT scan and x-ray results of clinical trial after 6-m implantation showed no inflammation, and newly conducted bone tissues had grown into the gaps. Moreover, the patients recovered very quickly and could move their necks almost 180°.

Conclusion : *Porous HA and HA/TCP ceramics with flexural strength and porosity ranging from 9-17 MPa, 45-37 vol% and 8-15 MPa, 46-36 vol%, respectively were successfully produced in our laboratory. The results of in-vivo test on animal and the clinical trial on patients' anterior cervical neck bones (13 cases , 24 pieces of ceramics) using porous HA specimens proved positive.*

cknowledgements : The authors would like to acknowledge the financial supports of Rachdaphisekasompot endowment, MTEC, and the technological support through the OECF-TJTTP the collaborations from the team of orthopedic surgeons from Chulalongkorn Hospital and the team of pathologists from Siriraj Hospital are much appreciated.

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Porous Calcium Phosphates: Fabrication, in-vivo and clinical studies

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* Department of Orthopedic, Chulalongkorn Hospital.

**Department of Forensic Medicine, Siriraj Hospital, Bangkok, Thailand

ract : High purity DCPD and DCPA were synthesized from bovine bone ash or the waste from the manufacture of ucts derived from it. Then DCPD and DCPA were each mixed with $CaCO_3$ in a stoichiometric molar ratio to form HA A/β-TCP composite. After pressing into shaped specimens and sintering at 1200°C, the resulted specimens became us HA or HA/β-TCP depending on the employed conditions. Chemical composition of the materials involved was rmined by ICP, phase analysis by XRD and FT-IR, and morphology by SEM. The porous specimens having 32-37 porosity composed of interconnected micro (< 5 μm) to meso (>20 μm) pores and a flexural strength of 17 MPa, were anted in a canine for 3 to 6 months. Histology study of the implant was performed and the result showed that there new bone tissues penetrated into the implants which still survived after 6 months. Also some resorption of the ants was observed.

ical application : D-cage was used as a replacement of the patient's anterior cervical neck bone

Porous HA specimen

In vivo study and histology test

fter 3 months

SEM micrograph indicated that there was no gap between the porous calcium phosphate specimen after implantation in the canine tibia for 6 months, and there was some noticeable resorption shown in the implant.

After 6 months

The generation of new bone (paler color) tissues was appreciable and more clearly detected in the 6-month specimen, interpenetrating into the implanted specimen and interconnecting with the old tissues (deep red color).

The specimens showed a good bone conduction and biocompatibility.

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