THE EFFICACY OF NATURAL CORAL WITH AND WITHOUT PLATELET RICH PLASMA IN PREVENTION OF POST-DENTAL EXTRACTION ALVEOLAR BONE LOSS

By

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ABSTRAK

KAJIAN TULANG GANTIAN DENGAN MENGUNAKAN KARANG SEMULAJADI DAN FAKTOR PERTUMBUHAN DALAM BIDANG PERGIGIAN KLINIKAL

Resorpsi tulang alveolar berlaku dalam jangkamasa setahun setelah gigi dicabut dan terjadi dengan kadar yang cepat pada tiga bulan yang pertama. Kehilangan tulang akibat cabutan gigi memberi kesan negatif pada kosmetik, kebersihan, prostetik dan struktur gigi. Oleh itu setelah gigi dicabut, pengekalan kontur tulang dengan bantuan bahan tulang gantian adalah penting untuk implan dental, rekaan pontik, kestabilan dentur, estetik tisu gusi dan mengekalkan status gigi bersebelahan. Tujuan kajian ini ialah mengekalkan ketinggian tulang alveolar dengan menggunakan karang semulajadi and faktor pertumbuhan. Pesakit diberi penerangan mengenai kajian ini dan sebelum persetujuan penyertaan diperolehi. Sembilan puluh enam pesakit berumur diantara 18 hingga 45 tahun dibahagikan kepada tiga kumpulan yang terdiri daripada 32 orang setiap kumpulan. Sebelum gigi yang rosak atau patah dicabut x-ray orthopantomograph digital (OPG) dilakukan untuk dijadikan ukuran sebelum pembedahan. Kumpulan I adalah penerima karang semulajadi (NC) sebagai bahan rawatan pada soket gigi yang dicabut, kumpulan II menerima karang semulajadi yang ditambah dengan faktor pertumbuhan (PRP) yang akan diambil dari darah pesakit sendiri dan kumpulan III tanpa apa-apa rawatan. Selepas proses rawatan berakhir, kawasan soket yang berkenaan dijahit dan pesakit diberi arahan penjagaan selepas cabutan dan antibiotik. Selepas cabutan, rawatan susulan klinikal dijalankan. Pengukuran aras tulang dilakukan dengan OPG susulan. Keputusan kajian ini menunjukkan terdapat pertambahan ketinggian tulang alveolar dalam kumpulan I dan II. Purata pertambahan tulang kumpulan I ialah 1.33 mm, SD (1.54) dan kumpulan II ialah sebanyak 0.88 mm, SD (0.92). Kumpulan III mengalami purata kehilangan tulang sebanyak -2.98 mm, SD (1.37). Ini menunjukkan resorpsi tulang alveolar sebanyak 3 mm dalam masa kurang

daripada 6 bulan. Keputusan kajian ini menunjukkan tiada perbezaan signifikan dalam kumpulan I dan II dalam mengekalkan ketinggian tulang alveolar. Kajian ini juga menunjukkan karang semulajadi dapat mengekalkan ketinggian tulang alveolar selepas gigi dicabut. Dalam kajian ini didapati bahawa penambahan faktor pertumbuhan untuk mempercepatkan pertumbuhan tulang tidak dapat dikaji kerana ujian histologi tidak dapat dilakukan keatas pesakit.

Chapter one

Introduction

Chapter One

Introduction

1.1. Background

The quantity and quality of maxillary and mandibular bone have long been a focus of attention for odontologists and implantologists for maintenance of oral and craniofacial health (Hausmann & Allen, 1997).

Alveolar bone plays a key role in providing support to the teeth, which are anchored to the bone by periodontal fibers. Most of the bone mass in the mandible consists of cortical bone in the basal portion. Mechanical stimulation of alveolar bone during mastication is crucial in keeping the teeth and underlying bone healthy. However, it has been estimated that the alveolar bone is subjected to mechanical loads for only 15–20 min per day (Bates *et al*., 1975). These mechanical loads that are basically the masticatory forces, have a direct effect on the bone mineral density (BMD) and thickness of the cortical bone mass in the mandible (Klemetti *et al.,* 1993).

Loss of teeth is followed by irreversible alveolar bone resorption. Alveolar bone resorption can be detected and measured by taking impressions and constructing study casts or by intraoral radiographs as shown in Figure 1.1.

