SPEAKER NOTES
PhD Training Course 15-18 September 2013, Hamburg

Geert Carmeliet (Leuven, Belgium)

Biography

Geert Carmeliet is Professor in Medicine and head of the Laboratory of Clinical and Experimental Endocrinology in the department of Clinical and Experimental Medicine at the KU Leuven, Belgium. She obtained her MD, Board Certification in Paediatrics and PhD at the KU Leuven. Dr Carmeliet's research studies the role of angiogenic factors, including VEGF and Placental Growth Factor, in bone development using several genetic mouse models. Recent studies address the contribution of oxygen sensors in bone metabolism. Findings are translated in preclinical models of bone metastases, fracture repair and tissue engineering. The therapeutic use of molecules targeting these pathways to improve vascularisation, cell survival and bone formation in tissue engineered constructs is evaluated. Her laboratory also studies several aspects related to calcium homeostasis and calcium signalling. The tissue-specific effects of vitamin D action with respect to calcium and bone homeostasis are investigated. The role of calcium transporters in the intestine and in bone cells is studied, linking extracellular calcium homeostasis with intracellular calcium signalling.

Hypoxia and Bone Development – Geert Carmeliet

Abstract

Adequate oxygen supply is critical for cellular function and survival. Because of their dependence on oxygen, aerobic organisms have developed mechanisms to sense oxygen availability. When oxygen supply is insufficient, cells can activate a number of adaptive responses to match oxygen delivery with metabolic, energetic and redox demands. The hypoxia-inducible factor (HIF) is recognized as a key modulator of the transcriptional response to hypoxia. Under hypoxic conditions, the HIF□ subunit heterodimerizes with the stable HIF□ subunit and enhances transcription of numerous genes, leading to alterations in oxygen delivery and cellular metabolism. In a well oxygenated environment, HIF□ subunits are hydroxylated by prolyl hydroxylase domain (PHD) proteins. This modification targets HIF□ for proteasomal degradation. The PHDs use oxygen as a direct substrate and the oxygen availability thus regulates the enzymatic activity of the PHDs. Because of this dependence on oxygen, PHDs have been proposed to be 'oxygen sensors'

The important roles of HIF1 and HIF2 in the regulation of skeletal development, bone homeostasis and bone repair have been identified using genetically altered mice. Part of the effects of HIF are attributed to the increased expression of vascular endothelial growth factor, VEGF, a potent angiogenic factor that ensures the timely and adequate vascularization during bone development and bone repair. HIF is in addition a survival factor for chondrocytes, likely by regulating adaptations in cell metabolism. Adequate levels of HIF are thus vital to permit bone cells to respond accurately to hypoxia. Whether increased levels of HIF in normoxia are beneficial seems to depend on the bone cell type. An overview of recent findings on the role of hypoxia signalling in bone development and bone repair using transgenic mouse models will be given.

Simone Cenci (Milan, Italy)

Biography

Born: 16 November 1971, married, one daughter.

1995: M.D. degree

2000: Specialization, Geriatrics and Gerontology

1998-2003: Research Associate, Dept. Of Internal Medicine, Division of Bone and Mineral Diseases, Washington University School of Medicine, St. Louis, MO, USA. Here, Dr Cenci discovered novel immune mechanisms of osteoporosis (in 6 first-authored papers, including *PNAS*, *Blood* and 2 *J Clin Invest)*, and earned 2 Young Investigator Awards from the American Society for Bone and Mineral Research (ASBMR).

2003-2006: Junior Scientisit, San Raffaele Scientific Institute, Milano, Italy. Here, Dr Cenci provided novel insights into terminal plasma cell (PC) differentiation, discovering proteotoxic stress as a novel inbuilt counter setting PC lifespan (*EMBO J 2006, FEBS Lett 2007).*

2006-present: Staff Scientist & Group Leader, San Raffaele Scientific Institute, Milano, Italy. As an independent group leader, supported by the European Calcified Tissue Society (ECTS), the Multiple Myeloma Research Foundation (MMRF) and the Italian Ministry of Health, Dr Cenci focused his research on the biology of normal and malignant PCs, with particular interest in the role of protein homeostasis in myeloma cell survival (*Blood 2009, Curr Opin Cell il 2011, J Leuj Biol 2012, Semin Hematol 2012)* and of autphagy in PC ontogenesis (*Nature Immunol 2013, Autophagy 2013)*. Dr Cenci's articles have been cited 1,531 times (H index: 16), and he filled 2 international patents. The Cenci Lab comprises 10 members (4 postdocs, 1 MS students, 1 PhD student, 2 graduate students, and 2 technicians).

Academic activities: training of (under)graduate and PhD students. Dr Cenci teaches Cell Biology, Gerontology and Geriatrics, and serves as a board member in the Immunology PhD Program at Universitá San Raffaele. He delivers international invited lectures, and serves as an active member of many international scientific societies, including the American Society of Hematology (ASH), the ECTS, and the ASBMR.

Metabolomics – Simone Cenci

Abstract

Metabolomics is a new wide-scope approach that provides a comprehensive profile of small metabolites within a biological system. Resulting from the complex interaction between cells and molecules, the *metabolome* yields an unbiased, integrative view of the environment. Hence, metabolomics - the "systems biology of small molecules" - demonstrated unprecedented power for biomarker discovery and dissection of pathogenic mechanisms in human complex diseases (Spratlin JL et al, *Clin Cancer Res* 2009;15:431-40; Sreekumar A et al, *Nature* 2009;457:910-14; Kettunen J et al, *Nat Genet* 2012;44:269-76).

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Multiple myeloma (MM) is a malignancy of plasma cells, which grow at multiple foci in the bone marrow (BM), secrete monoclonal immunoglobulins, and induce skeletal destruction, hypercalcemia, anemia, immune suppression, and renal failure. Although novel therapies improved overall survival, this cancer remains incurable, causing ~2% of all cancer deaths. The development and progression of MM rely on vicious interactions with the BM environment (Palumbo A & Anderson K, *N Engl J Med* 2011;364:1046-60; Hideshima T et al, *Nat Rev Cancer* 2007;7:585-98). MM enhances osteoclastogenesis and neoangiogenesis, and reduces osteoblast differentiation and activity (Roodman GD, *J Bone Min Res* 2010;28:244-50). The diversion of the BM milieu sustains cancer cell survival, proliferation, migration and chemoresistance (Palumbo A & Anderson K, *N Engl J Med* 2011). However, our understanding of the interplay between MM and the BM environment in its complexity and of its exact role in disease evolution remain still elusive.

MM is a unique paradigm to investigate complex bone-cancer relationship through metabolomics. We employed a high throughput platform, *ultra-high performance liquid and gas chromatography followed by mass spectrometry* (UHPLC/GC-MS), to define the metabolome of the BM environment in MM pathophysiology. Data processing, unsupervised/supervised data analyses, metabolic modeling, multiple discriminant analyses, combined with hypothesis-driven *in vitro* studies, disclosed a number of microenvironmental changes associated with myeloma evolution. These studies identified unanticipated prognostic markers and putative pathomechanisms and specific therapeutic targets.

Christian Dierkes (Trier, Germany)

Biography

Current position: Specialist in pathology at MVZ für Histologie, Zytologie, Molekulare Diagnostik , Trier, Germany

1999	German Medical Exam at the Justus-Liebig-University, Giessen
1999-2008	Specialization in surgical pathology at St.-Vinzentius-Kliniken, Karlsruhe, Germany (Arzt im Praktikum) and Institute for Pathology, Justus-Liebig-University, Giessen, Germany
2002-2003	Research assistant project B1 SFB 547„Kardiopulmonales Gefäßsystem“, Justus-Liebig-University, Giessen
2008-2009	Specialist in Pathology, Institute for Pathology, Justus-Liebig-Universtiy, Giessen
2009-2011	Specialist in Pathology, Gerhard-Domagk-Insitut für Pathologie, University-Clinic, Muenster, Germany

Main interest in bone, joint and soft tissue pathology.

Doctoral thesis: Laser-assisted microdissection for molecular analysis of human endosteal lining cells from native human bone

Short list of journal articles:
Immunohistochemical expression of cyclooxygenase isoenzymes and downstream enzymes in human lung tumors.
Ermert L, Dierkes C, Ermert M. Clin Cancer Res. 2003 May;9(5):1604-10.
Interindividual variability, pathological changes and decomposition as an impediment to the morphological determination of human specificity of bone finds. Verhoff MA, Rensing N, Kreutz K, Dierkes C, Ramsthaler F. Arch Kriminol. 2008 Mar-Apr;221(3-4):99-112.
Catabolic properties of microdissected human endosteal bone lining cells. Dierkes C, Kreisel M, Schulz A, Steinmeyer J, Wolff JC, Fink L. Calcif Tissue Int. 2009 Feb;84(2):146-55.
Genomic alterations and allelic imbalances are strong prognostic predictors in osteosarcoma. Smida J, Baumhoer D, Rosemann M, Walch A, Bielack S, Poremba C, Remberger K, Korsching E, Scheurlen W, Dierkes C, Burdach S, Jundt G, Atkinson MJ, Nathrath M. Clin Cancer Res. 2010 Aug 15;16(16):4256-67.
Indications for tissue biopsy. Diagnostic histopathology in rheumatological diseases. Krenn V, Poremba C, Dierkes C. Z Rheumatol. 2012 Jun;71(4):297-311
MicroRNA-34c inversely couples the biological functions of the runt-related transcription factor RUNX2 and the tumor suppressor p53 in osteosarcoma. van der Deen M, Taipaleenmaki H, Zhang Y, Teplyuk NM, Gupta A, Cinghu S, Shogren K, Maran A, Yaszemski MJ, Ling L, Cool SM, Leong DT, Dierkes C, Zustin J, Salto-Tellez M, Ito Y, Bae SC, Zielenska M, Squire JA, Lian JB, Stein JL, Zambetti GP, Jones SN, Galindo M, Hesse E, Stein GS, van Wijnen AJ. J Biol Chem. 2013 May 29.

Ewing sarcoma dissemination and response to T cell therapy in mice assessed by whole body magnetic resonance imaging

Lennart Liebsch, Sareetha Kailayangiri, Laura Beck, Bianca Altvater, Raphael Koch, Christian Dierkes, Marc Hotfilder, Nina Nagelmann, Cornelius Faber, Hendrik Kooijman, Janine Ring, Volker Vieth, and Claudia Rossig. Br J Canc, *in press*

Claus-C Glüer (Kiel, Germany)

Biography

Dr. Claus-C. Glüer was born in Hamburg, Germany, studied physics at the University of Hamburg, Germany, and earned his Diplom-Physiker degree in 1982. In his dissertation at HASYLAB DESY, Germany, he first began working on medical applications of X-ray, developing synchrotron-based techniques for coronary angiography. He received his Dr. rer. nat. degree from the University of Hamburg, Germany, in 1986.

