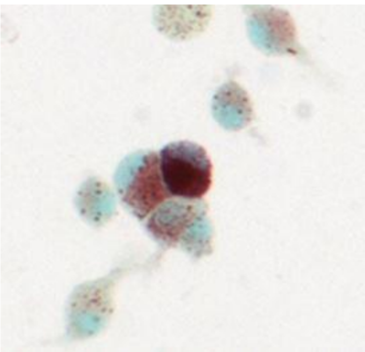
Study of the influence of a static MAGnetic field on the BONE remodeling.

MAGBONE



Jean Boisson, Unité de Mécanique de l'ENSTA-Paris

Amélie Coudert, BIOSCAR INSERM U 1132, Hôpital Lariboisière, Université de Paris-Cité

Abstract:

In certain medical situations, such as pediatric congenital facial deformities, it is necessary to perform mandible lengthening by a procedure called osteogenic distraction. The surgical operation consists of an osteotomy (surgical fracture) of the bone to be lengthened, followed by the placement of a distractor on either side of the fracture site. Recently, a new type of mandibular distractor was developed in ENSTA-Paris: the magnetic distractor [1, 2]. In this device, a cylindrical magnet ensures the activation of the osteogenic distraction. This device is one example of new medical devices using the magnetic performances of Neodymium allowing improvement of medical procedures. We can mention for example the PRECICE® device that is nowadays used for long bone distraction [3], or the LINX® Reflux Management System [4].

All these systems integrate a permanent magnet for their remote action properties. Generally, the influence of the static magnetic field on the biological environment is considered to be negligible [5]. However, few literature articles [6,7] observe effects of static magnetic field on bone cells that can give unpredicted therapeutic advantages for these new devices. It can give some insights for the development of future devices. The fundamental objective of this project is to improve knowledge on the influence of the magnetic field of magnets on bone healing. Biology aspects will be explored by submitting primary bone cells cultures (osteoblasts, osteoclasts) to different magnetic field intensity and orientation. Impact on bone tissues will be determined by extracting mechanical properties of femur bones and calvaria grown under a static magnetic in organotypic cultures.

Keywords: Magnetic field, bone remodeling, bone cells, bone organotypic cultures, mechanical properties

State of the art

Osteogenic distraction is a procedure allowing bone lengthening by elongation of a callus following osteotomy [8]. This technique is used to correct growth defects related to congenital facial deformities. In order to facilitate the distraction activation, a collaboration between Necker Hospital and ENSTA-Paris has allowed the development of a magnetically activated distractor (Figure 1A) [1, 2]. This project is an example among others of medical devices using the mechanical properties of Neodymium magnets (e.g. centromedular magnetic distractors, Precice®, Nuvasive [3]). These devices induce the presence, desired or not, of a static magnetic field of the order of one Tesla on healthy biological tissues or on tissues undergoing reconstruction, in particular, in the case that concerns us here, on bone. Indeed, the static magnetic field created by the magnet integrated to the distractor is within the normative limits to be innocuous. Few articles in the literature observed possible benefic effects on the bone remodeling from a static magnetic field. So, in this context, understanding the magnetic field influence on the different aspects of bone remodeling is crucial.

Studies have shown that static magnetic fields appear to have a beneficial effect on bone formation [9,10]. Indeed, from a biological point of view, some studies suggest that it promotes the differentiation of mesenchymal stem cells into osteoblasts, while increasing their proliferation [6,7,11]. The orientation of the bone matrix also seems to be modified in the presence of very strong magnetic fields (Figure 1B) [10]. Most of the studies were conducted on osteoblast cell lines. The effect on osteocytes and osteoclasts is not yet clearly identified [6]. From a macroscopic point of view, studies have shown that the density of bones increases in the presence of magnetic fields to such an extent that therapeutic use is being considered [9].

Objectives

The objective is to study the effect of a static magnetic field on bone remodeling biological process and mechanical properties of formed bone, using primary bone cell cultures (osteoblasts and osteoclasts) and organotypic cultures of mouse bones (calvaria and long bones).

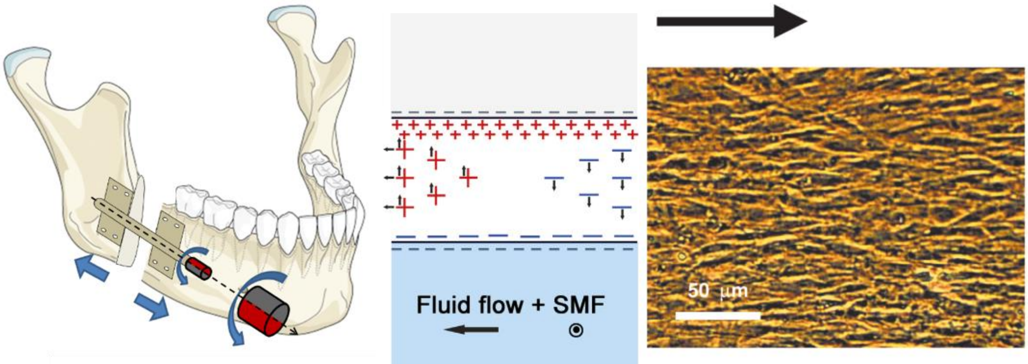


Figure 1 : On the left, principle of the VIVODOGMA device [1,2], in the middle, schematic diagram of the Hall effect in the bone matrix [6], on the right, alignment of bone cells according to the orientation of an intense magnetic field [5].

