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Structural Aspects of Human Bone

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Besides genetic, biochemical and environmental factors, mechanical loading and deformations resulting thereof as well as eventual microdamage due to fatigue have been shown to exert a significant influence on growth, remodeling and healing of bone. In vivo imaging procedures, laboratory experiments and mathematical modeling of bone (micro-) mechanics are aimed at extending and deepening our knowledge with respect to largely unexplored aspects of bone physiology and skeletal disorders.

From a clinical point of view, the assessment of "bone quality" or the "competence of bone" is of primary interest whereby the fracture risk is mainly of concern. Skeletal structures mostly involved in fractures, in particular in osteoporotic patients of advanced age, are the spine, the femur and the wrist. Bone mineral content (BMC) and trabecular structure (characterized by a number of parameters) have been shown to be major determinants in view of fracture risk. While integral measurement methods such as DEXA or ultrasound enable an assessment of the state of overall mineralization (BMC), the determination of trabecular structure requires the application of special imaging procedures. Methods to investigate the microstructure of bone samples nondestructively include peripheral Quantitative Computed Tomography (pQCT, applied in vivo), furthermore micro CT (μCT, laboratory instrument), CT based on synchrotron radiation as well as micro-magnetic resonance imaging (μMRI). μCT allows furthermore the analysis of deformation patterns of trabecular bone samples which are subjected to controlled loading. The measurements are accompanied by large-scale finite element calculations in order to relate BMC and structure elements with mechanical properties. Such calculations can be extended today to include detailed models, e.g., of the human wrist.

Perfusion of bone is a further important aspect in bone mechanics, in particular, as bone as such is relatively impermeable for fluids. Instead, fluid flow in bone is mainly induced by deformations of the solid phase (forced convection), accordingly, the primary driving mechanism for fluids derives from the mechanical loading and deformation properties.

Key words: *Bone, remodeling, pQCT,μCT, bone analysis, bone perfusion*

1. Introduction

The skeletal system consisting of bones and joints serves two major purposes in the human body, viz.,

- to support the body, secure stability, provide protection and facilitate motion;
- to act as a reservoir for Ca^{++}.

This communication is devoted to a number of aspects relating to the structural properties of bone which are of importance with respect to its mechanical functionality and in particular to the gradual loss thereof which is frequently observed with increasing age or in case of disease.

The ability to fulfill the supporting and stabilizing function of the human body adequately, i.e., the capacity to withstand mechanical loads and enable motion, is often denoted as "competence of bone" or associated with the term "bone quality" (Cooper 1993, Dequeker 1994, Mosekilde 1994, Ulrich et al. 1997, Majumdar & Bay 2001, Madsen et al. 2002, Müller and van Lenthe 2004). Typical incidents which are observed in case of insufficient competence or bone quality are a fracture of the femoral head, a fracture of the wrist or a collapse of a vertebra (Fig. 1). In the case shown, a thoracic vertebra collapsed due to osteoporosis causing pain and spinal instability. As can be seen from the dates of the X-ray projections, such a collapse is a rather rapid, sudden process and does not necessarily extend over long periods of time; this is in contrast to degradation associated with loss of competence of the skeleton which may progress over years (Kahn 1995).

A distinction is often made in biomechanics between "soft" and "hard" tissues. While there are numerous kinds of soft tissues, hard tissue appears essentially in the form of calcified tissue, in particular bone. Thereby, the calcium is contained in hydroxyapatite crystals $[Ca_5(PO_4)_3OH]$ which are embedded in a collageneous matrix. Aside from the integrity and mechanical loading capacity of bones, a physiologic calcium balance is eminently important for the overall homeostasis of the human body in that calcium is

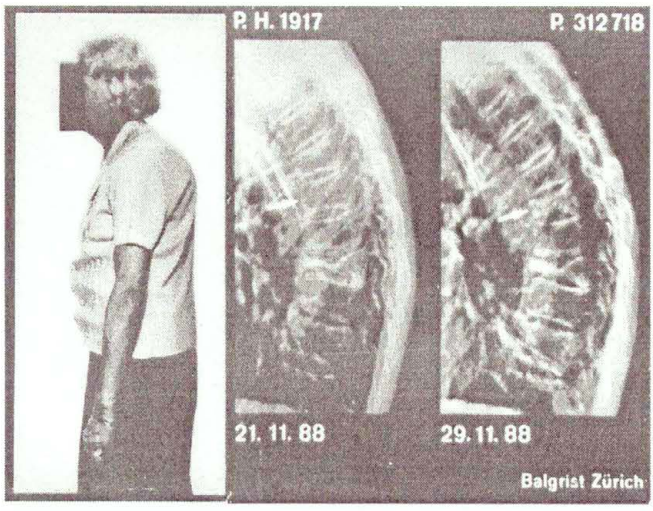

FIGURE 1. Collapse of cervical spine vertebra (arrow) due to osteoporosis in 71 year old woman (note the dates of the X-ray examinations)

essential for many physiologic processes, among them, the action of muscles, the transmission of nerve signals or the coagulation of blood. As such, calcium is by far the most abundant bone mineral ("calcium reservoir"), others, e.g., phosphor, being much less concentrated. Therefore, the terms "calcification" and "mineralization" of bone are often used synonymously.

Bone exists in various forms: cortical bone makes up the shaft (diaphysis) of the long bones as well as the outer layer of other bones while trabecular or cancellous bone is located mostly in the medullary canal of long bones, particularly in regions close to joints (epiphysis and metaphysis) as well as in the spine and in bones whose primary task is not to support loads (e.g., skull, iliac crest). The metaphysis denotes the zone of growth between the dia- and epiphysis of young bones. The terms Haversian bone and woven bone, in turn, refer to the internal structure of the bone material. Haversian bone consists of cylindrically shaped osteons which along their longitudinal axis exhibit a canal (Haversian canal), whereas woven bone grows during growth and healing processes. For a more detailed description of bone, the reader is referred to the literature (e.g., Bilezikian et al. 2002).