After 7 years of research as *Assistant* and *Associate Adjunct Professor* in the Department of Radiology at the University of California, San Francisco, in 1995 he became a Professor of Medical Physics in the Department of Diagnostic Radiology, University Hospital Schleswig-Holstein in Kiel, Germany. He is the president of the *Deutsche Gesellschaft für Osteologie* and president-elect of the *European Calcified Tissue Society*. He is associate editor of Osteoporosis International and member of the editorial board of three other professional journals.

Dr. Glüer's research is focused on the development of innovative parametric imaging techniques and their quantitative evaluation. Working in the field of osteoporosis for more than 25 years, he has contributed specifically to the development of bone densitometry, quantitative ultrasound, and high-resolution computed tomography approaches. He has coordinated several multicenter studies including OPUS, a European project on epidemiology and optimized diagnostic assessment of osteoporosis. He also has a strong research interest in developing multimodal methods for molecular imaging with applications in oncology, inflammation, and skeletal research. Since 2010 he is head of the *Molecular Imaging North Competence Center* (MOIN CC) at the Christian-Albrechts-Universität zu Kiel.

Dr. Glüer is a member of 15 professional societies and is the current president of the Germany Academy of Bone and Joint Sciences. He has published more than 170 original papers.

Innovative skeletal imaging techniques – Claus-C Glüer

Abstract

Imaging techniques play an important role in the diagnostic assessment of patients as well as in the preclinical characterization of bone disease and response to treatment. A very large number of imaging modalities can be employed each of them with specific advantages and disadvantages. In this overview lecture I would like to briefly review current trends in imaging of the skeleton addressing both preclinical as well as clinical methods applicable in vivo.

<u>Imaging approach categories</u>

Imaging provides localized information about pathophysiological processes and the effect on the skeleton and, if performed non-destructively in vivo, permits monitoring of temporal development of these processes over time. Two-dimensional imaging approaches have a long history, including transmission and reflection techniques. Today, images acquired with almost any of the modalities can be reconstructed in three dimensions, resulting in tomographic data, either rendered in 3-D or reformatted in arbitrary 2-D sections.

A large variety of features of biological materials can be visualized as summarized below.

Morphological imaging provides insight into the detailed anatomy with a spatial resolution of at best around 0,1 mm in humans and 0,01 mm in animals. Moving beyond these limits is technically feasible but induces inacceptable radiation levels or imaging times, depending on the technique. When applied clinically, better resolution can be achieved at peripheral measurement sites but these may or may not be representative of pathological changes at relevant central sites. For some techniques tissue contrast is limited but can be enhanced by using *contrast agents*, e.g. angiography to depict the vascular system.

Functional imaging permits depiction of organ, tissue or cell function. In the case of bone, mechanical competence is one of the most important aspects. It can be assessed by using the image data to simulate mechanical loads using the engineering methods of *finite element analysis*. This permits calculation of the response of whole bones to specific loads and generates estimates of bone fragility and fracture risk. Another functional aspect relevant for the skeleton is its vascularity. A characterization of the vascular network beyond its micromorphology can be performed by perfusion techniques. While perfusion techniques are well established in soft tissue studies assessment of bone vasculature is extremely challenging due to physical characteristics of bone and the spatial resolution required. The characterization of angiogenic processes is of particular relevance in oncological imaging but also for the assessment of inflammation or fracture repair. Another aspect of generic importance is imaging of cell or tissue viability.

Property mapping is another category in imaging. For a variety of techniques the graylevel of the image reflects a specific physico-chemical property of the tissue or material under study. Employing calibration techniques permits analysis in a quantitative fashion. Examples include bone densitometry, reflecting calcium hydroxyapatite concentration or fat/water ratios but also a large variety of spectroscopic imaging approaches.

Specific labelling. For most imaging modalities few molecules have a strong specific inherent contrast and even tissues may lack specific native contrast. Efforts to overcome this issue by labelling molecules with high contrast markers have been very successful in recent years, moving substantially beyond classical contrast agents, which have limited specificity. The most well-known approach is termed *molecular imaging* which can be broadly defined as the in vivo characterization and measurement of biologic processes at the cellular and molecular level by means of imaging methods (1). Most molecular imaging methods rely on some type of marker that binds to the molecule in question, providing image contrast. Labelling antibodies, receptors or drugs permits detailed insight into the molecular biology of skeletal physiology, pathological processes and their treatment. Using molecular imaging technique cells, drugs and drug carriers can be imaged. The techniques can be used for micro morphological and functional imaging, e.g. by visualization of enzyme activity. Genetic engineering methods permit labelling promoters of specific genetic encoding processes, resulting in genetic reporters. For the most part such approaches are limited to preclinical studies, in part because of the restrictions in employing genetic engineering methods, i.e. to generate transgenic animal models, but also due to limitations in the imaging depth of even the most powerful imaging technologies. However, recent improvements permit sensitive and specific imaging of some aspects of human skeletal metabolism in vivo.

Imaging modalities

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The range of imaging modalities used in the life sciences is very broad and in this lecture only a short overview of the techniques most relevant for skeletal imaging and of the most exciting innovations can be provided. Virtually any of the modalities can be used to study morphological, functional or molecular aspects, albeit with specific strengths and limitations. The techniques most commonly used include x-ray based approaches (including radiography, computed tomography, bone densitometry), magnetic resonance imaging and spectroscopy, sonography, optical imaging including fluorescence and bioluminescence, and imaging methods employing nuclear medicine techniques. In this script I will point the reader to relevant review articles covering the major applications of these modalities in skeletal research along with a few paper highlighting recent advanced methods.

X-ray based approaches

Morphological imaging: Micro-CT (µCT) (2-4) and High Resolution (peripheral) Computed Tomography (HRQCT (5, 6) and HRpQCT (7, 8))

Functional imaging: Finite Element Analysis (9), usually based on QCT (10), HR(p)QCT (11) or µCT (12). Application also for assessment of bone implants (13).

Property mapping: bone densitometry based on Dual X-ray Absorptiometry (DXA) and Quantitative Computed Tomography (QCT) (14).

Magnetic Resonance (MR)

Morphological imaging: high resolution MR imaging (HRMRI) (15, 16), bone erosions (17)

Functional imaging: Finite Element Analysis based on MRI (18), Contrast agents (19, 20){Daldrup-Link, 2009 #93

Property mapping: MR susceptibility imaging of trabecular microstructure {Majumdar, 1991 #2;Majumdar, 1991 #3}, MR spectroscopy for measuring bone marrow fat content and composition (21), ultrashort TE for measuring cortical bone properties (22), diffusion imaging of musculoskeletal tumours (23).

Specific labelling approaches: Stem cell tracking (24-26), Macrophage tracking (27)

Sonography

Morphological imaging: acoustic microscopy for assessment of porosity (28), bone ultrastructure (29)

Property mapping: bone strength and fracture risk (30, 31), mechanical properties of cortical bone (32-35), assessment of trabecular bone microstructure (36), antiangiogenic treatment assessment (37)

Optical imaging

Functional imaging: cathepsin K (38), VEGF expression in bone grafts (39), bone metastases (40)

Labeling approaches: review (41), mineral labelling (42), bone metastases (40), bone metastases treatment monitoring (43), multiple myeloma model (44, 45), fracture healing (46), genetic reporters (47, 48), osteoclast labelling (49), estrogen receptor reporter (50)

Nuclear medicine

Functional imaging: bone turnover (51)

Property mapping: bone metastases from prostate (52), breast cancer (53) and misc. cancers (54).

Specific labeling approaches: bone scan (55), Fluoride PET (56-58), choline PET (58, 59), FDG PET (57, 58, 60).

Multimodality reviews

Osteoporosis (61, 62), glucocorticoid induced osteoporosis (63), bone metastases imaging (43, 64), myeloma (65), skeletal engraftment (66), molecular imaging review (beyond bone) (1, 67, 68)

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Jenny Gregory (Aberdeen, UK)

Biography

Jenny Gregory obtained her PhD in 2005 from Aberdeen University, following an undergraduate degree from Leeds University in Artificial Intelligence and Operational Research.

My research interests lie in medical image processing and understanding. My main research area is examining the shape of bones and joints on Dual Energy Xray-Absorptiometry (DXA) scans, radiographs and MRI scans and seeing how they relate to bone disorders such as Osteoporosis and Osteoarthritis. I am currently working on a Medical Research Council (MRC) New Investigator Award looking at bone shape in osteoarthritis of the knees. However I have worked and am interested in working with images from many different sources, including microscopy, radiographs and photographs.

Please note, the Image J course is a practical workshop. Please bring your own laptop with you (PC/Apple/Linux all acceptable). If you have any specific topics you would like included, please contact me directly before the course and I will try to incorporate them.

Image Analysis (Image J) – Jenny Gregory

Abstract

This interactive workshop will provide an introduction to working with digital images using ImageJ/Fiji. Although there are many different imaging packages available, ImageJ was chosen because it is free, works on any standard computing system (Unix, PC, Macintosh) and is widely supported by the biomedical research community, by both papers and 'plugins' (small add-on programs written by users for a specific task). "Fiji" may also be used, as it is essentially the same program (The Fiji acronym stands for Fiji Is Just Imagej).

The course will start at a very simple level, presuming no previous knowledge of image analysis or ImageJ/Fiji. It will give the user experience of the common steps required in any imaging tasks (how to open, view and save different types of images, adjust brightness and contrast, calibrate your image, perform basic measurement and counting procedures and export the results) alongside an overview of some of the more advanced steps (how to remove noise and background illumination, automate counting and measurement, write macros, use plugins, work with 3D or timelapse datasets). A set of example images will be provided to work with, but attendees are welcome to bring their own examples too (and can also email me at j.gregory@abdn.ac.uk if there are any specific topics they would like to cover).

References

The online user guide for ImageJ (1) is a good introduction. The list of available plugins (2) will give a good idea the wide scope of work going on and is a valuable resource. Finally, I recommend a paper by Doube *et al.* describing the imageJ plugin "BoneJ" (3). This is an excellent example of how ImageJ can be used to develop and share image analysis measures and may also be a useful tool for many members of ECTS.

