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Original Article

The Development of a Novel Bone Filler, Titanium Wire Ball

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In designing bone fillers, hardness has heretofore not been a major concern. Fillers are typically very hard and thus often accelerate the collapse of the adjacent bones. We developed a novel, relatively soft bone filler, whose hardness is similar to the human cancellous bone. The structure is simple: a 0.14-mm-diameter pure titanium wire was rolled and folded with both ends buried in the central portion, resulting in a ball of 4-mm diameter with 83% internal void ratio, having 300-500 μm internal gaps. The balls are chemically washed in an acidic solution at the end of the manufacturing process. We call this new filling device titanium wire balls (TWBs). We implanted TWBs into the medial condyle of the right tibiae of twelve adult Japanese white rabbits, and histologically evaluated the results. Four weeks after implantation, the spaces in the TWBs were fully calcified; the TWBs, the calcified tissues in them and the cancellous bones surrounding them were all connected with each other. In conclusion, we developed a novel bone filler that has similar hardness to the human cancellous bone and an 83% internal void ratio, with 300-500 μm internal gaps. Four weeks after implantation, the spaces in TWBs were fully calcified and connected to the surrounding cancellous bones.

Key words: bone filler, hardness, void, titanium, bone conductivity

In developing bioactive bone fillers, bone conductivity, porosity and mechanical toughness have been major concerns. However, comparative hardness to the adjacent bone has not been considered as an essential issue. The recent increase in fractures in elderly people, for example vertebral compression fractures and distal radial fractures, has increased the opportunity to use several kinds of bone fillers. Several bone fillers, such as polymethyl methacrylate (PMMA) bone cement, calcium phosphate paste, and small hydroxyapatite blocks have been introduced. PMMA cement is known for adverse effects such as leakage of the semi-hardened cement or monomer into the vertebral canal, resulting in radicular or spinal compression, and into the systemic circulation, resulting in pulmonary embolism, respiratory and cardiac failure, and even death [1,2]. PMMA cement can also become very much harder than the adjacent trabeculae, especially they were osteoporotic, and thus might accelerate their secondary collapse [3]. Calcium phosphate paste does not generate any heat and is considered to be replaced by the host bone [4]. However, the final hardened material, hydroxyapatite, is also very hard. In one instance, small blocks of hydroxyapatite were packed percutaneously into a painful cleft [5], but each of them was highly crystallized and also hard.

Hence, we designed titanium wire balls (TWBs) as a novel bone filler that has similar hardness to the human cancellous bone along with good bone conductivity.

Materials and Methods

Titaniumm wire balls. TWBs were made at Horie

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Corporation (Niigata, Japan) using pure titanium wire with an oxygen content of 0.25%. The wire was processed into 4-mm-diameter balls, with both ends of the wire buried in the central portion to avoid self-relaxing. We constructed 3 types of TWBs that had 74%, 83% and 87% internal void ratios using 3 types of titanium wires, having diameters of 0.134, 0.140 and 0.142 mm, respectively. They were then chemically washed in an acidic solution. The TWBs with 83% internal void ratio were used for implantation in experimental animals, and X-ray computerized tomography (Latheta LCT-200, Hitachi Aloka Medical, Tokyo, Japan) was performed before implantation.

Animals. Twelve male Japanese white rabbits were purchased from Hamaguchi-Dobutsu Inc. (Hyogo, Japan). Rabbits were fed individually in compartmental cages under standard conditions. All experiments were conducted according to the guidelines of the Animal Welfare Committee of Kindai University.

Human cancellous bones. Human cancellous bone specimens were obtained from the tibial condyle during total knee replacement, after obtaining the patients' consent. Seven cancellous bone blocks (10 × 10 × 5 mm) were subsequently prepared from 7 different individuals who had not been diagnosed with osteoporosis. There were 3 knees of males and 4 knees of females with a mean age of 71 years old (range, 65-78 years old). All 3 patients were diagnosed with osteoarthritis. The mean spinal bone mineral density of the patients was 0.833 ± 0.069 g/cm^2 (range, 0.771-0.987 g/cm^2). All experiments were conducted according to the Ethical Committee of Kindai University.