The human skeleton is subjected to a constant turnover or remodeling in that up to 25% of the hard tissue (bone) is replaced each year. It is the combined action of osteoclasts (multinucleated cells performing bone resorption), osteoblasts (cells producing new bone material, i.e., bone apposition) and

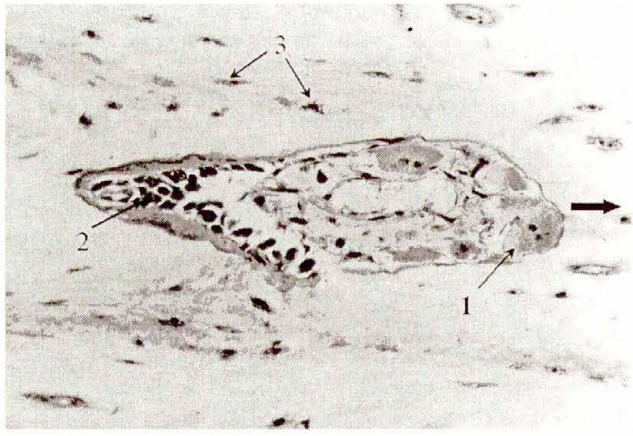

FIGURE 2. Osteclasts (multinucleated cells, 1), osteoblasts (2), osteocytes (3). The arrow indicates the direction of resorption and apposition (Courtesy: St. Perren, MD, AO-ASIF, Davos, Switzerland).

osteocytes (mature osteoblasts which are embedded in the bony surrounding) that causes and controls bone remodeling, cf. Fig. 2. The osteocytes are housed in small cavities within the bone material, called lacunae, and they are connected to one another through an extensive network of tiny channels (canaliculi) through which osteocytic processes extend such that each osteocyte has numerous connections with adjacent osteocytes. It is believed that the osteocytes communicate by way of their processes and thereby direct osteoclast and osteoblast activity (Johnson & Highison 1983, Moss 1991, Cowin et al. 1991, Aarden et al. 1994).

An unbalance between bone resorption and apposition in the course of the natural remodeling process may lead to pathologic conditions. In particular, excessive bone resorption or demineralization may cause osteoporosis (NIH 2000) which in turn may compromise the competence of bone (a certain amount of slow demineralization after the age of 35–45 years, especially in postmenopausal women has however been found in large population statistics and may be physiologic). Besides osteoporosis, a number of other pathologies are associated with mineral loss and loss of competence.

According to a general paradigm, demineralization of the skeleton manifests itself more rapidly in trabecular than in cortical bone. The reason for this derives from the way that bone is remodeled: osteoclasts act at the surface of bony structures and not within intact bone material, and the surface of trabecular bone structures is much greater than that of cortical sections.

Also bone healing is a surface-driven process. This is in contrast to soft tissues, where remodeling, if it occurs at all, is observed throughout the tissue.

A comprehensive understanding of the processes including their mutual interaction that govern bone remodeling is not available to date. Nevertheless, a number of factors have been identified that exert an influence on the activity of the various bone cells and the state of mineralization or calcification, mainly,

- genetic, biochemical, immunological and endocrine factors,
- molecular and cell biology related factors,
- mechanical loading (or, more specifically, a lack thereof),
- bone perfusion,
- diseases.

In the following, bone mechanics and bone perfusion are considered in more detail along with selected pathologies. Thereby, we will restrict ourselves to mechanical and structural aspects.

From a clinical point of view, however, the competence of bone as introduced above is of primary concern and bone remodeling should be viewed mainly under this aspect. The question therefore arises with respect to the relation between bone remodeling and the competence of bone as well as with respect to the assessment of these processes and properties under *in vivo* conditions. Two determinants have been identified in the past which are of significance with respect to the ability of bone to support mechanical loads, viz.,

- bone mineral content (BMC) and
- structure of the cortical/trabecular bone architecture.

It is important to note at this point that BMC alone is insufficient to assess the competence of bone, although this is often attempted under clinical constraints (time, cost) (Melton et al. 1989, Parfitt 1992, Ahmed et al. 1997, Rüegsegger 1996). The reason is that it is much easier to determine BMC than the often complex architectural characteristics of the skeleton. Nevertheless, several studies have shown that bone microarchitecture strongly influences its stiffness and strength, and predictions of trabecular bone competence can greatly be improved by including architectural parameters in the analysis (Turner et al. 1990, van Rietbergen et al. 1998b, Yang et al. 1998, Müller et al. 2004).

Various methods which will be discussed in the next paragraph have been devised to quantify the determinants mentioned above. With all such methods, however, extrapolations based on mathematical modeling or estimations are necessary with respect to the competence of the skeleton which is ultimately decisive. Yet, the procedures which lend themselves for this purpose are not at all straightforward, because "competence of bone" or "bone quality" are rather qualitative characterizations and defy a direct measurement.

2. Measurement

The most important methods which are of use for the investigation of the geometrical and mechanical properties of bone are:

- X-ray, in particular computed tomography (CT),
- magnetic resonance imaging (MRI),
- ultrasound (US),
- atomic force microscopy (AFM),
- nanoindentation measurements,
- compression and tensile tests.

