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Development of Revolutionizing Methods for Hybrid-Type Artificial Organs by Means of Bioceramics and Biomechanics

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Abstract

The phylogenetic transition of branchiae into the lung as well as the migration of hemopoietic activity from the spleen to the bone marrow cavity demonstrate the concurrent evolutionary changes of organs structures and functions in vertebrates in response to the effect of gravity during terrestrialization from a water environment. The present study introduces a hybrid-type artificial bone marrow chamber that uses sintered hydroxyapatite (HA) and titanium electrode, which is able to induce hemopoietic nest formation in conjunction with osteogenesis not only heterotopically in mammalian muscle, but heterospecifically in archetype vertebrates, which have no bone marrow hemopoiesis in inner skeletons [1-2].

As well the gomphotic tooth system developed during the third revolution of the vertebral evolution by biomechanical stimuli, i.e., by corresponding of teeth and jawbones of animals to the masticatory movements.

The authors developed artificial root of corrugated cone shape using sintered compact hydroxyapatite (HA), which could induce to be gomphotic as well as ankylotic binding system depending on biomechanical stimuli of occlusal function or complete rest of implanted root after operation [3-7]. Following results are obtained: Bone marrow hemopoiesis in artificial chambers could be observed heterotopically in all cases. Gomphotic artificial roots with cementum were developed by biomechanical stimuli in the solid diet group. Ankylotic artificial roots were obtained in complete rest roots by the soft diet group.

Introduction

From evolutionary researches on vertebral skeletons authors disclosed that the bone marrow hemopoiesis developed during the second revolution of the vertebral evolution, i.e., terrestrialization by biomechanical stimuli, i.e., by corresponding of animals to the gravity action. As well the gomphotic tooth system developed during the third revolution of the vertebral evolution by biomechanical stimuli, i.e., by corresponding of teeth and jawbones of animals to the masticatory movements. Authors set the hypothesis that the cementoblasts as well as bone marrow hemopoietic cells, i.e., highly differentiated skeletal tissues can be induced in vivo not only by a harmony of the shape, function, and material effects of artificial bone implanted but elevated streaming potential, which had been brought about by correspondence of the organisms to the gravity action of the earth during landing (terrestrialization) of the vertebrates, i.e., physico-chemical stimuli (biomechanics in broad sense). Shape and functional effects mean biomechanical actions.

Materials and Methods

Two kinds of in vivo experiments were carried out to develop hybrid type artificial organs, i.e., (1) artificial bone marrow chamber and (2) gomphotic artificial root. Artificial bone marrow chambers made of hydroxyapatite (HA) and titanium (Ti) electrode as well as artificial roots were fabricated by Asahi Optical Co. and N.I.R.I.M. They were implanted into dorsal muscles and the jawbones of shepherds with 35kg and these of Japanese monkeys with 12kg respectively.

(1) The following development and experimental evolutionary studies were carried out using artificial skeletal organs.

1) Development of several kinds of artificial bio-chambers using:

① Conventionally sintered HA by Asahi Optical Co.

② High pressure sintering collagen-HA composite (by National Inst. for Research in Inorganic Mat.; collagen was extracted from cow skin.)

③ Titanium (Ti) mesh with a 10μA current by National Inst. for Research in Inorganic Mat.

2) Implantation of the artificial bone marrow chambers into cyclostomata (hogfish), chondrichthyes (dochishark), amphibian (xenopus), aves (chicken) and mammals (dogs and Japanese monkeys) were carried out.

3) Transplantation of Ti bio-chamber with a 10 μA current into muscles of sharks and dogs were carried out.

(2) Development of hybrid type gomphotic artificial root

The authors developed artificial root of corrugated cone shape using sintered compact hydroxyapatite (HA), which could induce to be gomphotic as well as ankylotic binding system, which are depending on biomechanical stimuli with occlusal function or complete rest of implanted root after operation. Artificial roots of sintered compact HA were implanted into jawbones of dogs and monkeys to verify how gomphotic can be induced by biomechanical stimuli. For this purpose, artificial roots of same configuration were implanted into mammalian jaws with or without occlusal functions. These animals were separated into two groups during the experiment. The first group was fed with a soft diet. The second group was fed with a solid diet. After a fixed period specimens were taken out and recovered. Histopathological observation by light microscopy as well as by SEM was carried out. Specimens with a mirror surface were prepared for microanalyzer using Kevex 8000.