Measurement of hardness. Compression tests were performed on TWBs for each void ratio group and also on cancellous bone blocks, respectively, at a crosshead speed of 1 mm/min (Shimadzu Autograph AGS-J, Shimadzu Co., Kyoto, Japan). The measurements were performed five times.

Calculation of hardness. The compression tests showed the steepness of the linear portion of the stress-strain curves between 6 Newton (N) and 7 N. We subsequently used the reductions in diameter under 6 N to 7 N for calculation. We showed the hardness as N/reduction in diameter (mm) (N/mm).

Implantation and observation of TWBs. TWBs with 83% internal void ratio were implanted into the cancellous bone of the medial condyles of the right tibia of 8-month-old adult male Japanese white rabbits via 4-mm-diameter and 6-mm-depth drill holes. Sham operations, making voids of 4-mm diameter and 6-mm depth, were also performed in the cancellous bone of the medial condyles of the left tibia. Soft X-ray pictures of the implanted TWB were exposed by a soft X-ray radiator (Softex M-100, Softex Co. Kanagawa, Japan). At one, 2 and 4 weeks after implantation, rabbits were euthanized using pentobarbital and the TWBs were excised with the surrounding bone, fixed in 10% formalin neutral buffer solution for 24 h, mounted in epoxy resin, and sliced into serial sections of 5 µm. Serial undecalcified sections were observed with Villanueva Goldner staining using a light microscope (BZ-9000, Keyence, Osaka, Japan). The sites of the sham operation were also excised with the surrounding bone, fixed, mounted, sliced and observed similarly 4 weeks after implantation. Some sections including TWB were also observed using a Scanning Electron Microscope (SEM) (Hitachi SC-900, Hitachi Co. Ibaragi, Japan).

Statistical analysis. Results are presented as means ± SDs and were processed using Microsoft Excel 2007 (Microsoft Corp, Redmond, WA, USA) and a statistical calculating add-in (Statcel 4; OMS Ltd, Saitama, Japan). Differences between results were evaluated using Student's *t* test, and $p < 0.05$ was considered statistically significant.

Results

TWBs looked like small balls and were chemically washed in an acidic solution at the end of the manufacturing process (Fig. 1A). The X-ray computerized tomography of TWB also showed that the 83% internal void ratio comprised 300-500 µm gaps in the TWBs (Fig. 1B). The compression tests showed the steepness of the linear portion of each of the stress-strain curves between 6 and 7 N (Fig. 2A). So, we calculated the hardness under 6.5-N compression. The mean hardness of TWBs with 74% internal void ratio was 47.1 (SD7.5) N/mm (Fig. 2B). The mean hardness of TWBs with 83% internal void ratio was 39.4 (SD5.3) N/mm (Fig. 2B). The mean hardness of TWBs with 87% internal void ratio was 21.8 (SD2.0) N/mm (Fig. 2B). The mean hardness of the seven cancellous bone blocks was 41.9 (SD17.1) N/mm (Fig. 2B). The mean hardness of TWBs with 83% internal void ratio was most similar to the mean hardness of seven human cancellous bone