Due to the fact that the relatively heavy element calcium is abundant in bone material (soft tissues do not contain large amounts of any heavy elements), X-ray, in particular X-ray Computed Tomography (CT) is the method of choice to image and quantify bone. Procedures and instruments for CT extend from whole-body down to microscale. While whole-body CT has insufficient resolution for a quantitative analysis of trabecular structures, specialized small scanners have been developed that enable the scanning of bony architectures, first *in vitro* (Feldkamp et al. 1989), later also *in vivo* (Müller et al. 1994, Rüegsegger et al. 1996). Thereby, human extremities, or, as the term peripheral Quantitative CT or pQCT suggests, appendicular segments of the human anatomy (arms, legs) are scanned quantitatively with a spatial resolution better than 100 μm, high accuracy and minimal radiation dose. pQCT allows to quantify BMC as well as to image the intrinsic structures in peripheral bones (ulna/radius and tibia/fibula) in great detail (Fig. 3). Regions close to joints (wrist and knee, respectively) are thereby especially interesting because of the trabecular bone which is mostly located in these areas. A drawback of this method derives however from the mechanical constraints

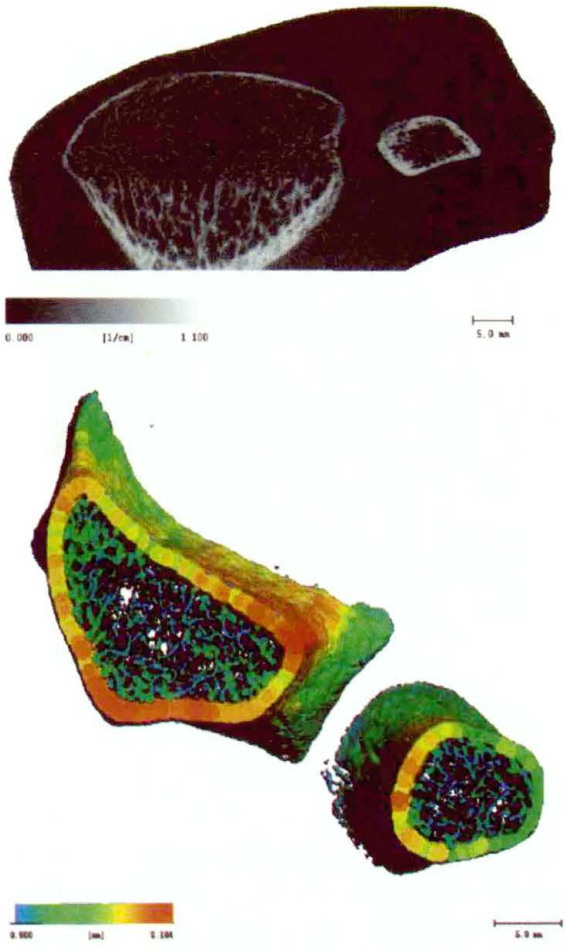

FIGURE 3. pQCT of human wrist (Courtesy: Scanco AG, Bassersdorf, Switzerland).

associated with high resolution, i.e., the opening of the scanner along with the scanned area does not allow to perform whole-body scans. The radiation dose furthermore increases with the fourth order of the resolution (Barrett & Swindell 1981) such that high-resolution whole-body scans for screening purposes could not be justified. Accordingly, particularly interesting objects such as the spine or the femoral heads cannot be imaged with these instruments. Yet, pQCT has been evaluated and has established itself as a reliable clinical means to determine BMC along with the structure of the bone architecture (Neff et al. 2000). Even smaller instruments, μCT, are used as a laboratory

method to image small bone samples at high resolution reaching 10 μm typically. The highest resolution is however obtained with synchrotron CT, where the advantage of the monochromaticity and parallel beam characteristics of synchrotron radiation enable resolution below 1 μm (Fig. 4) (Stampanoni et al. 2002, Stampanoni et al. 2003).

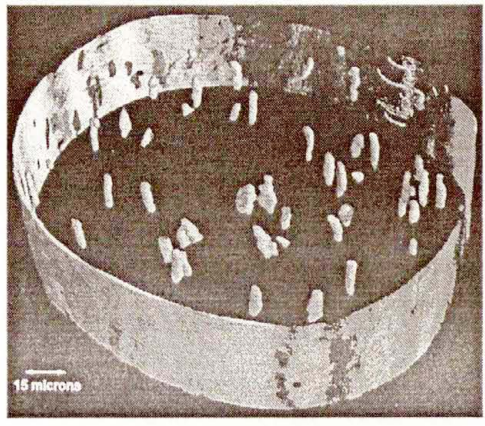

FIGURE 4. Edge-enhanced synchrotron CT of human trabecula exhibiting lacunae (Courtesy: Dr. M. Stampanoni, PSI, Switzerland).

Dual energy X-ray absorptiometry (DEXA) (Mazess et al. 1991, Prince et al. 2001, Kirk et al. 2002), in turn, can be applied to all bones of the body, in particular to the spine. The measurement is restricted to BMC, however, and architectural aspects remain in essence unresolved.

Magnetic resonance imaging lends itself to be used for the analysis of the morphology of bone (Majumdar et al. 1996, Hipp et al. 1997, Wehrli et al. 1998) in that the bony tissue as such yields no detectable signal and appears therefore as "void". The method has the advantage of not only being noninvasive but also nonionizing. Yet, it is expensive and BMC cannot be determined directly.