Ferreira, T and Rasband, W ImageJ User Guide 1.45m http://rsbweb.nih.gov/ij/docs/user-guide.pdf

Multiple authors ImageJ Plugins http://rsb.info.nih.gov/ij/plugins/index.html

Doube M, Klosowski MM, Arganda-Carreras I, Cordelieres FP, Dougherty RP, Jackson JS, et al. BoneJ: Free and extensible bone image analysis in ImageJ. Bone 2010 Dec;47(6):1076-9

Núria Guañabens (Barcelona, Spain)

Biography

Professor of Medicine, University of Barcelona, Spain

Place of Birth: Barcelona, Spain.

Education/Training/Positions: MD, University of Barcelona, Spain (1978); Fellow in Rheumatology, Hospital Clinic, Barcelona (1979-1982); PhD, University of Barcelona (1987); Visiting scientist/Postdoctoral Research Fellow, The Jewish Hospital, St. Louis, MO (1988) and The Johns Hopkins University School of Medicine (1992); Associate Professor of Medicine, University of Barcelona (1996-2007); Aggregate Professor of Medicine, University of Barcelona (2007-present); Staff member of the Department of Rheumatology, Hospital Clinic, Barcelona (1985-2004); Head of the Department of Rheumatology (2004-present).

Honors/Awards: Residency Hospital Clínic Award**,** Barcelona (1983); Professional Excellence Award from the Medical Association of Barcelona (2009).

Editorial Duties/Peer Review Panels: Editorial Boards: Medicina Clínica, Reumatología Clínica, Revista de Osteoporosis y Metabolismo Mineral. Ad Hoc reviewer of Manuscripts: Bone, Osteoporosis International, Nature Reviews Gastroenterology & Hepatology, Journal of Hepatology. Panel Member/External reviewer: Spanish Society of Rheumatology; Catalan Agency for Health Technology Assessment and Research; Health Research Fund, Health Institute Carlos III; The Efficacy and Mechanism Evaluation (EME) Programme (UK).

Professional Societies: Membership: American Society for Bone and Mineral Research, International Bone & Mineral Society, Spanish Society for Bone and Mineral Research, Spanish Society of Rheumatology, Catalan Society of Rheumatology. Treasurer (1999-2003) and President (2003-2007) Spanish Society for Bone and Mineral Research; Councilor Spanish Society of Rheumatology (2000-2004) and Catalan Society of Rheumatology (1995-1997). Member, International Bone & Mineral Society Meetings Committee.

Current Research: Bone markers, liver and bone; bone disease of organ transplantation, particularly after liver transplantation; premenopausal osteoporosis, male osteoporosis, Paget's disease of bone, bone cell cultures.

Meetings organizer: Local co-organizer, 35th European Symposium on Calcified Tissues and ECTS Training Course; Co-chair, Spanish Society for Bone and Mineral Research Annual Scientific Meetings (2003-2007).

Osteoporosis - Núria Guañabens

Abstract

This PhD training session will cover the most relevant clinical aspects of osteoporosis. The questions to be answered will be: what is osteoporosis?, what are the clinical features and diagnostic tools? and finally, which drugs are in use and under development?.

The main fractures (wrist, vertebra and hip) in postmenopausal women with osteoporosis will be described, as well as their timing. In this context, the assessment work-up will be carefully developed, including the risk factors, symptoms and clinical findings, as well as the recommended lab tests, x-rays, bone densitometry and other diagnostic tools, such as the FRAX tool. FRAX estimates the 10-yr probability of a major osteoporotic fracture or a hip fracture. When considering lab tests, the reasons for performing them and the most recommended tests will be addressed, as well as the clinical utility of the biochemical markers of bone turnover. In addition, basic concepts regarding bone densitometry and its interpretation will be provided.

Treatment of osteoporosis will be discussed from the bisphosphonates to denosumab, as well as the future of drug therapy in osteoporosis. Bisphosphonates will be addressed taking into consideration their history, mechanisms of action and their main characteristics. Similarly, denosumab (monoclonal antibody which inhibits RANKL) and teriparatide (PTH 1-34) will be introduced, pointing to their mechanisms of action. Finally, an overview of future therapeutic targets in osteoporosis such as anti-cathepsin K (osteoclast-specific colagenase) and anti-sclerostin (Wnt signalling antagonist) will be given.

A vignette of a postmenopausal woman with osteoporosis will close the session, after discussion.

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Russell RGG. Bisphosphonates: The first 40 years. Bone 2011;49:2-19.

Christine Hartmann (Muenster, Germany)

Biography

1985-1991	Studies in Biology, Universities of Ulm and Munich
1991	Diploma in Biology with Dr. Herbert Jäckle, Ludwig-Maximilians University Munich, Germany
1991-1996	Ph.D. studies with Dr. Herbert Jäckle, Max-Planck Institute for Biophysical Chemistry, Göttingen, Germany
1996	Ph.D. rer. nat. in Genetics, Ludwig-Maximilians University Munich, Germany
1996-2002	Postdoctoral fellow, Harvard Medical School, Boston, USA
2002 – 2012	Group leader, Research Institute of Molecular Pathology, Dr. Bohrgasse 7, 1030 Vienna, Austria
2012 – now	W3 Professor at the Medical Faculty, WWU Münster
2010	Habilitation ("Signaling Pathways in Vertebrate Skeletogenesis") in Developmental Biology and Genetics at the University of Vienna, Austria

Selected publications:

Lyashenko, N., Winter, M., Migliorini, D., Biechele, T., Moon. R., and **Hartmann, C**. (2011). Differential requirement for the dual functions of β-catenin in embryonic stem cell self-renewal and germ layer differentiation. **Nat. Cell Biol**, 7, 753-761

Fujimori, S., Novak, H., Weissenböck, M., Jussila, M., Gonçalves, A., Zeller, R., Galloway, J., Thesleff, J., and **Hartmann, C**. (2010). Wnt/β-catenin signaling in the dental mesenchyme regulates incisor development and number by regulating Bmp4. **Dev Biol**, 348/1, 97-106

Später, D, Hill, T.P., O'Sullivan, R.J., Gruber, M., Conner, D.A., and **Hartmann, C**. (2006). Wnt9a signaling is required to maintain joint integrity and regulates Ihh expression during chondrogenesis. **Development**, 133, 3039-3049

Hill, T.P., Taketo, M. M., Birchmeier, W., and **Hartmann, C**. (2006). Multiple roles of mesenchymal β-catenin during murine limb bud development. **Development**, 133, 1219-1229

Hill, T. P., Später, D., Taketo, M. M., Birchmeier, W., and **Hartmann, C**. (2005). Canonical Wnt/beta-catenin signaling prevents osteoblasts from differentiating into chondrocytes. **Dev. Cell** 8, 727-38

Hartmann, C., and Tabin, C. J. (2001). Wnt-14 plays a pivotal role in inducing synovial joint formation in the developing appendicular skeleton. **Cell** 104, 341-51

Hartmann, C., and Tabin, C. J. (2000). Dual roles of Wnt signaling during chondro-genesis in the chicken limb. **Development** 127, 3141-59

Bone Development – Christine Hartmann

ECTS PhD TRAINING 15-18 September 2013, Hamburg

Abstract

All higher vertebrates have an internal support system, the bony endoskeleton. Bone is a highly specialized form of connective tissue, with a mineralized extracellular matrix, which contains type I collagen as the major component. Bone confers rigidity and strength to the vertebrate skeleton. The cell type that builds up the bone tissue is the osteoblast. Osteoblasts are, like the cell type that forms the cartilage - the chondrocyte, of mesenchymal origin. The bones of the vertebrate skeleton are formed during embryogenesis by two different processes - intramembranous and endochondral ossification. In the first, mesenchymal cells condense to differentiate directly into osteoblasts, which secret an unmineralized matrix, the osteoid, that later becomes mineralized and reorganizes into either compact (bone collar) or cancellous (trabecular) bone. In the second process, mesenchymal cells condense and cells within the condensations differentiate into chondrocytes, these form the cartilaginous anlagen of the future skeletal elements. Indian hedgehog, a secreted signal from the chondrocytes is required to induce osteoblastogenesis through the induction of the expression of the transcription factor Runx2/Cbfa1 in the cells of the surrounding perichondrium, the region where those differentiate is then referred to as the periosteum[1,2]. For the final replacement of the cartilaginous template with bone, chondrocytes are required to mature into hypertrophic chondrocytes, which eventually mineralize their extracellular matrix. It is then assumed that the final fate of hypertrophic chondrocytes is to undergo apoptosis or to be removed by chondroclasts and osteoclasts, leaving behind a mineralized scaffold that is then used by osteoblast to build the primary spongiosa / trabecular bone[3,4]. Lineage tracing studies revealed that the osteoblasts which build up the trabecular bone originate from the periosteum and that they migrate in as precursors together with blood vessels[5]. There is more and more evidence that some trabecular / endosteal osteoblasts may have an alternative origin and originate from hypertrophic chondrocytes by transdifferentiation.

Numerous signaling pathways and transcription factors have been identified which either control osteoblastogenesis or are essential for bone formation, some of which will be discussed in this lecture.

References:

1. Long, F. *et al.* (2004). Ihh signaling is directly required for the osteoblast lineage in the endochondral skeleton. *Development* 131, 1309-1318

2. St-Jacques, B., Hammerschmidt, M. & McMahon, A. P. (1999). Indian hedgehog signaling regulates proliferation and differentiation of chondrocytes and is essential for bone formation. *Genes Dev* 13, 2072-2086

3. Mackie, E. J., Tatarczuch, L. & Mirams, M. (2011). The skeleton: a multi-functional complex organ: the growth plate chondrocyte and endochondral ossification. *J. Endocrinol* 211, 109-121

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5. Maes, C. *et al.* (2010). Osteoblast precursors, but not mature osteoblasts, move into developing and fractured bones along with invading blood vessels. *Dev Cell* 19, 329-344

Miep Helfrich (Aberdeen, UK)

Biography

Miep Helfrich is Professor of Bone Cell Biology at the University of Aberdeen. She studied cell biology at the University of Wageningen in the Netherlands, where she became interested in microscopical imaging. She obtained her PhD in 1988 at the University of Leiden, the Netherlands, where she studied osteopetrosis and developed an interest in ultrastructural imaging of osteoclasts. As postdoc at the Imperial Cancer Research Fund in London she studied the role of integrins in bone cell function and then moved to the University of Aberdeen where she worked on the etiology of Paget's disease of bone. As independent fellow, funded by the Arthritis Research Campaign, she worked at the University of Aberdeen from 1996-2004 on the role of nitric oxide as mediator of mechanical signalling in bone. She was appointed to a senior lectureship in Aberdeen in 2004 and to chair in 2009. She has been the academic lead of the microscopy core facility in Aberdeen since 2002. In 2010 she became leader of the Musculoskeletal Research Programme in Aberdeen. Her current research interests are in understanding the etiology of osteoclast diseases, such as osteopetrosis and Paget's disease of bone, with a recent interest in the role of autophagy in bone physiology and pathology. Another key interest is in improving ultrastructural imaging of bone cells and bone tissue. She was board member of ECTS from 2009-2012 when she chaired the Training Committee and organised previous PhD training courses. She has extensive experience with writing and reviewing funding applications for various funders, largely in the UK, but also for various other national agencies, for ECTS and for the EU.

