News Release

Title

Heat-stimuli-enhanced osteogenesis using clinically available biomaterials

Key Points

- Hyperthermia with clinically applicable materials effectively induced newly formed bone in the rat and rabbit.
- Hyperthermia effectively enhanced osteogenesis in two different animal models
- Appropriate heat-stimuli (45℃ 15min, once a week) can promote enhanced osteogenesis.
- Heat-stimuli induced enhanced osetogenesis mediated via osteoblastic differentiation.

Summary

Assoc. prof. Yoshihiro Nishida, Dr. Takehiro Ota at Department of Orthopaedics surgery, Nagoya University Graduate School of Medicine (Dean: Kenji Kadomatsu, MD, PhD), revealed the Hyperthermia with clinically applicable materials (Resovist® and REGENOS®) effectively induced newly formed bone in the rat and rabbit.

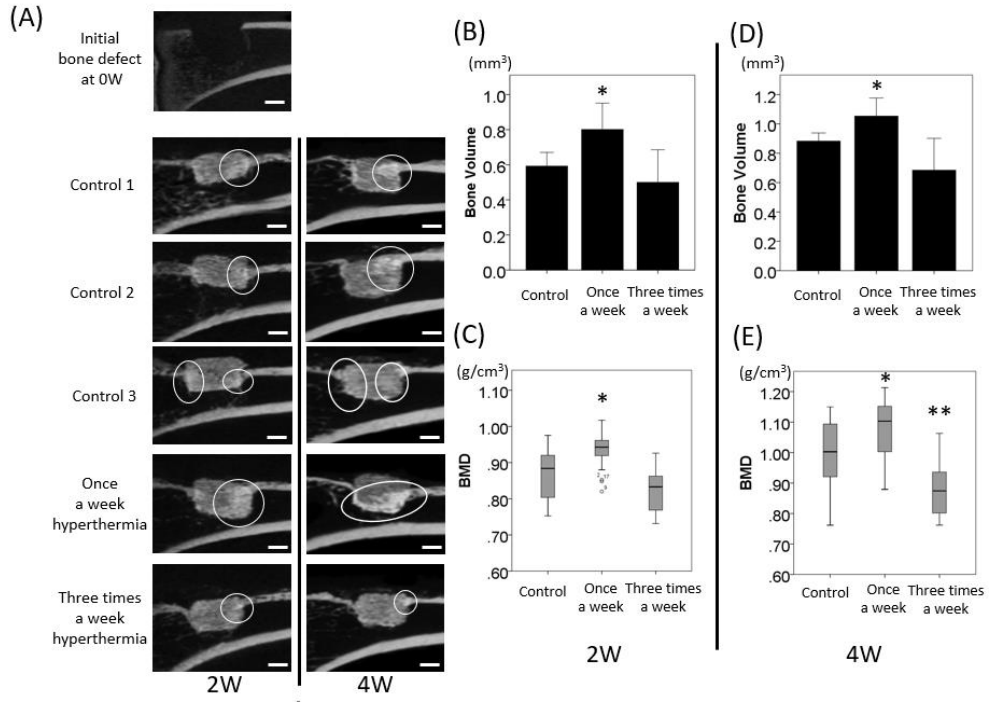
Several methods are used for patients with bone defects after resection of bone tumors, complicated bone fractures, and chronic bone infections. There is an urgent demand for the development of more effective novel methods for the promotion of osteogenesis. Our previous study reported that heat stimulus induces osteogenesis using alginate gel and magnetite cationic liposomes. However, for clinical use, the efficacy for promoting osteogenesis needs to be investigated using clinically approved materials. The researchers established a tibial defect model in rats and rabbits and evaluated heat stimuli-triggered (45℃ 15min) osteogenesis using alternating magnetic fields with already clinically applicable materials. The researchers also examined the effects of hyperthermia in two cell lines, osteoblast-like MC3T3 cells and ATDC5 in vitro. Micro-CT assessment and Histological examination revealed that a significantly stimulated osteogenesis was observed in the once a week hyperthermia group of both rats and rabbits as compared to the control group at two and four weeks after the start of hyperthermia. In contrast, the three times a week hyperthermia group did not show enhanced osteogenesis. Heat stress also enhanced alkaline phosphatase expression in cultured osteoblastic cells, MC3T3. On the other hand, heat stress had no obvious stimulatory effects on endochondral ossification using cultured ATDC5 cells. Our study demonstrates that heat-stimuli with clinically applicable heating materials can promote significant osteogenesis, and may thus be a promising novel treatment option for diseases associated with bone defects.