Ultrasound densitometry (Mazess et al. 1992, Langton and Langton 2000, Wunsche et al. 2000, Adler et al. 2001, Nicholson et al. 2001) offers the advantage that mechanical properties are measured directly (speed of sound, attenuation), but the applicability and accuracy are limited (mostly calcaneus), and the biological variability is relatively high. Even as a screening method, the usefulness of ultrasound has been questioned (Sim et al. 2000). AFM and nanoindentation measurements, in turn, act on histologically prepared bone surfaces. Thereby, AFM is a method to image the surface topography

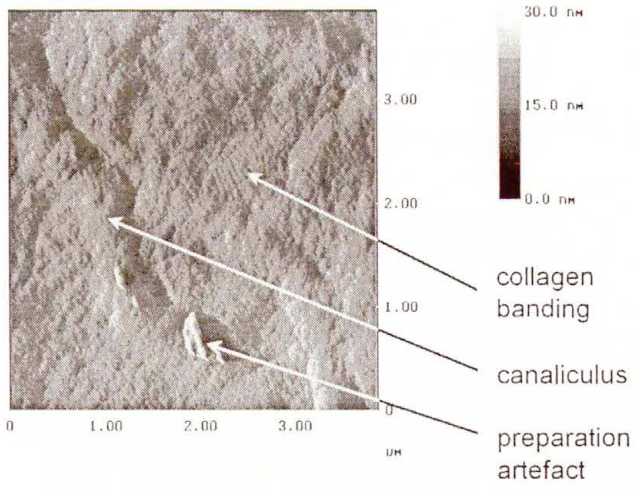

FIGURE 5. AFM image of cast of bone surface showing a branching canaliculus (preparation technique according to Knapp et al., 2002). The diameter is around 400 nm. The periodicity of the partially visible pattern corresponds to the periodicity of the collagen molecule.

(Fig. 5) (Reilly et al. 2001, Knapp et al. 2002), whereas, by nanoindentation, local deformation characteristics can be determined and related to mechanical parameters of the bony tissue (Hengsberger et al. 2002, Hengsberger et al. 2003). Compression and tensile tests, finally, can be performed on excised whole bones or bone samples of various sizes (Ciarelli et al. 1991, Zhu et al. 1994, Keaveny 1997).

While BMC can be determined from X-ray absorption and expressed as a numerical value, structural properties of trabecular bone architecture cannot be quantified in a straightforward fashion. A number of indices have been defined to this end, among them, densitometric parameters such as trabecular bone density (Dtrab), or metric parameters, in particular trabecular number (Tb.N), trabecular thickness (Tb.Th), cortical thickness (Ct.Th) or bone volume density (BV/TV) (Parfitt et al. 1983, Müller et al. 1996). In addition, geometrical characteristics relating directly to the intrinsic shape of the trabecular network, *i.e.*, how rod- or plate-like the structures are, can be quantified by the determination of non-metric parameters such as the structure-model-index, SMI (Hildebrand and Rüegsegger 1997), the trabecular bone pattern factor (TBPf) (Hahn et al. 1992) or bone connectivity (Odgaard and Gundersen 1993). A general morphometric analysis of trabecular bone has been performed by Hildebrand et al. (1999).

In order to be able to determine the progress of remodeling processes, in particular as they relate to osteoporosis, or the effectiveness of therapeutic procedures to control the mineralization of bone within clinically useful periods of time (typically 6 months), a reproducibility of at least 0.3% of typical quantities which are measured such as BMC is necessary. To the overall reproducibility, two effects contribute, namely the absolute accuracy of a single measurement as well as the precision of the repositioning of the measured section of bone from measurement to measurement which are a few months apart. At this time, only specially developed methods based on pQCT allow to perform measurements which meet the desired limits with respect to accuracy and precision (Rüegsegger & Laib 2000).

3. Geometrical Aspects of Bone Remodeling

As mentioned in the introduction, according to a general rule, trabecular bone is lost faster than cortical bone in case of skeletal demineralization. Thanks to pQCT such processes can be scrutinized with sufficient accuracy to demonstrate that this rule might have to be revised. Laib et al. (1998) found in a systematic analysis of eighteen postmenopausal women measured over one year that four different groups could be discerned (Fig 6.),

group 1: "Fast loosers", i.e., women with whom both cortical and trabecular parameters decrease rapidly

group 2: Normal age-related demineralization, primarily affecting trabecular bone

group 3: Calcium loss primarily in the cortical bone while the trabecular structure remains intact

group 4: No significant loss

There is no straightforward explanation at this time for the remarkable finding that in certain cases cortical bone is lost primarily rather than trabecular one. Further investigations are necessary which include in particular genetic, environmental and endocrine factors in order to arrive at a deeper understanding of such processes.

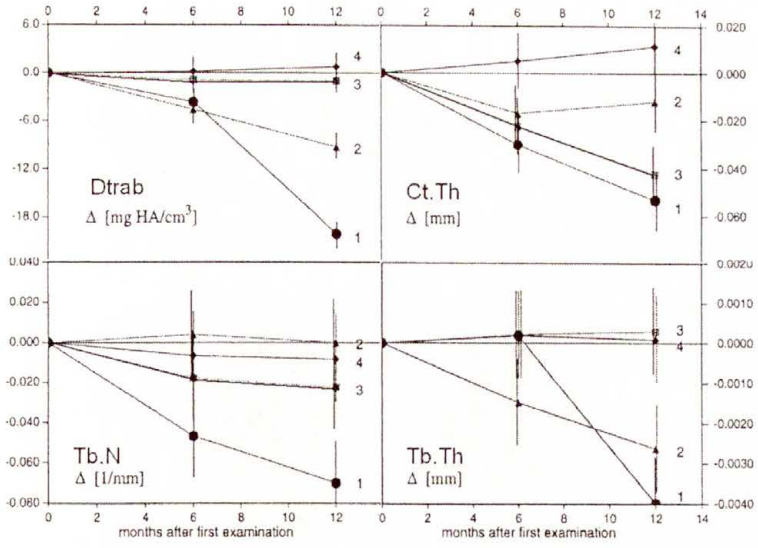

FIGURE 6. Trabecular density (Dtrab), cortical thickness (Ct.Th), trabecular number (Tb.N) and trabecular thickness (Tb.Th) as function of time in 18 postmenopausal women. Groups 1-4 see text.

