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Title	ナノ秒レーザ光の骨形成作用:骨粗鬆症モデルおよび 健常ラットにおける検証
Author(s)	二宮,禎
Citation	
Issue Date	2002-09
Туре	Thesis or Dissertation
Text version	none
URL	http://hdl.handle.net/10119/2127
Rights	
Description	Supervisor:西坂 剛,材料科学研究科,博士



Bone Formation by Nano-second Laser Irradiation in Unloading and Loading Rats

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Near infra-red light is permeable to the skin and the soft tissue reach the bone tissue and marrow and then the light is absorbed the tissue layer containing pigments such as hemoglobin. Therefore, a short pulse and high peak fluence light can be directly generated a stress on the bone tissue and marrow. If a LISW was generated in bone tissue or bone marrow, it may stimulate bone formation in a similar fashion to mechanical loading or ultrasound. I have hypothesized that pulsed laser light irradiation stimulates bone formation through stress. The purpose of this study was to demonstrate the effects of high-intensity pulsed laser irradiation on bone formation and to be clearly the mechanism of this bone formation.

The anterior facies femoralis of the hind-limb of the unloading and loading rats were irradiated with laser light from a Q-switched Nd: YAG laser with a wavelength of 1064 nm. Histomorphometry of specimens of trabecular bone was also performed using a confocal laser scanning microscope (CLSM). The mean bone volume and mineral apposition rate of laser irradiated group (Laser) were significantly higher than those of the non-irradiated group (Control). These values under the Laser of unloading rat were about 1.52 and 1.25-fold those of the Control, respectively. In loading rat, a value of bone volume under the Laser was about 1.27-fold that of the Control. These findings revealed that laser irradiation accelerates bone formation.

The number of bromodeoxyuridine (BrdU) positive cells and tartrate-resistant acid phosphate (TRAP) positive cells were measured in the distal metaphyseal trabecular bone of the femur. BrdU positive cells were observed on bone marrow cells and chondrocytes in growth plate. Number of BrdU positive cell of the Laser was higher than that of the Control. BrdU labeled bone marrow cell was the highest on the third day after laser irradiation. Number of TRAP positive cell of the Laser was lower than that of the Control. TRAP positive cell was the lowest on the third day after laser irradiation. These findings revealed that laser irradiation increase a number of

osteoblasts and decrease a number of osteoclasts.

When laser light is applied to tissue, its wavelength and intensity should be considered because these parameters are related to its penetration depth into tissues and the magnitude of stress. Low-energy laser irradiation with a He-Ne laser, which has a wavelength of 632 nm and with a Ga-Al-As laser which has a wavelength of 830 nm, stimulates bone cells. However, the mechanism by which low-energy laser irradiation affects bone is not clearly known. It was recently found that light at a low radiation dosage is absorbed by intracellular chromophores, such as porphyrins and cytochromes. Chromophore that has absorbed light with 1064 nm is involved in hemoglobin. However, a major role of this protein is to deliver oxygen molecules into cells. This protein is probably not associated directly with bone formation. Therefore, it was considered likely that the light with 1064 nm does not have a photochemical influence on bone cells. Even if this light is absorbed into other intracellular chromophores, its influence will not be reflected in the present result.

The applied intensity of the low-energy laser light that was previously reported was 1.27-8.91 mW. In this study, high-intensity pulsed laser light with 6.25 or 12.5 MW/pulse intensity was employed. The laser light used in this study which a standard light (wavelength; 1064 nm, repetition rate; 10 Hz) with Q-switched Nd: YAG laser, repeats transient thermal expansion and relaxation every 100 ms. Moreover, because the light is scattered in irradiated tissue, the absorption volume becomes large. In short, the region where the pressure wave breaks out in bone tissue is sufficiently wider than the area of the irradiated spot. The generated pressure wave would therefore increase the marrow pressure. It was assumed that the observed effects of bone formation on the metaphysis are mainly due to this phenomenon. These bone formations induced by high-intensity pulsed laser irradiation might be due to laser-induced pressure waves.

This study indicated that high-intensity pulsed laser irradiation accelerates bone formation on metaphysis, enhances the activity of bone formation and inhibits resorption.