Grant Writing – Miep Helfrich & Lynne Hocking

Abstract

In this workshop we will take you through the process of writing a grant application assuming that for most of you it is likely to be your first ever grant application. We will focus therefore on the type of grant you are most likely to write as a graduated PhD student: an application for your own salary, a type of fellowship application. Elements of such proposals are relevant to many other situations too and therefore this workshop is useful even if you do not immediately put pen to paper, but for example apply for a funded position.

For example: you will have to write about yourself, so CV presentation will be covered. You will have to be able to summarise your project, including for an audience that consists of non-experts, so your "elevator speech" will need to be perfected. You will have to be able to write convincingly about the importance and feasibility of the project you propose and the relevance of the questions to be addressed to society as a whole. Here, we will cover how to write concise and clear text that engages the reader.

We will use elements of various funding schemes (National and EU) as examples during the workshop to illustrate different aspects of the application process.

You will be asked to do some writing and reading during the workshop and you will be asked to do some preparation. We will hand out this homework the day before the workshop as you will be able to complete this in a relatively short time.

What you should do before coming to Hamburg:

ECTS PhD TRAINING 15-18 September 2013, Hamburg

- Prepare a 300 word abstract of the project you are currently working on, or of another study (perhaps the one you wish to find funding for ?). Your abstract has to be in English and understandable to a scientific readership. Print out 2 copies on A4 and bring these along to the workshop. Your abstract should inform the reader of the purpose of your study, the aims, the methodology and whom the results may benefit. Feel free to discuss this with others, including your supervisor.
- Read webpages and have a look at the guidance for applications of funders you may consider applying to.
  Please bring such information with you when attending this workshop, or have the web address to hand.
- Consult general books about scientific writing, or general writing.
  We like:
  1. Rowena Murray, R & Sarah Moore, S *The handbook of academic writing: A fresh approach*. Open University Press-McGraw-Hill
  2. Lynne Truss. *Eats shoots and leaves.* Profile Books or Fourth Estate.
  A humoristic and useful book about correct use of punctuation.
  Both books will be available during the workshop to browse.
- There are many books about grant writing, but few about writing scientific grants. *If you have come across a useful book, please bring it along for others to browse.*

Christian Hellmich (Vienna, Austria)

Biography

Education:

Studies at Vienna University of Technology (TU Wien), Civil Engineering, 1990-1995

Dipl.-Ing. (M.Sc.), 1995

Dr. Techn. (Ph.D.), 1999

Habilitation, TU Wien, 2004

Positions:

University Assistant, TU Wien, 1995-1999

Postdoctoral Fellow at Massachusetts Institute of Technology, 2000-2002

Assitant Professor, TU Wien, 2003

Associate Professor, TU Wien, 2004-2010

Full Professor, TU Wien, 2011-

Director of Institute for Mechnias of Materials and Structures, TU Wien, 2012-

Scientific output:

>85 refereed journal papers in the fields of Civil an Biomedical Engineering, with >1300 citations and h=20 according to www.scopus.com; 19 book chapters, >200 contributions to conference proceedings, >200 conference and seminar presentations

Service to the community:

Since 2004: Associate Editor of Journal of Engineering Mechanics (ASCE)- SCI; since 2011: Corresponding Editor of Computer Methods in Engineering Sciences – SCI; since 2013: Editor-in-chief of Molecular and Cellular Biomechanics – SCI; member and/or chair of ASCE-EMI committees on Material Properties, Promomechanics, Multiscale Modelling, Biomechanics, RILEM committee on concrete modelling; reviewing for >75 refereed journals; Chairman of 8 international conference/workshops; organiser of 44 symposia at scientific conferences, member of 22 international conference committees, Coordinator of EU-FP7 research for SMEs project BIO-=CT-EXPLIT (2009-2011), Vice -Chair of COST Action NAMAGIO (2011-2015); teaching in engineering mechanics, biomechanics, computational mechanics, multiscale modelling at TU Wien, M.I.T., Austrian Academic of Sciences, Univ Paris-Estm Politecnico di Milano.

Awards (selection):

Kardinal Innitzer Advancement Award of the Archbishopry of Vienne (2004), Lower Austrian Science Award (2005), Zienkiewicz Award of European Community on Computational Methods in Applied

Sciences (2008), ERC Starting Grants of the European Research Council (2010), Member of Young Austrian Academy of Sciences (2011), Walter L Huber Research Prize of the American Society of Civil Engineers (ASCE) (2012).

Micromechanics and Systems Biology of Bone – Christian Hellmich

Abstract

ccording to the eminent Austro-American zoologist Rupert Riedl (1925-2005), "… the living world happens to be crowded by universal patterns of organization …". While Riedl, as "classical" biologist, typically took a descriptive approach to this issue, we ventured, over the last decade and in particular during the last few years, into an engineering science approach of mathematical nature, where we have indeed been successful in identifying „universal" rules/patterns in structural biology and their mechanical consequences. A majority of our investigations concerned mineralized biological tissues such as bones, for which we identified the following mathematically cast rules: (I) In extracellular bone tissues across different organs from different animals/humans at different ages, mineral (hydroxyapatite) and collagen contents are not randomly assgined to each other, but fulfill astonishingly precise bilinear relations[1], which follow from rigorous evaluation of dehydration, demineralization, ashing, and de-organifying test data collected over a time period of more than 80 years of experimental research. Furthermore, (II) the distribution of mineral throughout the extracellular bone matrix or ultrastructure, i.e. its partitioning into the fibrillar and extrafibrillar spaces is governed by the on-average uniformity of hydroxyapatite concentration in the *extracollageneous* space[2], as was evidenced from chemical tests like the ones mentioned before, in combination with transmission electron micrographs. Before mineralization (as well as in unmineralized collageneous tissues such as tendon or cartilage), the fibrillar and extrafibrillar spaces again obey another general rule: (III) Upon hydration, the extrafibrillar space grows propertional to the fibrillar volume gain due to accomodation of water in the intermolecular spaces[3], as evidenced from dehydration and neutron diffraction tests. Finally, (IV) mineralization of such tissues is driven by fluid-to-solid phase transformations in the extracollageneous space under closed thermodynamic conditions[4], predicting precisely the volume losses which the tissues undergo during mineralization. All these compositional and structural rules may serve as ideal input for multiscale mechanics models for the elasticity[5], strength[6], and creep[7] of bone tissues; enabling various clinical applications, such as Computed Tomography (CT)-based Finite Element (FE) analysis for biomaterial design[8].

On the other hand, these structural and multiscale mechanics models have a clear interface to mathematical systems biology: in terms of mass or volume of produced extracellular bone matrix and ist composition, systems biology may provide input values for the micromechanics models: this marls a new generation of mechanobiological models[9], holding the promise of predicting bome modeling and remodeling at an unprecedented level of precision.

References

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2 Hellmich, Ulm, *Biomech Model Mechanobiol* 2: 21–36, 2003.
3 Morin, Hellmich, Henits, *J Theor Biol 317,* 384-393, 2013.
4 Morin, Hellmich, *J Theor Biol*, 317: 384–393, 2013.
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6 Fritsch, Hellmich, Dormieux, *J Theor Biol* 260: 230 – 252, 2009.
7 Eberhardsteiner, Hellmich, Scheiner, *Comp Meth Biomech Biomed Eng* DOI:10.1018/10255842.2012.670227, 2013

8 Dejaco, Komlev, Jaroszewicz, Swieszkowski, Hellmich. *J Biomech* 45, 1068-1075, 2012.
9 Scheiner, Pivonka, Hellmich, *Comp Meth Appl Mech Eng* 254: 181–196, 2013.

Eric Hesse (Hamburg, Germany)

Biography

Dr. Eric Hesse studied medicine at Hannover Medical School in Germany where he became MD in 2003. He was trained in Orthopedic Surgery and graduated as PhD in 2007 in Genetics & Cell Biology in Hannover, Germany. In 2005, he moved to the laboratory of Dr. Roland Baron at Yale University School of Medicine as a Postdoctoral Fellow funded by the German Research Foundation. The laboratory was relocated to Harvard University Schools of Medicine and Dental Medicine in 2008, where he continued his work as a Postdoc and later as Junior Faculty until 2011. During this time, he worked on clinical and basic science projects focusing on osteoblast biology and bone homeostasis, leading to publications in top tier journals including JCB, JBMR, Dev Cell, Bone, and the NEJM. He received numerous awards and fellowships, including the ASBMR Young Investigator-, John Haddad- and Harold Frost Award, the ECTS New Investigator Award, the Harvard Deans Fellowship, and the Gideon & Sevgi Rodan IBMS Fellowship. In 2011, he moved to the University Medical Center Hamburg-Eppendorf in Germany, where he established an independent international research group as full, endowed, tenure-track Heisenberg-Professor. His research continues to focus on translational aspects of osteoblast function and bone remodeling and is funded by the German Research Foundation, the European Union, the Helmholtz Association, and several private Foundations. Dr Hesse is Director of Research of the Department of Trauma, Hand, and Reconstructive Surgery, in which he is practicing as an Orthopedic Surgeon. In addition, he was Co-Chair of the IBMS Young Investigator Committee, is a member of several IBMS, ASBMR, and ECTS committees and served as local organizer of the 2013 ECTS PhD Training course.

Bone Cells: Osteoblasts – Eric Hesse

Osteoblasts are the cells that form the mammalian bones, and are often implicated in diseases affecting the skeleton. The overall aim of this lecture is to provide an in-depth understanding of the origin of osteoblasts and of molecular mechanisms controlling lineage allocation, differentiation and function of osteoblasts at the cellular and tissue level. An overview of the most important signaling pathways and networks of transcription factors and co-regulators and their implication in physiological and pathological conditions will be presented.