In cases of disease, the situation becomes even more complex (Figs. 7a-e).

Figure 7a: Cross section exhibiting normal calcification of cortical shell (a somewhat reduced cortical thickness at those locations where radius and ulna are close is normal) and of trabecular region

Figure 7b: Type 1 osteoporosis (age-related osteoporosis, primarily affecting trabecular bone)

Figure 7c: Developmental disorder (irregular development of cortical shell, evidence of earlier stages, irregular calcification of trabecular section)

Figure 7d: Algodystrophy (highly irregular trabecular region, probably due to insufficient and inhomogeneous blood supply)

Figure 7e: Osteogenesis imperfecta and fracture loco classico (the mineralization is strong at the location of an earlier fracture which leads to an overall high BMC, but the bone quality is not necessarily adequate)

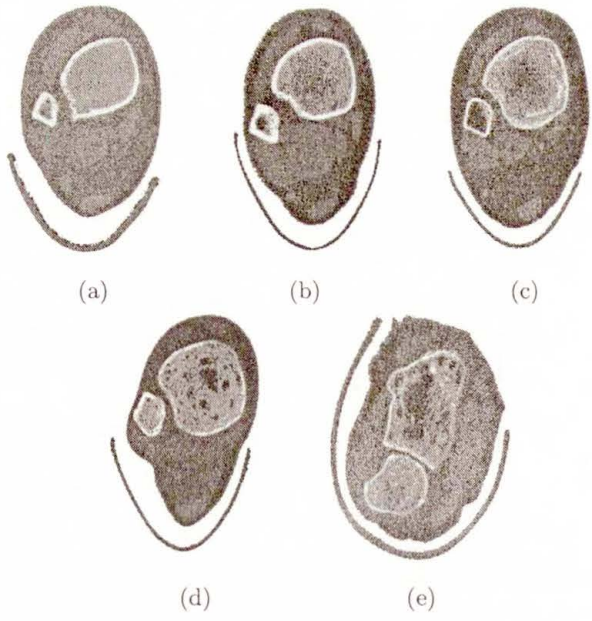

FIGURE 7. Typical cross-sections (radius-ulna), obtained with pQCT, normal and various disorders, see text.

4. Micromechanical Aspects

Trabecular bone sections exhibit various appearances (Fig. 8). The Structure Model Index (SMI) as introduced earlier serves as a quantity which can be used to classify trabecular structures according to their internal architecture. In particular, rod-like (SMI = 3) and plate-like (SMI = 0) structures can be discerned, a classification which can then be related to other geometrical quantities. In Fig. 9 the dependence of bone volume density (BV/TV) on SMI is shown as an example (Hildebrand 1997).

Because of its structure and geometry, bone is anisotropic. Both the shell-like architecture which is due to the formation of osteons in Haversian bone as well as the arrangement of the trabeculae in cancellous bone cause anisotropy. The fabric tensor (Cowin 1985) is thereby often used to relate geometrical and mechanical anisotropy (Turner et al. 1990, Zysset & Curnier 1995, Odgaard et al. 1997, Kabel et al. 1999).

Systematic measurements of the behavior of bone biopsies under mechanical loading were made by Müller et al. (1998) (Fig. 10). To this end, a method combining a mechanical deformation device and μCT was developed.

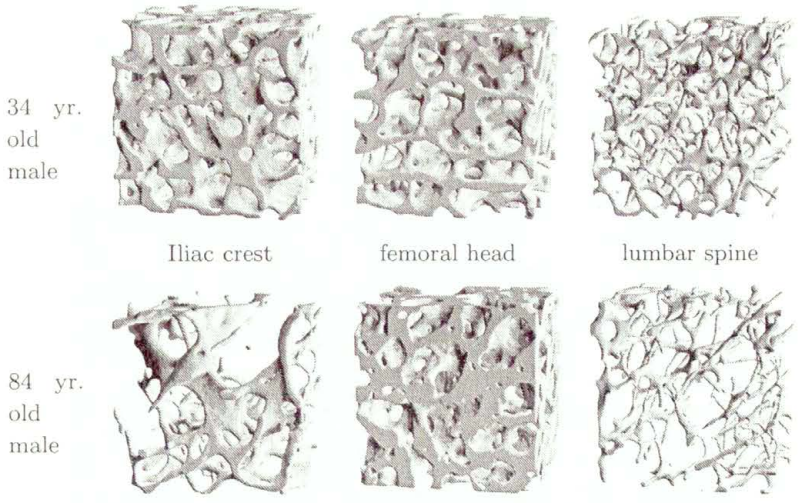

FIGURE 8. Typical trabecular bone structures at various anatomical sites. Measurements have been performed with a micro-tomographic imaging system providing 14 μm nominal resolution.