Untreated dental disease results in osteolysis that inevitably leads to loss of teeth. In addition to anchoring the teeth to the alveolar ridges, the maxillary and mandibular bone allows dental restoration procedures, such as construction of root-supported implants, fixed dentures or removable dentures. However, the functional and cosmetic results depend on the quantity and quality of the maxillary or mandibular bone (Fischer-Brandies & Dielert, 1986).

Local and systemic factors influence the overall healing process. However, regardless of the factors leading to alveolar lysis, the baseline characteristics of the alveolar bone play a crucial role. In patients whose alveolar bone is adequate in quantity and quality at the time of tooth loss, the adverse consequences of bone resorption are noticeable few weeks to months later.

Three clinical criteria are useful for evaluating alveolar bone prior to tooth restoration. These criteria's are shape, size and volume, and density of the mandible that vary widely across individuals (Atwood D, 2001). This variability translates into differences in mandibular bone responses to a given local or systemic insult (Kingsmill & Boyde, 1998).

Many normal and abnormal processes can cause alterations of the maxillary and mandibular bone tissue. The alveolar bone that supports the teeth is particularly fragile and labile. The outer cortex, which is thin in the maxilla and thick in the mandible, continues the basal bone seamlessly, with no difference in structure (Jeffcoat et *al.*, 2000).

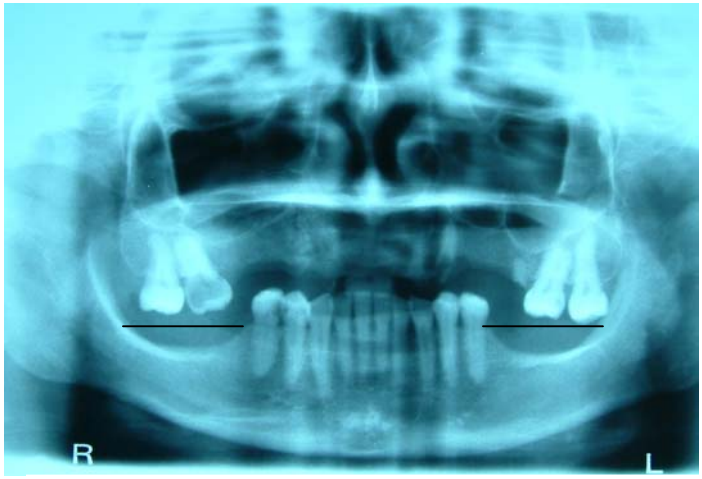

Figure 1.1. Orthopantomograph shows the radiolucent area (bone resorption area). Black line shows the level of the alveolar bone when teeth were present

Following tooth extraction resorption of alveolar bone is greatest during the first year and occurs at a particularly fast rate during the first 3 months, although up to fourfold variations have been reported across individuals over a 14 month period. In a given patient, the extent of bone resorption varies with age at tooth loss. In addition, marked differences can occur between the maxillary bone and the mandible. The amount of bone lost to the resorptive process has been estimated at

21% after 3 months, 36% after 6 months, and 44% after 12 month (Carlsson & Persson, 1967).

Bone stock preservation is a major concern for odontologists. The first step is prompt treatment of parodontal disease. However, tooth preservation is not appropriate when there is a risk of damage to the supporting tissues. When tooth extraction is necessary, trauma should be minimized during the procedure and bone preservation should receive careful attention. Similarly, to preserve bone stock, trauma should be kept to a minimum when fashioning the prosthesis. Healthy roots surrounded by healthy parodontal tissue can be preserved and used to anchor tooth implants, which delay and diminish post-extraction bone resorption (Vanwaas et *al.*, 1995).

Following an extraction, preservation of bone contour with the aid of bone graft materials are important considerations for dental implants as shown in Figure 1.2. Pontic design, denture stability, soft tissue aesthetics and maintaining the periodontal status of adjacent teeth are other important procedures of maintaining bone level. Bone loss following an extraction can have negative cosmetic, hygienic, prosthetic and structural consequences. Bone grafting into an extraction site can reduce these negative seqellae (Murray, 1998).