Within the bones, osteoblasts interact with nearby tissues including the vasculature, the hematopoietic- and the immune system, and in particular with other bones cells such as osteocytes and osteoclasts. All of these cross talks are important for preserving the integrity of the bones but the interaction with the bone-resorbing osteoclasts is of particular importance for the life-long remodeling of the skeleton and the maintenance of the bone mass. Thus, the seminar intends to explore molecular- and cellular interactions osteoblasts are engaged in to reach an integrative view of the osteoblasts acting as key players in the formation, maintenance, and regeneration of osseous tissue. This includes a conceptual understanding of bone remodeling and its alterations, the basis of the debilitating disease osteoporosis. In addition, we will learn about pharmacological principles to treat osteoporosis with special emphasis on bone anabolic approaches.

On a technical level, we will discuss *in vitro* and *in vivo* mouse genetic approaches to study the differentiation and function of osteoblasts using recent discoveries as examples. Based on the

lessons learned from human diseases and the laboratory approaches, we will then move on to discuss future scientific and clinical directions of the field of osteoblast biology.

References

1) Update on bone anabolics in osteoporosis treatment: rationale, current status, and perspectives. Baron R, Hesse E. J Clin Endocrinol Metab. 2012 Feb;97(2):311-25.

2) WNT signaling in bone homeostasis and disease: from human mutations to treatments. Baron R, Kneissel M. Nat Med. 2013 Feb;19(2):179-92

Histomorphometry - Eric Hesse & Hiroaki Saito

Abstract

The overall goal of this histomorphometry workshop is to initiate an interactive discussion about the technical and conceptual principles of histomorphometry as a very specialized but important method in the field. This lecture will include practical examples and exercises coming from recently published or ongoing hot topic projects in the field of bone research. The seminar intends to teach the technical principles of practical bone tissue handling including fixation, embedding, and cutting. Furthermore, an overview of the standard staining techniques and what they can tell us will be provided. We will then present and discuss the process of a structured histomorphometric analysis according to ASBMR standards and explain how the results can be interpreted. With the understanding that histomorphometry is based on a technical and an intellectual part, attendees will understand how bone tissue has to be processed for histomorphometry and what we can learn from it and how it can be of great help to advance the research in the musculoskeletal field.

References

1) Standardized nomenclature, symbols, and units for bone histomorphometry: a 2012 update of the report of the ASBMR Histomorphometry Nomenclature Committee. Dempster DW, Compston JE, Drezner MK, Glorieux FH, Kanis JA, Malluche H, Meunier PJ, Ott SM, Recker RR, Parfitt AM. J Bone Miner Res. 2013 Jan;28(1):2-17.

2) Bone histomorphometry: standardization of nomenclature, symbols, and units. Report of the ASBMR Histomorphometry Nomenclature Committee. Parfitt AM, Drezner MK, Glorieux FH, Kanis JA, Malluche H, Meunier PJ, Ott SM, Recker RR. J Bone Miner Res. 1987 Dec;2(6):595-610

3) Issues in modern bone histomorphometry. Recker RR, Kimmel DB, Dempster D, Weinstein RS, Wronski TJ, Burr DB. Bone. 2011 Nov;49(5):955-64.

Lynne Hocking (Aberdeen, UK)

Grant Writing – Miep Helfrich & Lynne Hocking

Abstract

In this workshop we will take you through the process of writing a grant application assuming that for most of you it is likely to be your first ever grant application. We will focus therefore on the type of grant you are most likely to write as a graduated PhD student: an application for your own salary, a type of fellowship application. Elements of such proposals are relevant to many other situations too and therefore this workshop is useful even if you do not immediately put pen to paper, but for example apply for a funded position.

For example: you will have to write about yourself, so CV presentation will be covered. You will have to be able to summarise your project, including for an audience that consists of non-experts, so your "elevator speech" will need to be perfected. You will have to be able to write convincingly about the importance and feasibility of the project you propose and the relevance of the questions to be addressed to society as a whole. Here, we will cover how to write concise and clear text that engages the reader.

We will use elements of various funding schemes (National and EU) as examples during the workshop to illustrate different aspects of the application process.

You will be asked to do some writing and reading during the workshop and you will be asked to do some preparation. We will hand out this homework the day before the workshop as you will be able to complete this in a relatively short time.

What you should do before coming to Hamburg:

- Prepare a 300 word abstract of the project you are currently working on, or of another study (perhaps the one you wish to find funding for ?). Your abstract has to be in English and understandable to a scientific readership. Print out 2 copies on A4 and bring these along to the workshop. Your abstract should inform the reader of the purpose of your study, the aims, the methodology and whom the results may benefit. Feel free to discuss this with others, including your supervisor.
- Read webpages and have a look at the guidance for applications of funders you may consider applying to.
  Please bring such information with you when attending this workshop, or have the web address to hand.
- Consult general books about scientific writing, or general writing.
  We like:
  1. Rowena Murray, R & Sarah Moore, S *The handbook of academic writing: A fresh approach*. Open University Press-McGraw-Hill
  2. Lynne Truss. *Eats shoots and leaves.* Profile Books or Fourth Estate.
  A humoristic and useful book about correct use of punctuation.
  Both books will be available during the workshop to browse.
- There are many books about grant writing, but few about writing scientific grants. *If you have come across a useful book, please bring it along for others to browse.*

Katharina Jähn (Hamburg, Germany)

Biography

Katharina Jähn studied biochemistry and graduated from the University of Leipzig, Germany in 2006. She started her PhD training at the AO Foundation in Davos, Switzerland focusing on *ex vivo* culture systems for osteoblastic cells and the role of TGFβ_3 and serum-free culture on cell phenotype and differentiation. After obtaining her PhD degree in 2010 from the University in Cardiff, UK she started as a postdoc in the laboratory of Dr Lynda Bonewald in the Department of Oral Biology at the University of Missouri-Kansas City, USA focusing on osteocyte function. During her postdoctoral training she found that secreted factors from skeletal muscle cells protect osteocytes from dexamethasone-induced cell death and started to investigate molecular mechanisms of osteocytic osteolysis. She has received several poster and presentation awards from ASBMR and the Swiss Bone and Mineral Society. Currently, Katharina Jähn works as a postdoctoral researcher in the laboratory of Dr Eric Hesse, Heisenberg-Group for Molecular Skeletal Biology, at the University Medical Center Hamburg-Eppendorf in Hamburg, Germany. Her research includes bone imaging techniques and is focused on investigating novel molecular mechanisms regulating bone homeostasis and remodelling.

Bone Cells: Ostecytes - Katharina Jähn

Abstract

Bone mass is tightly regulated throughout life but decreases with aging due to unbalanced bone remodeling activities[1]. Osteocytes, the most prominent cells within bone, have been shown to orchestrate the activities of bone-forming osteoblasts and bone-resorbing osteoclasts during bone remodeling. The canonical Wnt/ß-catenin pathway is involved in critical functions of osteocytes. These functions range from sensing mechanical stimulation to the regulation of apoptosis and bone remodeling activities of bone surface cells. Recent work demonstrated the importance of several pathway components i.e. Lrp5/6, sclerostin, and ß-catenin in osteocytes[2,3]. For instance the sclerostin antibody represents an emerging pharmacological treatment strategy that targets the Wnt/ß-catenin signaling in osteocytes to increase bone formation in osteoporotic patients. On the other hand PTH, the only currently available anabolic treatment for bone, exerts several of its effects via the osteocyte, where it regulates sclerostin and RANKL expression[4,5]. Thus, the PTH and Wnt/ß-catenin pathways crosstalk in the osteocyte to modulate the bone remodeling activities of osteoblasts and osteoclasts. Other novel functions of osteocytes implicate a major role in the control of calcium and phosphate homeostasis, which include osteocytic osteolysis and the secretion of FGF23.

This lecture will summarize the functions that have been attributed to osteocytes with special emphasis on recent findings on molecular pathways affecting bone homeostasis. Furthermore, the seminar will cover *in vitro* and *in vivo* methods to study osteocytes and discuss future scientific questions in the field of osteocyte biology.

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ECTS PhD TRAINING 15-18 September 2013, Hamburg

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Hank Kronenberg (Boston, US)

Biography

Henry M. Kronenberg is Chief of the Endocrine Unit at the Massachusetts General Hospital and Professor of Medicine at the Harvard Medical School. There he leads a research group that studies the actions of parathyroid hormone and parathyroid hormone-related protein, with a particular emphasis on bone development, bone biology, calcium homeostasis, and the roles of osteoblast-lineage cells in hematopoiesis. Dr. Kronenberg's laboratory in recent years has used a number of genetically altered strains of mice to establish the role of signaling by the PTH/PTHrP receptor in bone.

Dr. Kronenberg received his BA from Harvard University, his MD from Columbia University, his medical house officer training at the Massachusetts General Hospital, and post-doctoral training at NIH, MIT, and the MGH.

PTH Signalling in Bone – Hank Kronenberg

Abstract

The most important function of parathyroid hormone (PTH) is to regulate the level of calcium in the blood through its actions on bone and kidney. Strongly supporting this claim is the observation that calcium in the major regulator of PTH secretion and synthesis. PTH causes release of calcium for bone matrix primarily through stimulation of osteoclast production and activity, though recent studies demonstrate that, in some settings, PTH action on osteocytes to release calcium may be independent of osteoclastic activation ("osteocytic osteolysis"). This simple paradigm for PTH action, however, does not obviously predict the surprisingly complex actions of PTH on bone. These are most dramatically illustrated by the actions of PTH, when administered by once daily subcutaneous injection, to increase bone mass, despite the associated increase in bone resorption. Those so-called anabolic actions of PTH include PTH's action to suppress apoptosis of osteoblastic cells, to activate previously dormant bone lining cells, and to increase the commitment to and proliferation of early cells of the osteoblast lineage. All of these actions serve to increase the number of active osteoblasts. Mechanisms that accomplish these goals are poorly understood. Some may be direct actions through activation of the PTH/PTHrP receptor found on osteoblasts, osteocytes, and early cells of the lineage. But other actions may be indirect actions through activation of the wnt pathway at multiple steps, stimulation and secretion of growth factors, such as IGF1, FGF2, and amphiregulin. Speculations about the rationale for these complicated anabolic actions of a calcium-regulating hormone include the possibility that PTH simply increases bone formation to assure adequate stores of calcium and also the possibility that PTH is tapping into the functions of its cousin, parathyroid hormone-related protein. Here I will provide an overview of these issues, focusing on the multiple cellular targets of PTH action in bone.