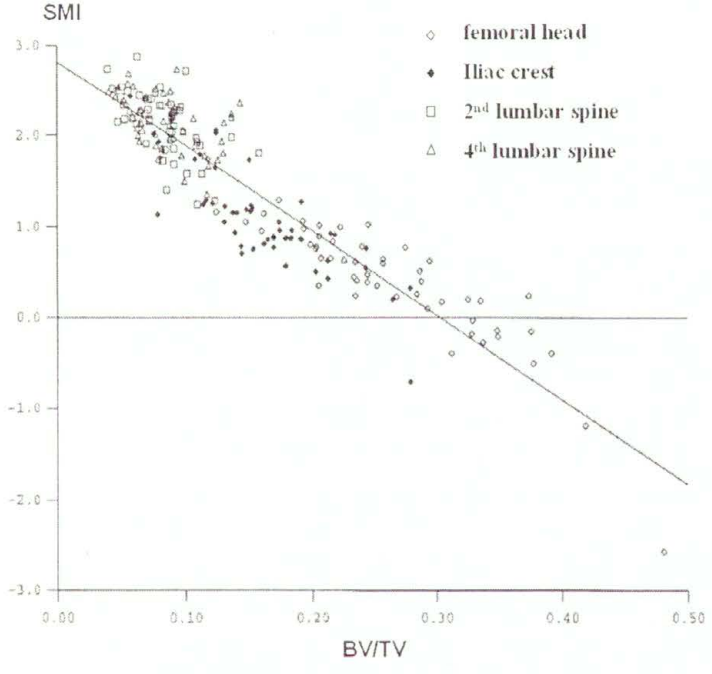

FIGURE 9. Correlation Structure Model Index (SMI) – Bone Volume/Total Volume (BV/TV, bone volume density)

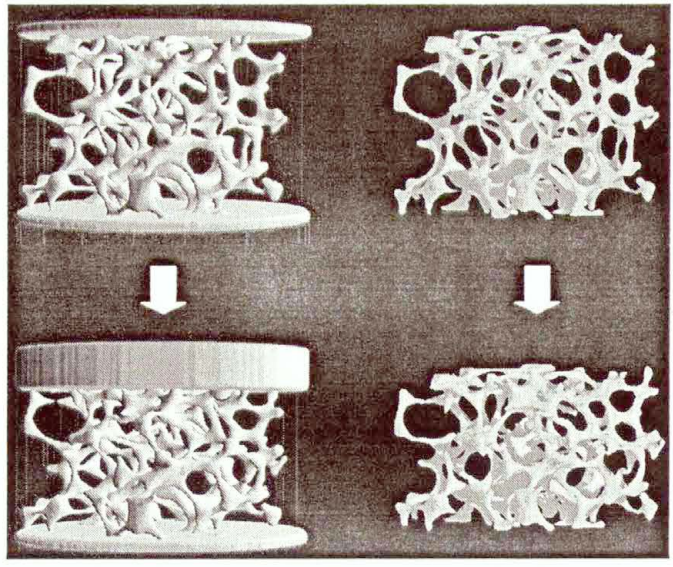

FIGURE 10. Compression of trabecular foam sample. Left: μCT recording during compression. Right: FE model replicating experimental compression sequence (FE images courtesy Dr. Mark Taylor, University of Southampton).

Figure 10 (left) shows a bone biopsy at the beginning (top) and end (bottom) of a deformation sequence. On the basis of a Finite Element (FE) model (Alonso Vázquez et al. 2004) which replicates the deformation process iteratively, the mechanical properties of the material can be recovered (Fig. 10 right).

At a larger scale, pQCT is used to image skeletal regions which are close to the wrist (Fig. 3). Since the bony material *per se* keeps in essence its mechanical properties also under most conditions of atrophy (Kahn 1995), in particular osteoporosis, a FE model of the reconstructed section of bone can be made utilizing average values for the mechanical behavior of bone material. In order to obtain a valid geometrical representation of the section to be modeled, the image stacks obtained from pQCT have to be segmented to yield a true 3D replica. This is not a straightforward procedure since the spatial resolution of pQCT is only slightly better than typical trabecular dimensions and carefully designed and validated algorithms have to be applied for this purpose (Müller et al. 1994, Oestreicher & Rüegsegger 2000).

As a consequence of the geometrical complexity of bony structures, in particular of cancellous bone, FE meshes have to be generated that exhibit a high degree of freedom if the morphology is modeled with a resolution matching

the one of the scanner. Large scale FE methods have been developed for this purpose (van Rietbergen et al. 1995, van Rietbergen et al. 1998a). Special algorithms are thereby necessary to deal with the high number of degrees of freedom (van Rietbergen et al. 1996, Ulrich et al. 1998b).

Whenever FE models are developed and applied, boundary conditions are critical. If the section under consideration is sufficiently large, the influence of the details of the assumed boundary conditions fade with increasing distance from the boundary (St. Venant's principle), and reliable results are obtained providing that the overall loading is correct. Such a loading can, e.g., be derived from a fall on the hand (Ulrich et al. 1998, Pistoia et al. 2002) which is a type of accident leading to Colles or Smith fractures which are frequently observed in the elderly population (Fig. 11).

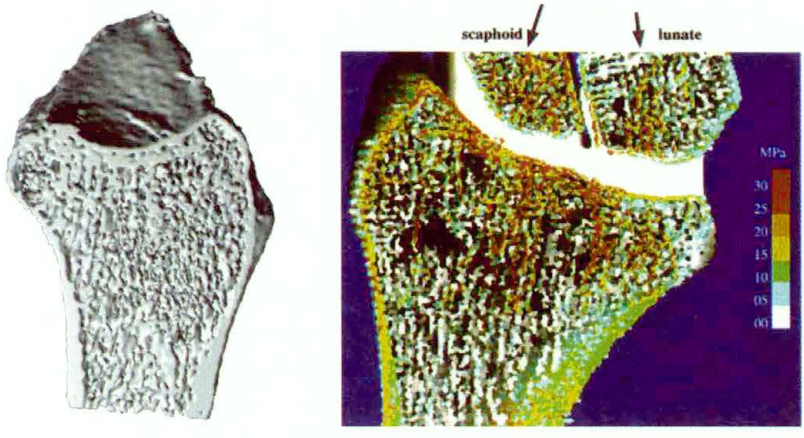

FIGURE 11. FE model of the human wrist (right), derived from a pQCT measurement (left). The loading consists of two forces, viz., 600 N on the scaphoid and 400 N on the lunate, simulating a fall on the hand. The colors (right) indicate the von Mises stresses.