Results

Following results are obtained: Bone marrow hemopoiesis in artificial chambers could be observed heterotopically in all cases. Artificial induction of hemopoiesis was carried out successfully in each of the newly developed chambers for all species. The leukocyte and lymphocyte induction around the Ti bio-chamber with a 10μA current in specimens of a dog 4 months after implantation was observed. As well leukocyte production induced from undifferentiated mesenchymal cells by the electrical current around the Ti was observed. Induction of hemopoiesis 12 months after surgery in conjunction with osteogenesis by the HA bio-chamber implanted in a Japanese monkey was observed. Osteogenesis as well as hemopoiesis could be observed around the chamber. In dog muscles around the collagen-HA bio-chambers implanted, marked tissue differentiation that resembled digestive tract formation could be observed. In specimen of the collagen-hydroxyapatite artificial bone marrow chamber implanted in shark muscle, hemopoiesis and osteoid formation 4 months after surgery around the hydroxyapatite were obseved. In a cross section of a shark (dochi), no bone marrow in the cartilaginous tissue around the spinal cord is evident. In a cross section of a shark with a Ti bio-chamber with a 10μA current. The histopathological findings associated with the dorsal cartilage with hemopoietic marrow induction by the adjacent Ti bio-chamber could be observed.

Gomphotic artificial roots with cementum were developed by biomechanical stimuli in the solid diet group. Ankylotic artificial roots were obtained in complete rest roots by the soft diet group, which had no tooth-supportive organ of periodontal tissues.

A layer of connective tissue rich in blood vessels was present in constant width around the artificial roots of hydroxyapatite and bone tissue similar to the alveolar bone proper formed around this layer. At the concave site of the artificial root, the root surface was fixed to the alveolar bone by means of fibrous connective tissues that ran at right or acute angles. In most cases, the bone was indirectly fixed with angled fibrous tissue to the artificial root surface. The layer of fibrous tissue was attached to the root surface with parallel orientation. At the convex site, fibrous connective tissues ran parallel with the artificial root surface. Observation by light microscope and SEM with Kevex 8000 of the mirror-polished surfaces of dog specimens revealed that in all artificial roots of the three different materials, alveolar bone proper (lamina dura) with trabeculae was observed, which was assumed to coincide with the principal stress trajectories as seen in specimens. The surface of the sintered hydroxyapatite appeared porous and seemed to remineralize. By SEM, calcified substance was observed attaching to the hydroxyapatite artificial root surface [5]. Calcium and phosphate contents similar to cementum or bone were detected by the Kevex 8000 microanalyzer system [5]. The surfaces of the titanium artificial roots showed no change and lacked calcium substance when studied by Kevex 8000.

At the attached gingival region of hydroxyapatite artificial root, no inflammation was found in the undecalcified specimen, in the decalcified specimen, and many cells resembling epithelium were observed by SEM. At the surface nearer the apex, various attaching structures made of microplatelets were observed.

At the surface of the hydroxyapatite artificial root, a laminated layer resembling calcified materials 20 μm in thickness was observed in a 11-month-postop specimen of a dog by SEM. Quite a few cells resembling cementoblasts were observed at the concave surface of clinically-applied hydroxyapatite roots 11 months postop. In some areas of a clinically used 24-month-postop artificial roots, these cells appeared to be covered with calcified substance. Occasionally, cementoblast-resembling cells were thought to be calcified with fibrous tissue. Cementoblast-resembling calcified cells observed by SEM at the concave surface 24 months postop, were corresponding to the numerous cells seen at the surface of the lamellae with light microscopic specimen of dog 6 months postop. The surface substance attached to the artificial root was analyzed by FT-IR. As a result, the attached substance proved to be a calcified thin layer of the cementum.

Microscopic findings also showed a calcified layer attached to the root surface 17 months postop. By microanalyzer, calcified substance attached to the artificial root surface was shown to be porous, which suggested remineralization at the surface of the sintered hydroxyapatite by mesenchymal cells like deposition was substance-specific to sintered hydroxyapatite.

Discussion and Conclusion

Heterotopical hemopoietic nests concomitant with ossification can be induced by fluid dynamics which are converted into streaming potential at the surface of bioceramics. Differentiation of mesenchymal cells was proved to be controlled by biomechanical stimuli. This differentiation coincides with the development of gomphotic tooth by biomechanical stimuli at the cellular level through mammalian jawbone evolution. From the experiments it is evident that development of bone marrow hemopoiesis as well as the gomphotic mammalian tooth system in phylogeny are dependent upon corresponding of animals to the gravity action. Therefore the evolution of the vertebrates is controlled by the biomechanical stimuli with the same genetic characteristics. As conclusion hybrid-type organs with highly differentiated tissue can be developed by means of bioactive ceramics conjugated with biomechanical stimuli, which trigger the gene expression of cells of the organs.

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References

[1] K. Nishihara, Tissue Engineering for Therapeutics Use 1 Organ, (1998), pp39-50.
[2] K. Nishihara, Bioceramics 12 (1999), pp253-256.
[3] K. Nishihara, S. Nakagiri and T. Akagawa, The First Congress of Biomechanics (1990), p114.
[4] K. Nishihara, T. Kobayashi and T. Akagawa, Bioceramics, 3 (1990), pp171-179.
[5] K. Nishihara, T Kobayashi and T. Akagawa, Phosphorous Research Bulletin 1991, pp185-190.
[6] K. Nishihara, and T. Akagawa, Bioceramics, 3 (1990), pp183-192.
[7]. K. Nishihara and T. Akagawa, Bioceramics 4 (1990), pp223-230.

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