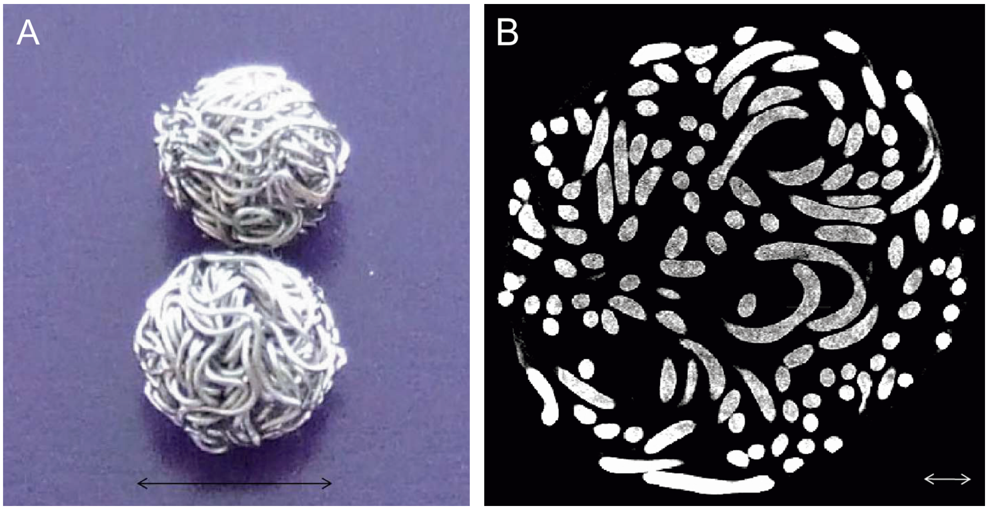

Fig. 1 Photograph and X-ray computerized tomography of a titanium wire ball (TWB). (**A**) TWBs were processed into 4-mm-diameter balls with 83% internal void ratio. (**B**) Both ends were buried into the central portion to avoid self-relaxing. The 83% internal void ratio created 300–500 μm gaps in the TWBs. Black arrow, 4 mm; white arrow, 400 μm.

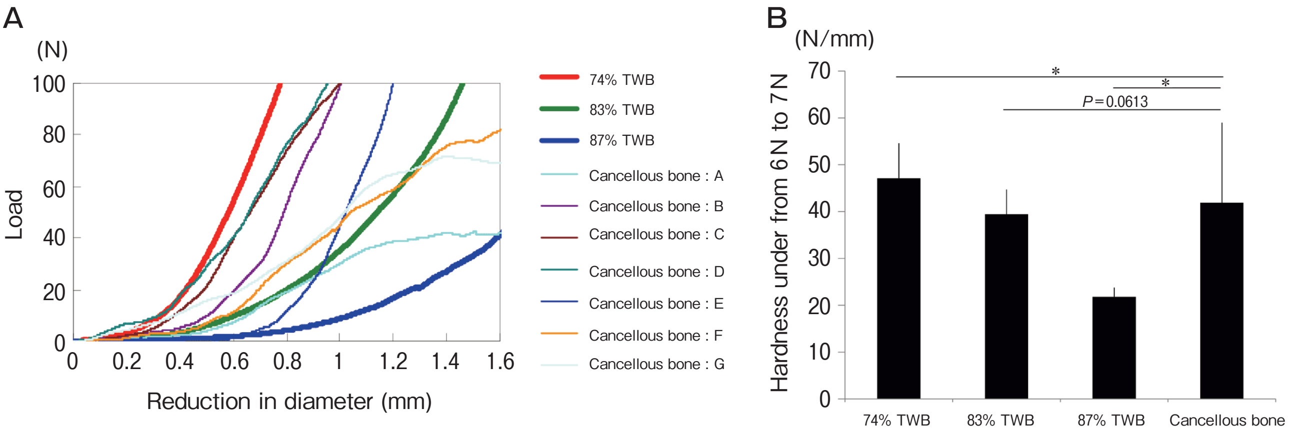

Fig. 2 **A**, Average compressive stress-strain curves of 3 kinds of titanium wire balls (internal void ratio: 74%, 83% and 87%) and 7 human cancellous bone blocks at a crosshead speed of 1 mm/min; **B**, Hardness of 3 kinds of titanium wire balls (internal void ratio: 74%, 83% and 87%) and human cancellous bone blocks under 6 Newton (N) to 7 N of force. The average compressive stress-strain curves showed the steepness of the linear portion of each of the stress-strain curves between 6 N and 7 N. The hardness of the titanium wire ball with 83% internal void ratio was not significantly different from that of human cancellous bone. The value of the cancellous bone presented in figure B was a mean of 7 human cancellous bone blocks. The values plotted into the figures were averages of 5 measurements. N, Newton; $^{*}p < 0.05$ between treatments.

blocks; no statistically significant difference was detected between these measures. So, we used TWBs with 83% internal void ratio in the subsequent examinations. A soft X-ray picture showed that TWBs were implanted into the medial condyles of the right tibia (Fig. 3). Micrographs of sections of the implanted TWBs showed that the osteoid tissues from the adjacent bone had already infiltrated the TWBs a week after implantation (Fig. 4A). Two weeks after implantation, the ingrown osteoid tissues were partially calcified (Fig. 4B). Four weeks after implantation, the spaces in the TWBs were fully calcified. The TWBs, the calcified tissues in them and cancellous bones surrounding them were all connected with each other (Fig.4C). Namely, in for weeks the artificial bone defects were reconstructed by structures having similar hardness to the surrounding trabecular bone including ingrown calcified tissue and TWB. The sites of sham operation were filled with granulation tissue; no calcified tissue was observed in the holes (Fig. 4D). The SEM of the cross section of the TWB, which was excised with the surrounding bone four weeks after implantation, showed that the internal titanium wire was infiltrated by osseous apatite deposits (Fig. 5).