The important aspect in the described procedure combining pQCT and FE analysis is however that this is the only valid method available to date in a clinical setting which allows to relate loading conditions and structural behavior of bone, *i.e.*, to assess the competence of bone (Ulrich et al. 1997, Ulrich et al. 1998a). Accordingly, great emphasis is given to the further development of methods which facilitate the measurement (by means of pQCT) and modeling (large scale FE methods) of increasingly larger bony sections which are close to joints (see, e.g., ADOQ, Advanced Detection of Bone Quality, European Union project, `www.medes.fr/adoq`).

5. Perfusion of Bone

Mechanical stress has long been recognized as an important stimulus within the framework of bone remodeling, in particular, lack of mechanical loading has been found to be a major factor causing remodeling of bone (Roux 1881, Wolff 1892, Frost 1964, Uhthoff & Jaworski 1978, Lanyon et al. 1982, Lanyon 1984, Rubin & Lanyon 1987). A number of hypotheses have been put forward in order to understand the mechanotransductive processes acting on a cellular level which underlie this fact (for an overview over mechanosensation and mechanotransduction in bone, see Cowin 2003). Among these hypotheses, bone perfusion or bone fluid flow has been identified as an important determinant within the framework of bone remodeling, and for the reasons outlined in the following, the structure and mechanical properties of bone are of importance with respect to bone perfusion.

The solid bony matrix as such is essentially impermeable for fluids. Yet, it is pervaded by the lacuno-canalicular system (Junqueira et al. 1995) (for the various degrees of porosity in bone, see Knothe Tate 2003). In Haversian bone, furthermore, bone appears in the form of cylindrical osteons which are organized in a shell-like fashion. A capillary runs in a longitudinal direction along the central canal, from which a dense network of canaliculi originates. The canaliculi connect the lacunae which are typically spaced at a distance of 10 μm – 30 μm from one another and which contain the osteocytes. These cells, as mentioned in the introduction, communicate by way of numerous processes which extend through the canaliculi such that they act as a functional syncytium (Johnson and Highison 1983, Moss 1991, Cowin et al. 1991, Aarden et al. 1994). The contacts are thereby provided by gap junctions (Doty 1981, Civitelli 1995, Lecanda et al. 1998). As most canaliculi have a diameter of less than about 500 nm (Fig. 5) and are in addition filled with the osteocytic processes there is minimal space for the metabolic traffic of the osteocytes although the number of canaliculi per lacuna is large. In particular, it has been established that diffusion alone is by far insufficient to fulfill the metabolic needs of the osteocytes (Bassett 1968, Piekarski & Munro 1977). Instead, the deformation that the bones undergo under the influence of daily activities cause pressure gradients to be built up in the fluid phase which drive the fluid through the canaliculi (load-induced fluid flow or forced convection) (Johnson et al. 1982, Kufahl & Saha 1990, Gatzka et al. 1999, Knothe Tate et al. 2000). The effect of this forced flow is mainly threefold, viz.,

- transport of nutrients and waste products,
- viscous shear loads (Weinbaum et al. 1994) and mechanical drag (You et al. 2001) acting on cellular membranes,
- streaming potentials (Pollack & Petrov 1984, Salzstein & Pollack 1987, Otter et al. 1992).

All of these effects have been brought in connection with bone remodeling. In particular, lack of mechanical loading evidently leads to a decrease in bone perfusion thereby causing, among other, cellular starvation, which in turn induces bone resorption.

The transport of substances covering a wide range of molecular weights and physico-chemical properties has been investigated extensively (see the table given in Knothe Tate 2003). Furthermore, the transport efficiency under various kinds of loading conditions has been studied as well as in the presence of fatigue-induced microfractures which is of importance with respect to bone healing processes (Tami et al. 2002).

Deformations which the long bones in the leg undergo during walking, *e.g.*, are of the order of magnitude of 500–2000 μstrain at the surface, where deformations are maximal. This has been verified, among other, in measurements made in the metacarpus of sheep (Steck et al. 2003a). These bones consist of Haversian bone which is considered as representative for human conditions.

Experiments using various tracer substances along with mathematical models demonstrated the efficiency of load-induced bone fluid flow (Knothe Tate et al. 2000). In Fig. 12 microscopic sections of the two tibiae of a rat are shown. In this experiment, the (anaesthetized) animal was perfused systemically during 5 minutes prior to the experiment with procion red (a fluorescent marker exhibiting no metabolic activity with a molecular weight around 200). While one of the two tibiae was then loaded mechanically, the contralateral side remained unloaded. The loading consisted of ten cycles of four-point bending (65 N) which leads to about 2000 μstrain (max.) at the surface of the bone. A FE model (Figs. 13, 14) confirmed the experimental findings qualitatively and demonstrated the influence of the various mechanical parameters such as viscoelasticity of the bone material on the efficiency of load-induced fluid flow (Steck et al. 2003b). This model was thereby based on a continuum mechanics' approach using Biot's theory of poroelasticity (Biot 1941) in combination with mass transport equations.