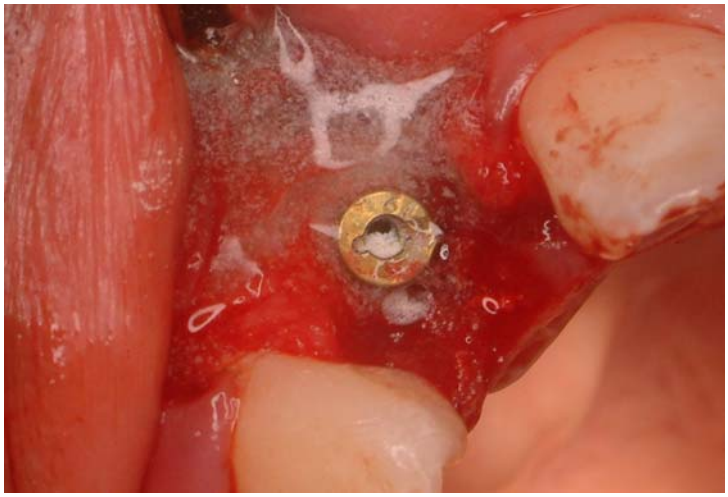

Figure 1.2. Bone grafting with NC prior to placement of dental implant

Augmenting post extraction sockets with bone graft material such as natural coral (NC) preserve alveolar bone height and volume until such time as the patient is ready to undergo definitive restoration with dental implant or prosthesis (Kurol & Odman, 1996). The bone grafting may act as an osteoinductor or osteoconductor or as both in supporting the new bone formation.

An alveolar sparing procedure using a bone graft substitute like NC could be beneficial to patients that undergo dental extractions. The time required for wound healing and incorporation of the coral granules could advantageously allow for the completion of jaw growth, all the while preserving the dimensions of the residual alveolar ridge. The goal would be to allow the placement of an implant, in an uncomplicated manner, without the need to harvest a bone graft from a second anatomic location, thereby minimizing postoperative morbidity. Therefore, ridge preservation techniques are often warranted after tooth extraction.

Over 25 years, the alveolar ridge may lose up to 10 mm in height at the mandible. Height loss is usually about four times smaller at the maxilla than at the mandible. This may be described to the fact that loads are distributed over a smaller surface at the mandible than at the maxilla. Bone resorption can be reduced, although not eliminated, by ensuring that the prosthesis applies balanced loads to the underlying bone (Sennerby *et al.*, 1988).

Bone mineral is composed of nano-crystalline platelets, originally described as hydroxyapatite. It is now agreed that bone apatite can be better described as carbonate hydroxyapatite (CHA). The composition of commercial CHA is similar to bone mineral apatite. It is generally accepted that natural and synthetic calcium carbonate & phosphate bioceramics are osteoconductive and have the ability to support tissue ingrowth and bone formation. They are not osteoinductive which means that they don't have the ability to form bone when implanted in non-osseous sites (LeGeros, 1988).

Bone contains macro- and micro-pores, which are interconnected to allow necessary body nutrients and fluids to be transported, making bone an extremely complex porous structure. Many have tried to mimic the unique qualities of bone; however nature has proved to be nearly impossible to adequately copy or emulate. Many considerations have to be addressed to produce an ideal synthetic bone material property such as the porous morphology and structure, type and volume fraction of the pores, the surface state and degradation rates (Kokubo *et al.*, 2000).

Dental and Maxillofacial applications of calcium phosphate bioceramics include repair of periodontal defects (Figure 1.3), alveolar ridge augmentation, maxillofacial reconstruction and implant coatings. Furthermore many challenging problems in maxillofacial surgery which include treatment of bridging diaphyseal defects, non-union, filling metaphyseal defects and mandibular reconstruction have been partially solved by the aid of these biomaterials.

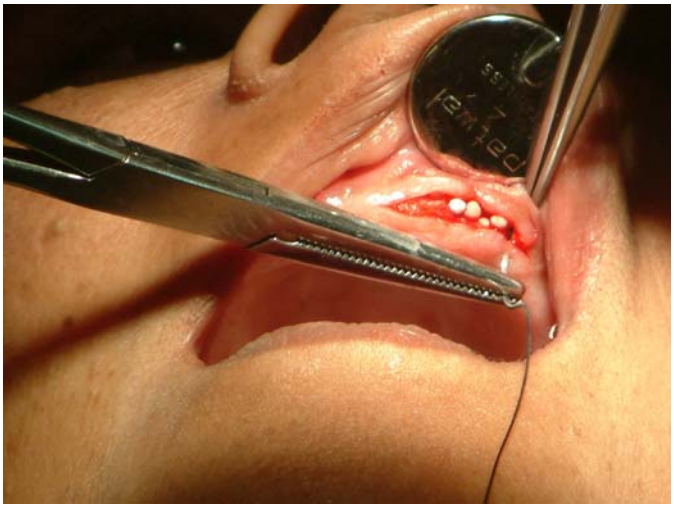

Figure 1.3. Repair of periodontal defects in the anterior maxilla with granular type Hydroxyapatite.