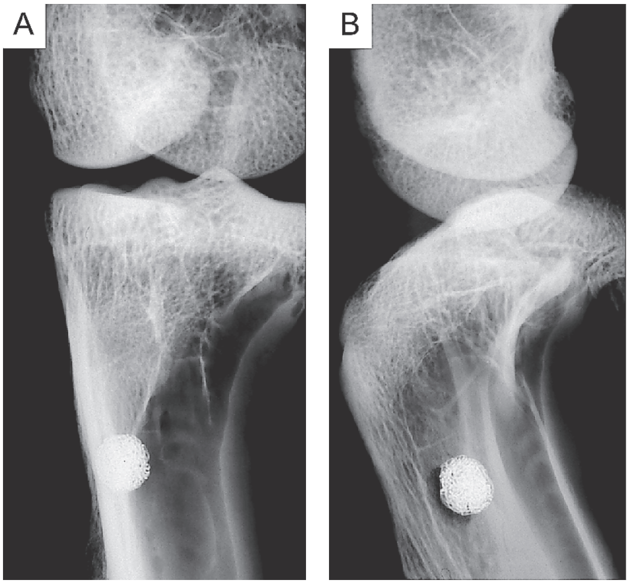

Fig. 3 Postoperative soft X-ray radiographs showing a titanium wire ball implanted into the medial condyles of the right tibia. **A**, Anteroposterior; **B**, lateral.

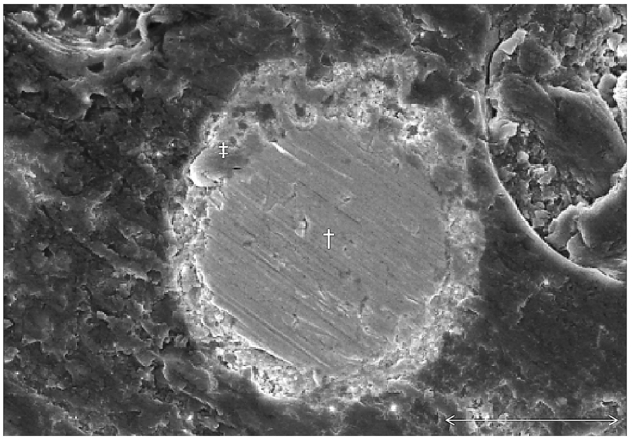

Fig. 5 Scanning Electron Microscope image of the cross section of the titanium wire ball extirpated with the surrounding bone 4 weeks after implantation. It is shown that the gaps in the titanium wire ball were occupied by osseous apatite deposits. † titanium wire, ‡ deposit. Arrow: 100 μm.