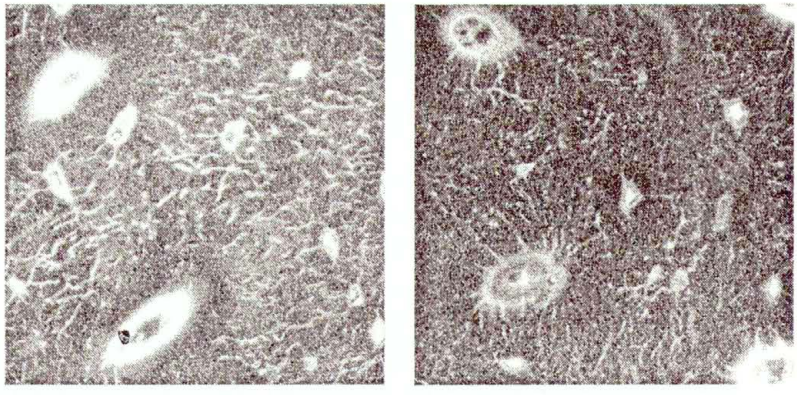

FIGURE 12. Micrograph of rat tibiae demonstrating the effect of load-induced fluid flow (forced convection). As fluorescing tracer substance, procion red was systemically administered. The loading consisted of cyclic four-point bending (see Fig. 13).

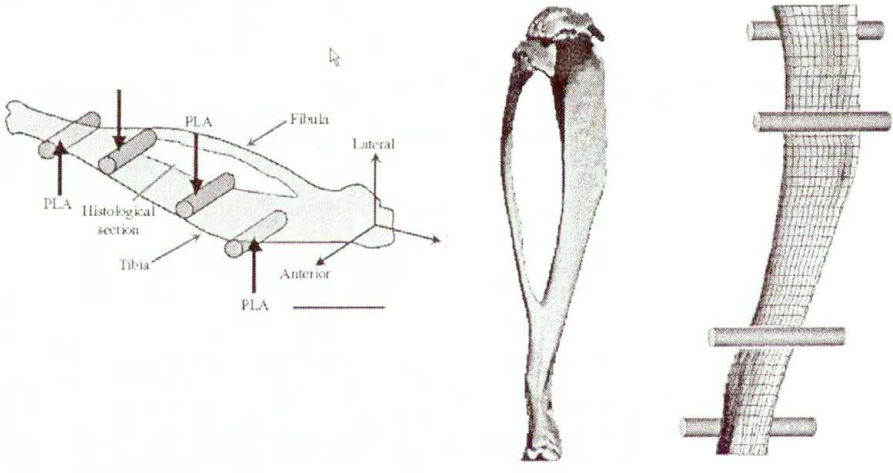

FIGURE 13. FE model of a rat tibia (right), derived from a μCT measurement (middle). The loading consisted of four-point bending as indicated left. Cyclic forces had an amplitude of 65 N. The histological section was taken as indicated (left), likewise, the results shown in Fig. 14 correspond to this section.

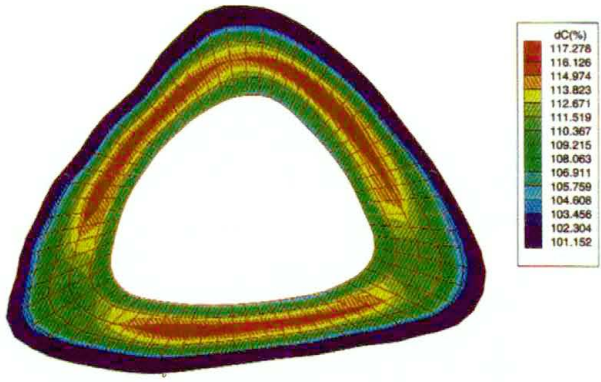

FIGURE 14. Results of FE calculation: Difference of the simulated tracer concentration between the loaded and unloaded rat tibia in percent after 10 load cycles.

6. Future Developments and Conclusions

This communication is limited to an outline of the measurement methods and the mathematical analysis of geometrical and structural properties of bone including bone perfusion. Genetic, environmental, endocrine and immunological factors as well as factors relating to molecular and cell biology have furthermore to be included in order to arrive at a more comprehensive understanding of bone remodeling. Yet, we are far away at this time from a unified and thorough concept of bone biology including all facets which would allow to predict the control and dynamics of bone remodeling, furthermore, interpret and understand disease processes. Likewise, therapeutic procedures are often based on a trial and error basis unless a clear endocrine or metabolic cause is diagnosed (e.g., calcium resorption deficit or specific endocrine disorder). The situation becomes even more complex if joints and their diseases are taken into account since this involves also cartilage, a tissue whose mechano-biological properties are likewise only partially understood. In addition, the cartilage-bone interface poses particular problems and exhibits characteristics which are important in view of diseases such as rheumatism or osteoarthritis.

In the future, higher resolution CT methods along with larger FE models than are available to date will enable the determination of mechanical conditions in bone with increasingly more detail and at the same time at an increasingly smaller scale. Of particular importance, thereby, is the pos-

sibility to make measurements in humans under *in vivo* conditions. From this we expect progress in several directions, likewise of a macroscopic and microscopic nature.

First, the competence of gradually larger skeletal sections within the human body can be assessed with increasing reliability.

Second, by including fluid dynamics, bone fluid flow and material transport can be modeled more realistically. A detailed knowledge of the micromechanical environment of bone cells will enable an analysis of local solid and fluid mechanics' influences on cellular membranes, membrane proteins and transport mechanisms along with processes down to the level of gene expression such that mechano-sensation and mechano-transduction will no longer be a conundrum.

Third, by formulating mathematical models which describe metabolic processes quantitatively, bone biology will become amenable to a comprehensive engineering analysis.

Along with animal models as well as with experiments involving cell cultures, all of these methods are expected to bridge the gap from the engineering side between bone biomechanics and the biological basis of bone physiology and pathology.

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