When a synthetic material is placed within the human body, the tissue reacts towards the implant in a variety of ways depending on the material type and applied functional loads. The only substances that conform completely are those manufactured by the body itself (autogenous) and any other substance that is recognized as foreign, initiates some type of reaction. In general, three terms describe or classify a biomaterial with respect to the tissue response. These are *bioinert, bioresorbable,* and *bioactive* (Degroot & Vincenzini, 1987).

Bioinert meaning that they must not trigger immune reactions or poison the implant environment. As for *Bioresorbable* that means the material should be degradable in order to be replaced with the new host cell. *Bioactive* as the name implies is the character of the material to exert a specific function.

The goal of this study was to increase bone volume, thereby promoting bone height and volume, as they produce loads within the normal range. Natural structural materials just recently started to surpass this by providing an abundant source of novel biomedical applications. Natural bioceramics such as animal skeletons (calcium carbonate or hydroxyapatite) are known to be designed through natural optimization methods to physically support and maintain the range of bone tissues (Tang *et al.*, 2003).

In this study we also combined Plasma Rich Platelets (PRP) with synthetic bone. These growth factors are well-established wound healing hormones. Growth factors have been shown to play important roles in the normal healing of bone. They play the role of osteoinductors within the osteoconductor scaffold (Hauschka, 1990).

1.2. Statement of the problem

Extraction of a tooth is followed by three dimensional bone resorption that hinders oral rehabilitation procedures. The resorption is lifelong, irreversible, chronic and cumulative (Hausmann & Allen, 1997).

Recent studies showed that there was a 25% decrease in the width of the alveolar bone during the first year and an average of 4mm decrease in height during the first year following multiple tooth extractions (Misch, 2000). Tatum and Misch have observed a 40%-60% decrease in alveolar bone width after the first 2 to 3 years post extraction and Christensen reported an annual resorption rate of at least 0.5% to 1% during the remaining period of the patient's life (Misch, 1999).

Therefore many techniques have been advocated for alveolar ridge augmentation after tooth extraction. Autologous bone grafting or implantation of phosphate- and calcium-based material are the current recommended solutions (Bianchi & Sanfilippo, 2001).

Autologous bone grafts are highly osteogenic and best fulfill the dental-grafting requirements of providing a scaffold for bone regeneration. However disadvantages such as the need for a second operative site, resultant patient morbidity, and the possibility of not being able to obtain a sufficient amount of material, led to the development of allografts and alloplasts as alternative or additional grafting materials (Hislop *et al.,* 1993).

NC has had considerable success considering that its porous structure is similar to cancellous bone and is one of the limited numbers of materials that has formed chemical bonding with bone and soft tissues (Kuhne *et al.,* 1994).

In this study natural coral processed by National Tissue Bank, Universiti Sains Malaysia (USM) was used for bone augmentation of post-extraction sockets. In addition PRP was extracted and mixed with NC to further accelerate wound healing and bone incorporation.

1.3. Hypothesis

1. The alveolar bone height of post-extraction dental sockets in the coral group was higher than the control group.
2. Combination of PRP with NC enhances bone formation therefore giving a higher alveolar bone height.

1.4. Objectives

1.4.1. General objectives

To study the use of natural coral and PRP in preservation of the post-extraction dento-alveolar defects.

1.4.2. Specific objectives

1. To determine the alveolar bone height before and after placement of graft material in natural coral group and natural coral with Platelet rich plasma group.
2. To determine the mean alveolar bone resorption in natural coral and natural coral and Platelet rich plasma and the control group.
3. To compare the alveolar bone resorption among the three groups.

1.5. Significance of the study

The results of this study will provide information on bone grafting with NC and application of Platelet-Rich Plasma. This information will aid clinicians to preserve the alveolar bone stock to facilitate the placement of implants and other dental prosthetic devices.

Chapter Two

Literature Review

Chapter Two

LITERATURE REVIEW

2.1 History of Bone grafting

Bone grafting has a long history and in legend goes far into antiquity. The first xenograft was possibly performed in Greek mythology when Demeter, the mother of Pelops, ate a portion of the young man's shoulder. To restore function, Zeus and the gods constructed a shoulder joint of ivory.