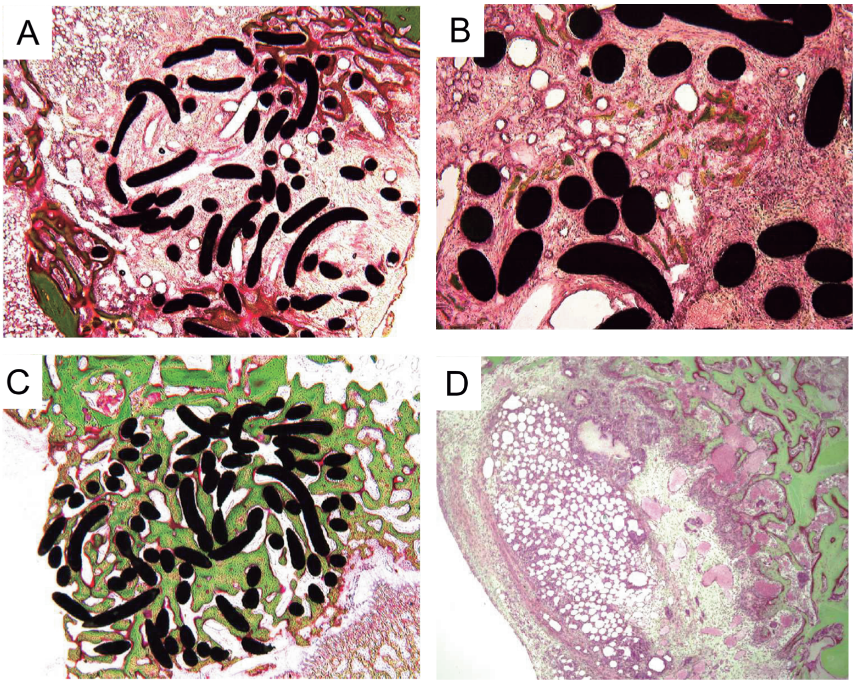

Fig. 4 Micrographs of sections of implanted titanium wire balls (TWBs). (**A**) One week after implantation. (**B**) Two weeks after implantation. (**C**) Four weeks after implantation. (**D**) Micrograph of the section of the sham operation 4 weeks after the operation. Each section was stained with Villanueva Goldner. One week after implantation, osteoid tissues from the adjacent bone had already entered the TWB. Two weeks after implantation, ingrown osteoid tissues were partially calcified. Four weeks after implantation, the spaces in the TWB were fully calcified. The TWB, calcified tissues in them and cancellous bones surrounding them were all connected with each other. The site of the sham operation was filled with granulation tissue; no calcified tissue was observed in the hole.

Discussion

In orthopaedic operations with bone defects, bone fillers represented by hydroxyapatites are now essential surgical devices. These fillers can be shaped freely, but it is usually not easy to change their hardness. The final product of calcium phosphate paste is biocompatible and can be degraded gradually by the bone tissue infiltrating the remaining cracks [6], but it is still very hard compared with the surrounding bone. Additionally, the hardness of bones naturally differs by many factors, including age, sex, underlying disease, part of the body and etc. Matching the hardness of implants to the hardness of patient's surrounding trabecular bone is usually not easy. So, orthopaedic surgeons implant ready-made bone fillers overlooking the unmatched hardness to the bones surrounding the implants. However, we were able to manufacture TWBs as a bone filler with hardnesses similar to that of the human cancellous bone. This means that using TWBs may reduce the mismatch between the hardness of the implant and the patient's surrounding trabecular bone.

Titanium is easily oxidized in the atmosphere or acid to have an oxidized titanium coat, which shows corrosion resistance [7-9]. Also, pure titanium is considered not to have the ability to bond to bone directly [10,11] while titanium with oxidized titanium coats bind to bone [12]. Namely, titanium is considered to bind to bone across oxidized titanium coats. Also, the layer of osseous apatite on the surface of titanium is necessary to obtain the ability to bind to bone [13-18]. Acid-treated titanium is easily deposited by Ca^{2+} and PO^{4-} in simulated body fluid [19-21]. SEM showed that osseous apatite is well deposited around the implanted TWB. This deposit was considered to be induced by the acid treatment in the manufacturing process.

Bone conductivity is one of the most important characteristics in the development of orthopaedic implants. On this point, researchers have tried various approaches. To obtain bone conductivity on a titanium surface, plasma-sprayed hydroxyapatite is most popularly used [22]. Additionally, some researchers previously reported that a close gap between titanium metals enhances apatite deposition [23-26]. Especially, apatite formation on two pieces of titanium was found to be most vigorous when they were arranged at a 300-500 μm gap [27-28]. To this end, TWBs have very advantageous internal structure. X-ray computerized tomography of TWBs showed that the 83% internal void ratio resulted in 300-500 μm gaps in TWB. Namely, the internal structure of TWB was considered to be especially favorable for apatite formation.

We acknowledge a limitation in our study. As a bone filler, TWBs showed some distinctive properties including their hardness being similar to the human cancellous bone and their internal structure being favorable for apatite formation. However, whether these profiles would improve clinical results remains uncertain. In order to investigate the clinical utility of TWBs, comparative studies to other established bone fillers are required.

We developed a bone filler made of titanium wire balls (TWBs) having similar hardness to human cancellous bone. Acid treatment and the internal structure with 300-500 μm gaps might promote apatite formation in TWBs.

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