Methodology

Development of the culture system in a static magnetic field

A structure of permanent magnets, allowing the application a static magnetic field of approximatively 1 T on the bone cells will be positioned under the culture dishes. The objective of the first magnetic structure will be to apply the most homogenous magnetic field on the cell cultures. This way, possible effects due to magnetic field gradient effect will be prevented. The second magnetic structure will be based on a Hallbach array, that will create an intense magnetic field with controlled gradient. In this configuration, the magnetic gradient locations will be controlled, giving us an easier way to identify its possible effects on bone cells. The use of CAD (Computer Aided Design) and 3D printing will allow the fabrication of the magnetic structure.

Effect of a static magnetic field on the differentiation and function of osteoblasts

Using bone marrow from 6-week-old mice, mesenchymal stem cells, subjected to a static magnetic field, will be differentiated into osteoblasts for 21 days. The effect of a static magnetic field on differentiation will be evaluated by alkaline phosphatase staining (marker of early osteoblastic differentiation) and on function by staining bone nodules with alizarin red (allowing the evaluation of osteoblastic function i.e. mineralized bone matrix formation). The osteoblastic marker gene expression will be analyzed by qPCR.

Effect of a static magnetic field on osteoclast differentiation and function

From 6 weeks old mouse spleen, splenocytes will be differentiated into osteoclasts in a static magnetic field in the presence of MCSF and RANKL (osteoclastic stimulating cytokines). The effect of a static magnetic field on differentiation will be assessed by counting TRAP positive multinucleated cells (3 or more nuclei) considered as osteoclasts compared to the control condition. The effect of a static magnetic field on osteoclast resorptive function will be analyzed by assessing the resorption of a resorbable surface on which the cells have been cultured. The osteoclastic marker gene expression will be analyzed by qPCR.

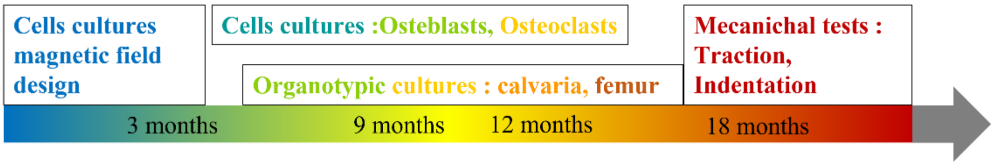
Effect of a static magnetic field on bone growth in organotypic model

Calvaria and femurs from newborn mice (3-5 days old) will be cultured according to the organotypic culture protocol developed by Kanczler et al [12]. The tissues are maintained in culture for 10 days in the presence of a static magnetic field. The bones are then analyzed by micro-computed tomography to evaluate the bone growth in the presence of a static magnetic field. This same bone growth will then be analyzed by histomorphometry to evaluate the effect of the static magnetic field on the rate of matrix deposition, on osteoblasts and on osteoclasts in these ex-vivo conditions.

Biomechanical study

We will apply mechanical tests on bone samples produced under a static magnetic field from organotypic cultures in comparison with those from the control condition. Given the millimetric size of the bone samples, first, indentation tests evaluating the hardness of the cortical material produced will be carried out in the Mechanical Unit of ENSTA. From the results, we will be able to determine the elastic (Young's modulus) and plastic (elastic limit) parameters of the bones under a static magnetic field. In a second step, tensile and bending tests will also be performed. The objective in a second time will be to correlate the results of these tests on bone fibers microstructure. For that, equipment from the LMS (école Polytechnique) will be used.

Project Timeline:



References:

[1] Boisson et al, J Craniomaxillofac Surg, 44, 684-688, 2016.
[2] Kadlub et al, BJOMS, 60, 767-772, 2022.
[3] https://www.nuvasive.com/procedures/limb-lengthening/precice-system/
[4] https://www.jnjmedtech.com/en-EMEA/product/linx-reflux-management-system
[5] Schenk, Biop Mol Biol, 87, 185-204, 2005*
[6] Zhang et al, Prog Biophys Mol Biol, 114, 146–152, 2014.
[7] Yang J et al, Biol Trace Elem Res, 184, 214-225, 2018.
[8] Mofid et al, Plast Reconstr Surg, 108, 1103–1114, 2001.
[9] Yan et al, Med Eng Phys, 20, 397–402, 1998.
[10] Kotani et al, J Bone Miner Res, 17, 1814–1821, 2002.
[11] Yamamoto et al, J Dent Res, 82, 962–966, 2003.
[12] Kanczler et al, Tissue Eng. - Part C Methods, 18, 747–760, 2012.

- **Project impact**

The design of high energetic permanent magnet in the 1980's lead to the creation of new medical devices decades later. The most famous of these being the magnetic resonance imaging. More recently, new devices using the remote force transmission of permanent magnet were created (PRECICE®, LINX®) opening the regulation way of other innovations.