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Bente Langdahl (Aarhus, Denmark)

Biography

Bente Langdahl graduated from medical school at Aarhus University in 1988 and did clinical training in internal medicine and endocrinology at Aarhus University Hospital. Bente Langdahl received her PhD at Aarhus University in 1995: "Investigations on a possible pathogenic role of thyroid hormones in postmenopausal osteoporosis" and received a DMSc at the same university in 2004: "The genetics of bone mass and risk of osteoporotic fractures". In 2004 Bente Langdahl was appointed consultant at the department of Endocrinology and Internal Medicine at Aarhus University Hospital and research lecturer at Aarhus University. In 2012 Bente Langdahl was appointed professor at Aarhus University. Bente Langdahl's main research interests are identification and further investigation of genetic variants that imply increased risk of osteoporotic fractures, osteogenesis imperfecta in adult patients, interactions between fat and bone tissues with a special interest in the PPARgamma pathway and the development of new treatments for osteoporosis.

Bente Langdahl is President of the European Calcified Tissue Society.

Vitamin D and Bone Health – Bente Langdahl

Abstract

The importance of vitamin D for bone health has been highlighted in situations where patients have too little vitamin D. In children this results in rickets with bowing of the long bones and in adults in osteomalacia characterised by bone pain and fractures. Both conditions are caused by insufficient mineralisation of bone and secondary hyperparathyroidism causing increased bone turnover.

Vitamin D is produced in the skin upon sunlight expose and subsequently hydroxylated (position 25) in the liver. Vitamin D can also be obtained from the diet. Particularly fatty fish contains high levels of vitamin D, but also meat and some dairy products contain vitamin D. The vitamin D produced by the skin and liver or obtained from the diet is inactive and needs another hydroxylation (position 1) before it is active and can bind to the vitamin D receptor. The hydroxylase responsible or this activation is found in the kidney and is responsible for the serum levels of activated vitamin D. However, the hydroxylase is also present in many cell types and activation can therefore also take place locally. The renal hydroxylase is mainly controlled by PTH, whereas the local hydroxylases probably are controlled by many different hormones and growth factors.

The definition of vitamin D insufficiency and deficiency has been the topic of many debates and publications. There seems to be consensus at the moment that vitamin D needs to be high enough to prevent seasonal variations in PTH. This level is 50 nmol/l. Others have suggested that the level of vitamin D should be higher in order to prevent osteoporotic fractures. This level has been suggested to be 60-75 nmol/l.

Studies on the effect of treatment with vitamin D with or without calcium have been numerous and with conflicting results. Meta-analyses have been performed and although not in complete agreement most of them suggests that there is positive effects of vitamin D on falls and fractures, specifically if the doses of vitamin D is high and if the population treated is elderly.

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Thomas Pap (Muenster, Germany)

Biography

Professor of Experimental Medicine and Director
Institute of Experimental Musculoskeletal Medicine
Westfälische Wilhelms-University Münster

Medical and academic career

1988 - 94	Medical School, Otto-von-Guericke University Magdeburg (Graduation with 'Auszeichnung', Best Graduate Award)
1994 - 97	Resident, Department of Rheumatology, Otto-von-Guericke University Magdeburg (Vogelsang Hospital)
1995	Medical degree 'Dr. med.' [Methotrexate and Sulfasalazine as second-line agents for the treatment of rheumatoid arthritis], Otto-von-Guericke University Magdeburg
1997 - 99	Postdoctoral Fellow, Center for Experimental Rheumatology and WHO Collaborating Center for Molecular Biology and Novel Therapeutic Strategies for Rheumatic Diseases, Department of Rheumatology, University Hospital Zurich
1999 - 04	Senior Research Fellow, Center for Experimental Rheumatology and WHO Collaborating Center for Molecular Biology and Novel Therapeutic Strategies for Rheumatic Diseases, Department of Rheumatology, University Hospital Zurich (2000-2004: 10%)
2000 - 04	Head, Division of Experimental Rheumatology and Orthopaedics, Otto-von Guericke University Magdeburg and Clinical Resident, Center for Internal Medicine
2001 - 04	Head of an 'Emmy-Noether' Group (funded by the German Research
since 2004	Professor of Experimental Medicine (tenure) and Head Division of Molecular Medicine of Musculoskeletal Tissue, Westfälische Wilhelms- University Münster
2006 - 2007	Associate Dean of the Faculty of Medicine, Westfälische Wilhelms- University Münster
since 2009	Director of the Institute of Experimental Musculoskeletal Medicine, Westfälische Wilhelms- University Münster

Relevant publications

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Bone Erosion in Rheumatoid Arthritis – Thomas Pap

Abstract

Rheumatoid arthritis (RA) is a chronic, autoimmune and inflammatory disease that primarily affects synovial joints. It results in the progressive destruction of articular structures, most prominently the bone. Bone destruction in RA not only is a pathophysiological hallmark of the disease, but determines the outcome of RA in most affected individuals. It is largely irreversible and the major cause for disability.

Development of rheumatoid bone destruction is triggered by the hyperplasia of the synovial membrane as caused by an infiltration and accumulation of inflammatory cells such as macrophages and lymphocytes as well as an increase in the number of resident mesenchymal cells. In this inflammatory pannus, there are high numbers of osteoclasts that differentiate from macrophages in the course of disease invade the adjacent bone. Their differentiation is triggered by a number of soluble factors, most prominently the receptor activator of the nuclear factor kappa B- ligand (RANKL) and different inflammatory cytokines. They are secreted both from inflammatory cells and from resident fibroblasts. These fibroblast-like synoviocytes (FLS) are a key part of the local immune system in the joints and integrate signals form different sources into a pathological tissue response. It has been shown that in the rheumatoid joint, FLS undergo a stable activation that is also called tumour- like transformation. This results in the imprinting of the specific phenotype of RA-FLS - most likely through a combination of genetic and epigenetic mechanisms - and is maintained in the absence of continuous stimulation by inflammatory triggers. As a consequence of this stable activation, FLS keep secreting factors that contribute to the differentiation and activation of osteoclasts.

Important information concerning mechanisms and consequences of increased osteoclastogenesis and subsequent bone resorption have come from the human TNFalpha transgenic (hTNFtg) mouse. These mice overexpress human TNFalpha from a gene construct with a modified 3’ UTR, and owing to increased local and systemic levels of TNFalpha develop an RA- like destructive arthritis. Studies using these mice have identified a number of key molecules involved in the regulation of osteoclast

differentiation and, thus, bone destruction in RA. These include factors that are secreted form FLS and that act on osteoclastogenesis directly as well as adaptor and signalling molecules in the osteoclasts that in response to well known cytokines and growth factors modulate pathologic osteoclast differentiation and activity in RA.

This talk will summarize key aspects of osteoclastogenesis in RA, point to recently identified mechanisms of bone resorption in the disease and discuss their therapeutic potential.

Fernando Rivadeneira (Rotterdam, Netherlands)

Biography

Fernando Rivadeneira is a medical doctor with a PhD in Endocrine and Genetic Epidemiology, currently an Associate Professor of the Department of Internal Medicine at Erasmus University Medical Center Rotterdam. He has been in charge of supervising the high-throughput genotyping of genome-wide association (GWAS) projects and running next-generation sequencing (NGS) efforts at the Genetic Laboratory of Erasmus MC. His main area of research focuses on the identification of genetic determinants of musculoskeletal traits applying high throughput technology, i.e., GWAS and NGS. His work is primarily embedded within the Rotterdam Study (a large population-based study of disease and disability in the elderly) and the Generation R (a multi-ethnic birth cohort in Rotterdam) epidemiological cohorts, also in the setting of large international consortia. He is coordinator of the Rotterdam Study GWAS project and has supported researchers from different epidemiological fields using GWAS and sequencing data, leading to hundreds of publications in the field. His work has led to the identification of the first novel genetic variants using the GWAS approach in the field of osteoporosis (Lancet 2008) and within the EU-FP7 funded Genetic Factors of Osteoporosis (GEFOS) Consortium, the identification of 20 (Nature Genetics 2009) and 56 (Nature Genetics 2012) loci influencing BMD variation. He has also co-lead the identification of 180 height loci, in one of the largest GWAS to date from the GIANT consortium involving >180.000 subjects (Nature 2010). His work on the Generation R study focuses on life-course trajectories of musculoskeletal traits and has recently identified WNT16 as a critical gene influencing peak bone mass accrual during childhood (PLoS Genetics 2012). His work towards the future aims in translating these GWAS and NGS discoveries into therapeutic applications for the treatment of osteoporosis and other musculoskeletal conditions.

GWAS and Next Generation Sequencing – Fernando Rivadeneira

Osteoporosis is a common disease characterized by systemic impairment of bone mass and microarchitecture increasing susceptibility to fragility fractures. With the advent of genome-wide association studies (GWAS) the number of genetic discoveries has increased dramatically for all complex diseases, and such is the case for osteoporosis. To date 56 BMD loci have been identified by GWAS as robustly associated with BMD, while the analysis of several other osteoporosis-related traits are currently underway. During my talk I will focus on the BMD loci identified within the setting of the Genetic Factors of Osteoporosis (GEFOS) consortium and the candidate genes likely (yet not unequivocally) to be underlying the GWAS signals. Many of the identified variants map in the vicinity of genes of unknown function (representing cutting-edge new biology), while several other factors cluster within critical biological pathways relevant for bone biology, like Wnt signalling, OPG-RANK-RANKL and mesenchymal cell differentiation. While at least 14 of these loci were also shown associated with fracture risk, these variants identified by GWAS together explain 5-6% of the variation in BMD, thus limiting its application for meaningful clinical risk prediction. On the other hand, the translation of the knowledge derived from these GWAS discoveries into therapeutic applications for the treatment of osteoporosis is warranted towards the future. Here the inclusion of new technologies like next generation-sequencing will play a key role providing further insight into the complex genetic architecture of BMD, osteoporosis and fracture risk.

Alexander Robling (Indiana, US)

Biography

My laboratory works on treatments for metabolic and genetic bone diseases. We use molecular genetic approaches in rodents to identify new ways to make bone stronger. One particular approach that we have focused on is mechanobiology. Mechanobiology merges the older science of mechanics with the newer and emerging disciplines of molecular biology and genetics. At the center of mechanobiology is the cellular process of mechanotransduction, or the way cells sense and respond to mechanical forces. We research how mechanical loads signal bone cells to build stronger bones. Our research program has led us to the Wnt signaling pathway as a major regulator of mechanotransduction. Current projects are centered around the particular steps in the Wnt cascade that are activated by mechanical stimuli and ultimately lead to increased anabolism.