Research Background

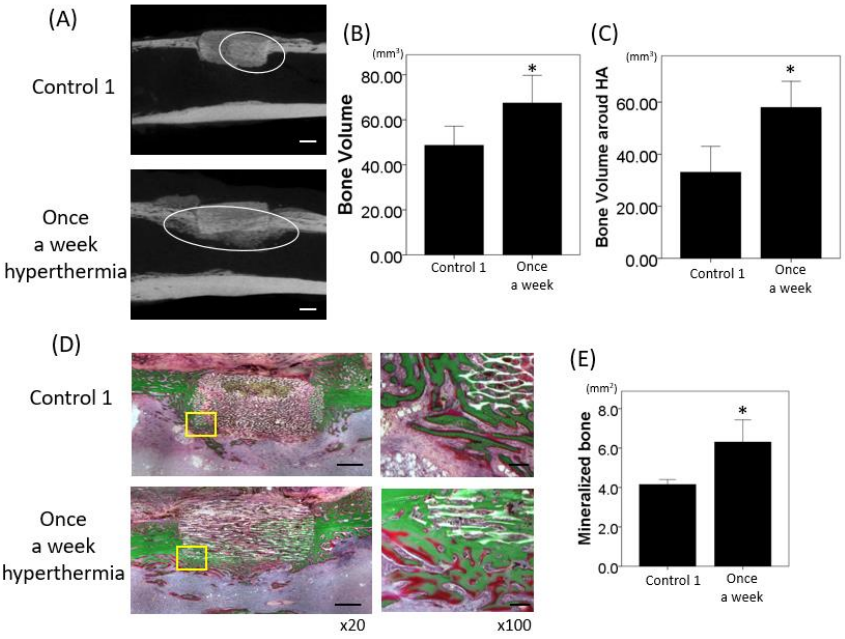
Intractable bone defects after resection of bone tumors, complicated fractures, and chronic infections bother both patients and physicians, and should be adequately treated. Autologous bone grafts have long been used because of their osteoinductive activity and subsequent osteogenesis. However, the collection of sufficient autologous bone is occasionally difficult, and pain and wound of the donor site are concerns for patients. Consequently, there is a demand for the development of more effective methods for the promotion of osteogenesis. Hyperthermia has been applied for inflammatory disease and metastatic disease clinically. Several previous reports using human mesenchymal stem cells, bone marrow stromal cells and some of cell lines, demonstrated that heat stress induces differentiation or proliferation of osteoblasts, alkaline phosphatase (ALP) activity, and osteogenic markers in vitro. However, there is no report demonstrating heat-stimuli induced osteogenesis using clinically applicable materials.

Research Results

Micro-CT assessment at two and four weeks after the start of hyperthermia (45℃ 15min) revealed that a significantly stimulated osteogenesis was observed in the once a week hyperthermia group of both rats and rabbits as compared to the control group. In contrast, the three times a week hyperthermia group did not show enhanced osteogenesis. Histological examination and image analysis showed consistent results that area of mineralized bone formation in the once a week hyperthermia group was significantly increased compared with that in the control group at two and four weeks. Newly formed bone was observed in the grafted materials from the periphery toward the center, and more osteoclasts were found in the once a week hyperthermia group. Heat stress also increased alkaline phosphatase expression in cultured osteoblastic cells, MC3T3, in vitro. On the other hand, heat stress had no obvious effects on endochondral ossification using ATDC5 cells.



Radiographic evaluation of newly formed bone with hyperthermia using a rat model. (A) Sagittal micro-CT image of the rat right tibial defect, control 1, 2, and 3, once a week, three times a week hyperthermia groups at 2 weeks and 4 weeks. White circling depicts newly formed bone at each time point. Measured bone volume (B) and BMD (C) in the rat models were graphed at 2 weeks. Bone volume (D) and BMD (E) in the rat models were graphed at 4 weeks. *p<0.05 compared with control group. **p<0.05 compared with control and once a week hyperthermia group.



Radiographic and histological evaluation of hyperthermia in rabbit model. (A) Sagittal micro-CT image of the right tibia in control 1 and once a week hyperthermia at 4 weeks. White circling depict the newly formed bone in (B) and around (C) the grafted materials at 4 weeks.

*p<0.05 compared with control 1 group. (D) Villanueva bone staining of the tibia of rabbit models in control 1 and once a week hyperthermia group at 4 weeks. Mineralized bone was stained as green and osteoid as red. Right column depicts the higher magnification of left column. (E) The areas of mineralized bone at 4 weeks was calculated and graphed. All data are expressed as the mean ±SD. *p<0.05 compared with control 1 group.

Research Summary and Future Perspective

We used clinically applicable materials, HA and Ferucarbotran composite, for hyperthermia in the rat and rabbit, and found that hyperthermia effectively induced significantly more bone formation in the defect. No severe side effects were observed in the experiments. These results show that appropriate heat-stimuli with clinically applicable heating materials can promote osteogenesis effectively, and that this procedure with combination materials may be a promising treatment option for bone defects in various skeletal diseases in the future.

Publication

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