The twin physicians, St Cosmas and St Damian, lived in the third century AD and were put to death in 255 AD by an angered Emperor Diocletian (Danilevicius, 1967). In the fifth century, a faithful church retainer, exhausted by the pain from cancer of the limb, had it removed and implanted one from a Moor who had just died. This was the first allograft. Because the Moor had darker skin than the recipient, the event became known as "The Miracle of the Black Leg". The twins were beautified and their spectacular surgery was used as a subject numerous Renaissance artists (Figure 2.1).

Figure 2.1 St Cosmas and St Damian, transplanting a limb from a Moor. The amputated leg lies on the floor in the foreground. Fifteenth Century painting, Stuttgart, Germany.

In 1668, Job Van Meekeren, a Dutch surgeon, described the first bone graft procedure (Figure 2.2). Graft was taken from a dog's skull and used for the successful repair of a traumatic defect in a soldier's cranium but the patient was excommunicated for this very barbaric treatment. The surgeon was asked to remove the graft so that the patient could be returned to the good grace of the church but the graft was found to have taken and he could not remove it (Van Meekeren, 1668).

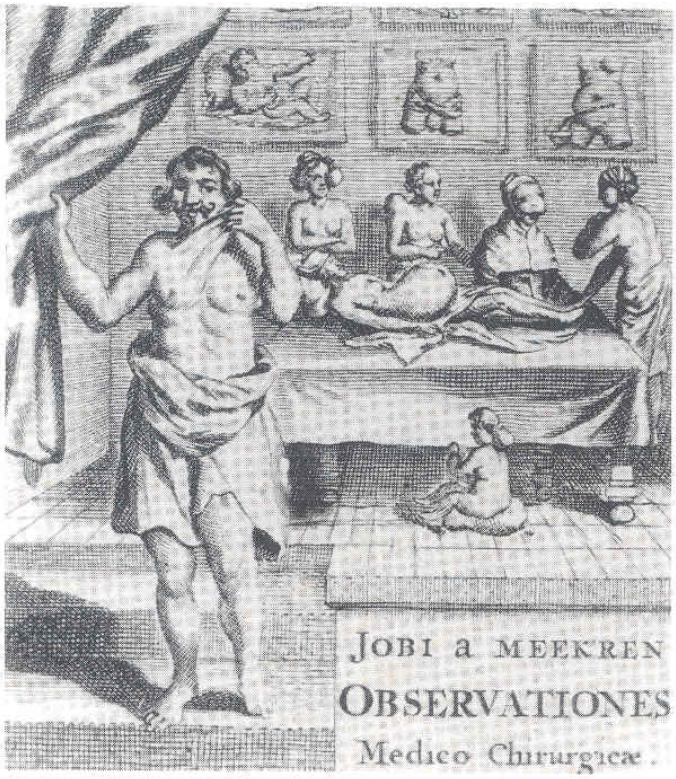

Figure 2.2 Job van Meekeren 1668, title page.

Anton van Leeuwenhoek, a Dutch scientist, was the first to describe bone structure in 1674. De Hyde and another Dutchman experimented on frog's legs and demonstrated his thought for the callus formation in blood clot around bone fractures (Van Leeuwenhoek, 1674).

Macween in Scotland 1880 performed the first allograft on a four-year-old boy, reconstructing an infected humerus with graft taken from the tibia of a child with rickets (Macween, 1881).

In 1867, the greatest surgeon Ollier, experimenting with rabbits and dogs in France, showed that autografts were viable surgical procedures. In this classic surgical work he recognized that separate living bone fragments within the periosteum could live and grow in a suitable environment (Ollier, 1867).

In 1893, Penski in Russia was the first to perform an allogeneic joint transplantation in animals (Aho 1973). Working independently in the same year as so often happens in medecine throughout the world. Barth in Germany and Curtis in America published work on bone transplantation. They described the absorption of dead tissue in bone graft and the formation of new bone that grew into the graft from the surrounding living bones. In 1914, Phemister described this process as “creeping substitution”.

Lexer described the use of joints in the reconstruction of traumatically injured knees in 1908. Seventeen years later he reported that half the patients still retained their grafts. Albee’s work in America on bone graft surgery was published in 1915, which resulted in an increase in bone transplantation.