Mechanical Stimulation and Signal Transduction in Bone – Alexander Robling

Abstract

Bone size, strength, and composition are modulated by the mechanical loading environment to which resident bone cells are exposed. Unraveling the biological processes that guide mechanobiological adaptation in bone tissue requires appropriate mechanotransduction models and molecular tools. Some fundamental rules and biochemical mechanisms have been established, based on engineering principles, organ physiology, and cell and molecular biology, that explain and account for the process of bone adaptation to the mechanical environment. The cells that sense mechanical inputs to the bone are probably the osteocytes, which have a number of specialized mechanisms, both morphological and biochemical, to convey mechanical information to the effector cells, i.e. those that change bone mass, size, and shape. We are now beginning to understand some of these cellular mechanisms that affect mechanical signaling in bone, and potentially bone tissue mechano-sensitivity. One such mechanism involves Wnt signaling—a pathway involved in many physiologic processes including body axis specification/patterning, and morphogenic signaling. Among bone cells, Wnt signaling through the Wnt co-receptor LDL-related receptor-5 (Lrp5) is crucial for load-induced bone formation. We have found that mechanical loading strongly regulates Sost/sclerostin expression—a potent inhibitor of Wnt signaling. Moreover, Sost, and its protein product sclerostin, are exclusive to the osteocytes—the cell type postulated to be the tissue's sensor apparatus in transducing mechanical loading events. The suppression of sclerostin expression by mechanical loading may provide a mechanism by which Wnt signaling is enhanced and bone formation is enacted. Furthermore, Wnt signaling modulation via Sost/sclerostin also appears to modulate the process of disuse osteoporosis. As the mechanisms of mechanical signal transduction become revealed, a greater range of targetable molecules will be manifest, that might ultimately be exploited to improve bone health among patients with a wide range of skeletal disease.

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Robling AG, Turner CH. (2009) Mechanical signaling for bone modeling and remodeling. *Crit Rev Eukaryot Gene Expr*.19(4):319-38

Hiroaki Saito (Hamburg, Germany)

Biography

Dr Hiroaki Saito received his PhD in Pharmacology of hard-tissue at Tokyo Medical & Dental University. During his PhD course, he investigated therapeutic options to treat Rheumatoid Arthritis using anti-TNF and anti-Rankl therapy. In 2006, Dr Saito started his postdoctoral training in the laboratory of Dr. Roland Baron, which was by the time located within the Department of Orthopaedics & Rehabilitation at Yale University School of Medicine. In early 2008, he relocated with the lab to the Harvard School of Dental Medicine, where he continued his training as a postdoctoral researcher focusing on Wnt signalling and osteoblast biology until 2012. During his training, Dr Saito obtained an in-depth expertise in bone histomorphometry and various animal models. He was awarded an ASBMR Young Investigator Award in 2008 and an ECTS New Investigator Award in 2010. Currently, he is a postdoctoral researcher in the laboratory of Dr Eric Hesse, Heisenberg-Group for Molecular Skeletal Biology, at the University Medical Center Hamburg-Eppendorf in Hamburg, Germany. He is supported by a fellowship of the Japanese Society for the Promotion of Science and continues to work on osteoblast biology and bone remodelling.

Histomorphometry - Hiroaki Saito & Eric Hesse

Abstract

The overall goal of this histomorphometry workshop is to initiate an interactive discussion about the technical and conceptual principles of histomorphometry as a very specialized but important method in the field. This lecture will include practical examples and exercises coming from recently published or ongoing hot topic projects in the field of bone research. The seminar intends to teach the technical principles of practical bone tissue handling including fixation, embedding, and cutting. Furthermore, an overview of the standard staining techniques and what they can tell us will be provided. We will then present and discuss the process of a structured histomorphometric analysis according to ASBMR standards and explain how the results can be interpreted. With the understanding that histomorphometry is based on a technical and an intellectual part, attendees will understand how bone tissue has to be processed for histomorphometry and what we can learn from it and how it can be of great help to advance the research in the musculoskeletal field.

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Cristina Sobacchi (Italy)

Biography

Cristina SOBACCHI (DOB 1974) started her work in the laboratory directed by Dr Anna Villa at the National Research Council (CNR) of Milan, in the Department of Human Genome. She was initially involved in the characterization of the molecular and cellular defect in a rare and peculiar primary immunodeficiency, Omenn Syndrome. In 2000, she contributed to the identification of the first gene responsible for human Autosomal Recessive Osteopetrosis (ARO), a rare bone genetic disease which in subsequent years proved to be genetically heterogeneous. Thereafter, she has been involved in the molecular dissection of human ARO. In particular, in 2007 she reported on Nature Genetics as a first author the identification of TNFSF11 as the gene responsible for a subset of osteoclast-poor ARO. More recently, she focused on the preclinical development of novel therapeutic approaches to RANKL-dependent ARO, for which there is no cure so far. Thanks to the collaboration with Amgen, owner of the RANKL patent, she demonstrated that the exogenous administration of this cytokine to the Rankl knock-out mouse model rescues the bone phenotype and also has beneficial effects on the hematolymphoid compartment. These results pave the way to the treatment of human patients and show the translational side of her research.

She's a member of the European Calcified Tissue Society, from which she received the Young Investigator Award in 2007 and the ECTS/ABBH Iain Boyle Award in 2009. Since 2007 she has been Principal Investigator in several research grants funded by Telethon Foundation, Cariplo Foundation and the Italian Ministry of Health. Since 2010 she's a Researcher of the CNR with a permanent position.

She has published 21 articles on osteopetrosis and other bone diseases.

Bone Cells: Osteoclasts – Cristina Sobacchi

Abstract

Bone is a dynamic tissue in which the concerted activities of different cell types (osteoclasts, osteoblasts, osteocytes), under the regulation of osteotropic and calciotropic hormones and cytokines, assure normal skeletal development (growth and remodeling) and the maintenance of its integrity throughout life. In particular, osteoclasts are giant, multinucleated cells specialized in bone resorption (1); they derive from precursor cells of the monocyte/macrophage lineage, as demonstrated for the first time in the early 1980s by the cure of an osteopetrotic child by bone marrow transplantation (2). On the other hand, osteoblasts are cells of mesenchymal origin which synthesize bone matrix. A cross-talk exists between osteoclasts and osteoblasts in order to regulate their antagonistic functions (3). Another cell type, the osteocyte, has recently come to the fore as an important regulator of bone homeostasis, through its mechano-sensing activity and the production of soluble factors (4). However, skeletal homeostasis is not only a matter of bone cells; on the contrary, the existing osteoimmunological network, that is the cross-talk between cells of the immune system and those of bone, importantly contributes, particularly in pathological conditions (5,6).

Alterations of this complex balance may cause either an increase or a decrease in bone mass (7). In particular, the osteopetroses are a group of rare diseases presenting with a generalized increase in bone density due to a defect in bone resorption because of either osteoclast absence (osteoclast-poor forms) or osteoclast dysfunction (osteoclast-rich forms) (8).

Based on studies in humans and in animal models, here we will focus on the various phases of an osteoclast life: from differentiation, particularly dealing with the RANKL/RANK/OPG axis (9); to adhesion, polarization and ruffled border formation (10); to resorption and finally switch-off (11).

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Gudrun Stenbeck (London, UK)

Biography

Gudrun Stenbeck studied chemical engineering at undergraduate level and received her PhD from the Technical University Darmstadt, Germany, in 1993 after PhD work in the laboratory of Prof Felix Wieland at the University of Heidelberg. She then undertook postdoctoral work with Prof James E. Rothman at the Memorial Sloan Kettering Cancer Centre in New York before joining the laboratory of Prof Mike Horton at University College London to study the molecular mechanisms of osteoclastic bone resorption. She was awarded an Arthritis Research UK fellowship in 2000 to establish her independent research and is now a lecturer in Biomedical Sciences at Brunel University in London, UK.

Her research focuses on the cell biology of bone homeostasis and extracellular matrix deposition. Her current research is aimed at the characterisation of the signalling events relaying extracellular stimuli to components of the intracellular trafficking machinery to develop new strategies for the treatment of osteoarthritis, osteoporosis and fibrotic diseases. Her research has been funded by Arthritis Research UK, the Royal Society and the Welcome Trust.

She teaches cell biology and biochemistry at both undergraduate and postgraduate levels.

Calcium Homeostasis and Transport – Gudrun Stenbeck

Abstract

Calcium plays an important role in the organism not only during bone formation but also as a signalling molecule. The concentration of ionised calcium in the blood is kept constant through the interplay of PTH, Vitamin D and calcitonin, which control calcium handling in kidney, intestine and bone.

The bone conserving hormone calcitonin has the unique property of inducing osteoclast quiescence without reducing osteoclast numbers. The recent discovery that osteoclasts play a significant role in providing bone-forming signals has led to renewed interest in calcitonin as a treatment for osteoporosis and osteoarthritis. This presentation will explore the molecular mechanisms of calcitonin action on osteoclasts.

During osteoclastic bone resorption digested extracellular matrix is removed from the resorption pit by endocytosis. Endocytosed material takes a transcytotic route to the bone avert side of the cell and is secreted. Signals leading to the cessation of resorption are poorly defined but flow through the different vesicular trafficking pathways could impart a monitoring function of its resorption activity to the osteoclast.

To establish the directionality and kinetics of trafficking events in resorbing osteoclasts, we devised an assay system using inert fluorescent low molecular weight markers as probes. Osteoclasts derived from either neonatal rabbits or generated in vitro from peripheral blood monocytes are plated on dentine discs and fluorescent marker distribution is monitored by confocal laser scanning microscopy.

Calcitonin acts through the calcitonin receptor which couples to several different trimeric G proteins thus activating distinct signalling pathways. We have shown that in osteoclasts, it blocks endocytosis from the ruffled border by phospholipase C (PLC) activation. Inhibition of PLC prior to calcitonin

treatment restores endocytosis to 75% of untreated rates. This effect is independent of PKC activation and can be mimicked by an increase in intracellular calcium. These intracellular oscillations in calcium ion concentration are essential to maintain endocytosis and transcytosis during resorption. Taken together our results define a link between systemic hormone signalling and intracellular trafficking and suggest the exciting possibility that an increase in intracellular calcium couples exo- and endocytosis in osteoclasts via calcium sensing proteins such as synaptotagmin VII.