Since 2016, our team develops a new surgical device: the magnetically activated mandibular distractor. This device design with pediatric maxillo-facial surgeons is intended to remove most of the complications of the facial osteogenic distraction. The magnetic distractor uses the interaction between two permanent magnets. The internal one, integrated to the implant, is on the patient mandible for at least 4-5 months. Due to the surgical configuration, the permanent magnet will be close to a fracture line for all the procedure. Therefore, its magnetic field could have a direct influence on the bone remodeling process. If, from a regulation aspect, a static magnetic field below 2 Tesla is considered as innocuous (directive 2013/35/EU), adequate data for proper risk assessment of static magnetic fields are almost totally lacking (SCENIHR 2007). Then, then production of such data will be in a great interest in this context of advent of new technologies. Especially since some articles of the literature [9,10] observed that a static magnetic field could have a benefic influence of the bone remodeling process.

The objective of the project is to quantify the influence of a static magnetic field of bone cells (osteoclasts and osteoblasts) and on the produced bone (organotypic cultures). If these two aspects are demonstrated in configurations close to the one in the magnetic distractor, this will shed a new light on this device by giving it a therapeutic advantage in addition to the complications removal of classic distractors. For example, the beneficial effects of the application of a magnetic field for bone consolidation could lead to a reduction in the consolidation period and an increase in bone volume after distraction. This could favor the use of this device developed in IP-Paris in future surgical routine

From a farer perspective, the understanding of the static magnetic field could give rise of new applications based on implanted permanent magnets. Indeed, during the development of the distractor, the group developed a rare know-how of use of permanent magnet in a medical context. Indeed, the biocompatibility and the resistance to sterilization processes are already solved problems giving real advantages to the team, if, at the end of this project, a new medical devices idea emerge. For example, we can imagine that static magnetic field based devices could be used in other indications, such as bone grafts, postoperative consolidation, and brittle osteopathies.

Finally, in a more fundamental perspective, we can imagine that understanding the influence of a static magnetic field on the bone cells will give a new insight o the bone remodeling process. Namely, the communication between cells (osteoblast, osteoclasts and osteocytes) is key to the remodeling. The magnetic field probably modifying this exchange, a good comprehension of its influence on the each cells lines could help to understand deeply how the cells communicate.

Environment

The department of engineering of ENSTA-Paris is equipped with two uniaxial machines dedicated to tests on biotissues that can be transportable if required. Those equipment are completed by tracking devices and conservation systems. For the microstructure part, LMS equipment will be used as a collaborative platform of the new mechanical center (ENSTA-Paris, Ecole Polytechnique) that will take place next year. The design of magnetic structures will be based on additive manufacturing that will give access to a large diversity of configurations.

5 chosen references over the past 5 years:

[A] Feudo S, et al, Nonlinear magnetic vibration absorber for passive control of a multi–storey structure, *J Sound Vibration*, 438, 2019.
[B] Heller U, et al, Decellularized vascularized bone grafts as therapeutic solution for bone reconstruction: A mechanical evaluation, *Plos one*,18, 2023.
[C] Rougier G, et al, Ramus Sagitall Osteotomy related biomechanical Properties, Br J Oral Maxillofac Surg., 58, 2020.
[D] Kadlub N, et al, Modeling of the human mandibular periosteum material properties and comparison with the calvarial periosteum, *Biomech Model Mechanobiol*, 19, 2020.
[E] Kadlub N, et al, Mandibular magnetic distractor: Preclinical validation, Br J Oral Maxillofac Surg., 60, 2022.

Selected reference

[1] Y. Chabbi-Achengli et al., “Decreased osteoclastogenesis in serotonin-deficient mice.,” Proc Natl Acad Sci U S A, vol. 109, no. 7, pp. 2567–72, Feb. 2012, doi: 10.1073/pnas.1117792109.

[2] A. E. Coudert et al., “Differentially expressed genes in autosomal dominant osteopetrosis type II osteoclasts reveal known and novel pathways for osteoclast biology,” Laboratory Investigation, vol. 94, no. 3, 2014, doi: 10.1038/labinvest.2013.140.

[3] A. E. A. E. Coudert et al., “Role of the captured retroviral envelope syncytin-B gene in the fusion of osteoclast and giant cell precursors and in bone resorption, analyzed ex vivo and in vivo in syncytin-B knockout mice,” Bone Rep, vol. 11, Dec. 2019, doi: 10.1016/j.bonr.2019.100214.

[4] N. Kadlub et al., “Molecular and cellular characterizations of human cherubism: Disease aggressiveness depends on osteoclast differentiation,” Orphanet J Rare Dis, vol. 13, no. 1, p. 166, Sep. 2018, doi: 10.1186/s13023-018-0907-2.

[5] N. Kadlub et al., “Defining a new aggressiveness classification and using NFATc1 localization as a prognostic factor in cherubism,” Hum Pathol, vol. 58, 2016, doi: 10.1016/j.humpath.2016.07.019.

INSTITUT
POLYTECHNIQUE
DE PARIS

ENSTA

IP PARIS