In 1953, Urist in America developed the theory of osteo-induction. This suggested that a chemical mediator from the bone graft could induce bone formation by the recruitment of cells with potential for bone formation. All this work set the stage for establishment of reliable bone banks, especially in the USA, which provided a source of materials for bone grafting. Some clinicians around the world have reintroduced the concept of massive allografts as an orthotopic replacement for traumatically lost, tumor–ridden and diseased bones (Urist, 1953).

In London during the 1960's, Burwell possibly more than anyone, established the histological and immunological events that ultimately defined the natural history of allo-implantation. Campbell and others tested and compared allo-implant systems against autografts and defined the fate of bone and cartilage grafts in experimental animals (Campbell et al, 1953). All this work set the stage for establishment of reliable bone banks, especially in the USA, which provided a source of materials. Some clinicians in the world have reintroduced the concept of massive allografts as an orthotopic replacement for traumatically lost, tumor-ridden and diseased bones.

In the past 88 years, cadaveric allograft implantation has waxed and waned in popularity as a clinical activity. The concept of bone banking became a reality during and after World War II. It was used mostly for fractures and later fell into disfavor, probably on the basis of bad surgical experiences such as infection risk and poor prescriptions.

2.2 Bone and bone structure

Bone is a specialized and mineralized connective tissue, makes up, with cartilage, the skeletal system, which serves three main functions: A mechanical function as support and site of muscle attachment for locomotion; a protective function for vital organs and bone marrow; and finally a metabolic function as a reserve of calcium and phosphate used for the maintenance of serum homeostasis, which is essential to life.

There are two types of bone tissue: compact and spongy. The names imply that the two types differ in density or how tightly the tissue is packed together. Compact and spongy bones are made up of the same cells and the same matrix elements, but there are structural and functional differences.

The primary structural difference is quantitative: 80% to 90% of the volume of compact bone is calcified, whereas only 15% to 25% of the spongy bone volume is calcified (the remainder being occupied by bone marrow, blood vessels, and connective tissue). The result is that 70% to 85% of the interface with soft tissues is at the endosteal bone surface, including all spongy bone surfaces, leading to the functional difference: the cortical bone fulfills mainly a mechanical and protective function and the spongy bone mainly a metabolic function.

Compact bone consists of closely packed osteons or haversian systems. The osteon consists of a central canal called the osteonic haversian canal, which is surrounded by concentric rings lamellae of matrix. Between the rings of matrix, the bone cells osteocytes are located in spaces called lacunae. Small channels canaliculi radiate from the lacunae to the osteonic haversian canal to provide passageways through the hard matrix.

In compact bone, the haversian systems are packed tightly together to form what appears to be a solid mass. The osteonic canals contain blood vessels that are parallel to the long axis of the bone. These blood vessels interconnect, by way of perforating canals, with vessels on the surface of the bone (Figure 2.3).

Spongy cancellous bone is lighter and less dense than compact bone. Spongy bone consists of trabeculae and bars of bone adjacent to small, irregular cavities that contain red bone marrow. The canaliculi connect to the adjacent cavities, instead of a central haversian canal, to receive their blood supply. It may appear that the trabeculae are arranged in a haphazard blood supply. This is because they are organized to provide maximum strength similar to braces that are used to support a building. The trabeculae of spongy bone follow the lines of stress and can realign if the direction of stress changes. The same concept applies in the maxillofacial region whereby the forces of mastication are dissipated upwards towards the skull base of the maxilla and mandible (Roberts *et al*., 1987).