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Hanna Taipaleenmäki (Hamburg, Germany)

Biography

Hanna Taipaleenmäki studied genetics and molecular biology at the University of Oulu, Finland where she graduated as a M.Sc. in 2005. Her PhD research in the Department of Medical Biochemistry and Genetics at the University of Turku, Finland and in the Endocrine Research Unit at Odense University Hospital, Denmark was focusing on transcriptional and post-transcriptional control of mesenchymal stem cell differentiation and bone homeostasis. After obtaining her PhD degree in 2010 she started as a postdoctoral fellow in the laboratory of Drs Gary Stein, Jane Lian, Janet Stein, and André van Wijnen in the Department of Cell Biology at the University of Massachusetts Medical School in Worcester, USA. During her postdoctoral training she expanded her research into the field of bone and cancer and investigated microRNA-mediated regulation of breast cancer bone metastasis and osteosarcoma progression. During her training, she has received several awards from the ASBMR, ECTS, and IBMS. Currently, she is a postdoctoral researcher in the laboratory of Dr Eric Hesse, Heisenberg-Group for Molecular Skeletal Biology, at the University Medical Center Hamburg-Eppendorf in Hamburg, Germany. Her research is focusing on the role of osteoblasts in regulating tumor cell activity in the bone marrow, and is funded by fellowships from EMBO, Humboldt Foundation, and the Finnish Cultural Foundation.

miRNA Approaches - Hanna Taipaleenmäki

Abstract

MicroRNAs (miRNAs) are small non-coding RNAs that regulate multiple biological processes, including bone formation and bone resorption, thereby contributing to the control of bone homeostasis. Deregulation of miRNA-mediated mechanisms is implicated in the pathogenesis of a large number of disease conditions, including osteoporosis and cancer. The aim of this lecture is to give an introduction to the role of miRNAs in bone physiology and pathology. In addition, the potential clinical use of manipulating miRNAs in therapeutic approaches will be discussed.

Since their discovery two decades ago, miRNAs have emerged as key post-transcriptional repressors that modulate cell signaling by interfering with mRNA stability and/or by blocking protein translation. By binding to specific complementary sequences in the 3'UTR of their target mRNAs, miRNAs control key components of osteogenic pathways leading to positive or negative effects on osteoblast and osteoclast differentiation and function. Conditional deletion of the miRNA processing enzyme Dicer in osteoblasts, chondrocytes, and osteoclasts results into severe skeletal defects, revealing the essential role for miRNAs in normal skeletal development and bone homeostasis. An increasing number of specific miRNAs have been identified that are regulated during osteoblast and osteoclast differentiation, and that modulate bone-specific regulatory networks. This presentation will discuss the cellular and molecular mechanisms by which miRNAs function, including experimental approaches to investigate miRNA-mediated control of osteogenic signaling pathways.

In addition to controlling physiological bone maintenance, miRNAs are involved in several bone-related diseases. For instance, aberrant miRNA expression has been implicated in the pathology of osteoporosis, osteoarthritis, and in primary bone tumors as well as metastases to bone. As a consequence, miRNAs have clinical potential as disease-specific biomarkers. Furthermore, *in vivo* delivery of miRNAs or miRNA antagonists provides an attractive therapeutic tool to reverse bone tissue degeneration or to prevent cancer-induced bone diseases. Although studies have shown promising results in using miRNAs in skeletal therapy, further investigations are required to better understand the advantages and limitations of this approach. Future perspectives of miRNAs in bone

biology, and the potential of applying miRNAs in clinical interventions will be discussed during the lecture.

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Anna Tampieri (Faenza, Italy)

Biography

Research Manager at the National Research Council - Institute of Science and Technology for Ceramics in Faenza (Italy) where she heads the Department of Bio-ceramic and bio-hybrid composites for Regenerative Medicine. Doctor degree in Chemistry at University of Bologna with Laude in 1985. Since 1988 Researcher at IRTEC-CNR and since 1996 coordinator of the Bioceramic Group. From 2006 in charge as Research Manager (full professor) at ISTEC-CNR. Co-author of 180 papers published on International Journals (**H index=26**). Inventor of 18 international patents.

Coordinator of 4 European Projects entitled:
AUTOBONE NMP3-CT 2003-505711 (2003-09),
TEM-PLANT NMP4-CT-2006-033277 (2006-11),
OPHIS FP7-NMP-2009 246373-2 (2010-2014),
SMILEY FP7-NMP-SL-2012-SMALL-6-310637 (2012-2015)

WP leader in the frame of the Large Project MAGISTER NMP3-LA-2008-214685 (2008-12). WP Leader of the Italian Project "Aging - INTERESSE_INVECCHIAMENTO" (2012-2015). Coordinator of National Project FIRB "Development of Articular Bio-prosthesis". Scientific advisor of Italian Ministry Ministry of Economic Development and Industry. Scientific Advisor of the French National Research Agency (MAtetPro projects). Carried out several qualified collaborations and consultancy services for chemical, biochemical and pharmaceutical companies (Johnson&Johnson, FIN-CERAMICA Biomedical Devices, Menarini Farm., etc.). Performed several courses and lessons within specialization degrees, PhD and masters for the Universities of Ferrara, Bologna, Modena, Padova, Palermo, Milano, Lione and La Cattolica (Rome).
Bestowed in 2005 with the Prize "Marisa Bellisario" by confindustria Italia for the Advancement in biomedical field.

In 1995 she founded the company FINCERAMICA Biomedical Solution and initially she was the Idea-woman, later the former CEO and today the head of the Scientific Advisory Board of Fin-Ceramica Faenza Spa, a company which owns worldwide patents in the regenerative surgery field. Dr. Tampieri has also carried out an intense activity of education and dissemination through Masters and Ph.D. lessons. Head of the commission for the evaluation of Spin-off generated by scientific project at Italian CNR. Her recent scientific activity is focused on the biomorphic transformations and the bio-mineralization processes in service for nanomedicine.

Organizer of several International Symposia and National Conferences on Biomaterials.
Since 2008 member of "Nanomedicine Platform of European Commission". In 2009 TIME Magazine selected the research "from Wood to Bone" by Anna Tampieri as the 30° research among the most important 50 researches of the year. Since 2011 she becomes Senior Affiliate Member of Methodist Hospital Research Institute, Houston Texas, where she is consultant in Material Science for Regenerative Medicine. From June 2012 to present, Coordinator of the Commission CNCCS "Bioeconomy Rome" Prize and Member of the National Research Council Advisory Board for the Emerging Companies and Start Up.

Nanotechnology in Regenerative Medicine – Anna Tampieri

Abstract

ECTS PhD TRAINING 15-18 September 2013, Hamburg

During the last decade, in the orthopedic field the well-established approach for curing diseased bone parts, based on replacement with inert substitutes, has progressively given way to new regenerative approaches, based on the use of bioactive and biomimetic devices. However, technological limitations exist which slacken the establishment of regenerative therapies, since the regeneration of organized and multi-functional tissues (like osteo-cartilaginous anatomical regions) requires scaffolds able to show compositional and structural complexity.

The presentation will illustrate how bio-mineralization, an amazing natural process with which nature has realized and optimized a profuse collection of living organisms endowed with astonishing abilities, can be used to guide efforts for developing biomaterials for bone and osteochondral regeneration.

In particular it will be illustrated how the self-assembling and bio-mineralization of natural polymeric fibers can be induced by reproducing the conditions of formation of new bone tissue in mammals, thus obtaining a collagen-based matrix where mineralization with nanoparticles of biomimetic apatite can take place. The reproduction of biomimetic conditions of bone synthesis allows to obtain hybrid constructs where the mineral phase is nucleated upon guidance by the chemical features and physical confinement imposed by the polymeric matrix, so that the mineral phase has physical, chemical and ultra-structural resemblance with mineral bone, thus providing very high osteogenic activity when implanted in vivo. Besides, the possibility to vary the degree of mineralization allows to obtain multi-layer graded devices able to regenerate the different districts of the articular region (subchondral bone, mineralized and hyaline cartilage). Finally, pinning on the recent development, performed by our research group, of intrinsic superparamagnetism exhibited by hydroxyapatite nano-powders upon crystallographic and chemically controlled doping with Fe(II)/Fe(III) ions, it will be illustrated how bio-hybrid bone-like devices with intrinsic magnetic properties can be obtained; such devices can increasingly assist the osteogenic and angiogenic capacity of biologically inspired bone and osteochondral scaffolds, through magnetically-driven release of specific growth factors.

Moreover, the use of nano-structured phases able to penetrate inside cells for the treatment of tumors and/or for image diagnostics in nanomedicine is currently one of the most innovative and promising approaches. Intelligent nanoparticles able to be moved and driven by “magnetic guiding” and even to enter into the cells as non-viral vectors, are very promising materials for therapies and diagnostics in case of various cancer diseases.

Andre van Wijnen (Minnesota, US)

Biography

Andre van Wijnen received training at the University of Utrecht (Netherlands), University of Florida (Gainesville, FL, USA) and University of Massachusetts Medical School (Worcester, MA, USA) where he earned a doctorate in Biomedical Sciences. His research focuses on patient-oriented applications of molecular knowledge to facilitate orthopedic tissue regeneration using human stem cells ('molecular stem cell engineering'), as well as to understand orthopedic cancers (e.g., osteosarcoma and chondrosarcoma) and metastatic cancers to bone (e.g., breast and prosate). The biological expertise of the PI encompasses stem cells, mammalian development, bone, cartilage and cancer. Research projects in his laboratory apply state-of-the-art approaches and innovative ideas to address both biological and mechanistic questions related to stem cell lineage commitment and mammalian development, as well as bone tumorigenesis and metastasis. Molecular topics of interest include microRNAs, epigenetics, gene regulation and cell signaling within the context of multipotent stem cell self-renewal, as well as proliferation and differentiation of normal and tumor cells. He has published >350 papers, has reviewed for >100 different journals, serves on a number of Editorial Boards (e.g., Journal of Biological Chemistry, Journal of Bone and Mineral Research) and is Editor in Chief of two journals (GENE and Molecular Biology Reports). He is an active member of the ECTS, ASBMR, IMBS and ORS.

Epigenetic landscaping in healthy bone and disease – Andre van Wijnen

Abstract

Patients with bone skeletal degeneration and/or injuries may benefit from innovative repair strategies with live or engineered tissues that depend on the activities of multi-potent mesenchymal stem cells (MSCs), osteoprogenitor cells and osteoblasts. One critical hiatus in our knowledge of the biological properties of mesenchymal cells is that there is no definitive and comprehensive description of the key molecular circuits that define multi-potent, pre-commited or mature cellular phenotypes. Although genetic differences contribute to biological distinctions in the properties of MSCs, we will discuss the concept that informative biological variation between healthy indivuals and patients is epigenetically mediated at the level of chromatin modifications, transcription factors and microRNAs. We will show new data indicating that mesenchymal cells have a unique epigenetic landscape of cellular regulatory proteins that can be accurately measured and leveraged to make informed decisions for clinical use of stem cells in tissue repair as either unmodified pristine cells or upon epigenetic modification with RNA based reagents or pharmaceutical drugs. We discuss these findings in the broader context of bone and stem cell biology, as well as indicate possibilities for immediate applications in research and clinical settings to improve 'point-of-care' applications to treat skeletal afflictions.

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