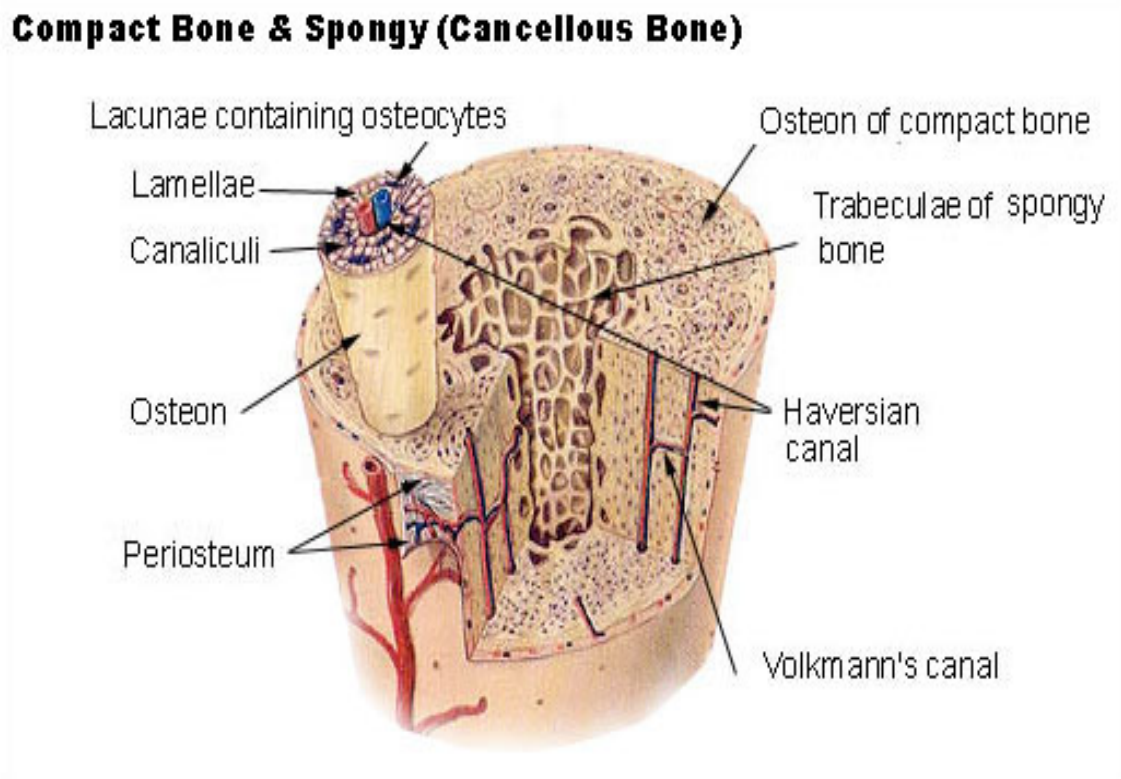

Figure 2.3 Microscopic structures of bone cells adapted from textbook, Bone diseases, by Claus-Peter Adler, 2000.

2.3 Bone contents

- Bone consists of 65% mineral (calcium + orthophosphate= hydroxyapatite)
- Organic matrix 25%
- Water 10%
- Collagen represents about 90% (dry weight) of the organic phase
- The remaining 10% consists of proteoglycans of small molecular size and non-collagenous proteins

2.4 Bone cells

Bone is composed of three cellular types: osteoblasts and bone lining cells, osteocytes and osteoclasts.

Osteoblasts are the bone lining cells responsible for the production of the bone matrix constituents, collagen and ground substance. Osteoblasts never appear or function individually but are always found in clusters of cuboidal cells along the bone surface (~100-400 cells per bone-forming site). At the light microscope level, the osteoblast is characterized morphologically by a round nucleus at the base of the cell (away from the bone surface), an intensely basophilic cytoplasm, and a prominent Golgi complex located between the nucleus and the apex of the cell.

Osteoblasts are always found lining the layer of bone matrix that they are producing, but before it is calcified (called, at this point, osteoid tissue). Osteoid tissue exists because of a time lag of approximately 10 days between matrix formation and its subsequent calcification. Behind the osteoblast can usually be found one or two layers of cells: activated mesenchymal cells and preosteoblasts, a mature osteoblast does not divide.

Osteoblasts are metabolically active secretory cells that express soluble signaling factors such as BMPs, TGF-β, insulin-like growth factors I and II, interleukin-1, PDGF that is crucial in the formation of osteoid. The active life span for human osteoblasts is believed to range from 1 to 10 weeks, after this period these cells may disappear. Some of these cells however become bone-lining cells and about 15% become osteocytes (Delmas & Malaval, 1993).

Osteocytes are found embedded deep within the bone in small lacunae because the calcified bone matrix is not metabolically inert. All osteocytes are derived from bone forming cells (osteoblasts), which have been trapped in the bone matrix that they produced and which became calcified. Even though the metabolic activity of the osteoblast decreases dramatically once it is fully encased in bone matrix, these cells still produce matrix proteins.

Osteocytes have numerous long cell processes rich in microfilaments, which are in contact with cell processes from other osteocytes, or with processes from the cells lining the bone surface osteoblasts or flat lining cells. These processes are organized during the formation of the matrix and before its calcification; they form a network of thin canaliculi permeating the entire bone matrix (Lynch *et al* 1999).